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Journal of Eco-Friendly Agriculture

Doctor's Krishi Evam Bagwani Vikas Sanstha
(Doctor's Agricultural and Horticultural Development Society)
Registered under Society Registration Act 21, 1860
Lucknow - 226 016, India

JOURNAL OF ECO-FRIENDLY AGRICULTURE

(A Bi-annual Scientific Research Journal)
Doctor's Krishi Evam Bagwani Vikas Santha
(Doctor's Agricultural and Horticultural Development Society)
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Subscription 2-years	1800.00	6000.00	250.00	400.00
Subscription 3-years	2500.00	8000.00	350.00	500.00
Processing Charges	1000.00 Per article	Not Applicable	50.00 per article	Not Applicable
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त्रिलोचन महापात्र, पीएच.डी.
सचिव, एवं महानिदेशक
TRILOCHAN MOHAPATRA, Ph.D.
SECRETARY & DIRECTOR GENERAL

भारत सरकार
कृषि अनुसंधान और शिक्षा विभाग एवं
भारतीय कृषि अनुसंधान परिषद
कृषि एवं किसान कल्याण मंत्रालय, कृषि भवन, नई दिल्ली 110 001

GOVERMENT OF INDIA
DEPARTMENT OF AGRICULTURAL RESEARCH & EDUCATION
AND
INDIAN COUNCIL OF AGRICULTURAL RESEARCH
MINISTRY OF AGRICULTURE AND FARMER WELFARE
KRISHI BHAVAN, NEW DELHI 100 001
Tel.: 23382629; 23386711 Fax: 91-11-23384773
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MESSAGE

Use of natural products in agriculture has assumed greater practical importance with the current thrust on environmental sustainability and organic farming. A large number of plant products, bio-pesticides, bio-agents and farm manure have been identified as one of the most potent sources of environmentally safe production. **I** am happy to note that **Doctors Krishi Evam Bagwani Vikas Sanstha (Doctors Agricultural and Horticultural Development Society),** a scientific society established in Lucknow since 1999, is fully dedicated towards promoting the cultivation of horticultural and agricultural crops in eco-friendly ways. The Sanstha is publishing a journal, namely **Journal of Ecofriendly Agriculture** covering different aspects of organic farming.

I congratulate the entire team of "Journal of Eco-friendly Agriculture" for raising awareness about organic farming.

(T. MOHAPATRA)

Dated the 10th September, 2021
New **Delhi**

Summary Proceedings of the International conference

On

Agriculture and Environmental Sciences (ICAES-2021)

September 01-02, 2021

THEME

Innovative Approach in Agricultural and Allied Sciences, Natural resource management, Food and Environmental Security & AGRI and Animal Husbandry Start-ups

ORGANIZED BY

Department of Agriculture, Mangalayatan University, Beswan, Aligarh, U.P, India, 202145

Doctors Krishi Evam Bagwani Vikas Sanstha, Lucknow, Uttar Pradesh

Krishi Rakshak Samiti, Uttar Pradesh

Summary proceedings of the International conference on Agriculture and Environmental Sciences (ICAES-2021)

COMPILED AND EDITED BY

Dr. Syed Danish Yaseen Naqvi,
Associate Professor & Head

Department of Agriculture, Mangalayatan University, Beswan, Aligarh, U.P, India, 202145

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Er. Ashish Jain, Lecturer Department of Civil Engineering, Mangalayatan University, Beswan, Aligarh, U.P, India, 202145

Dr. Harit Priyadershi, Assistant Professor, Department of Civil Engineering, Mangalayatan University, Beswan, Aligarh, U.P, India, 202145

SUMMARY AND PROCEEDINGS OF THE CONFERENCE

ABOUT THE INTERNATIONAL CONFERENCE

The huge population pressure on limited agricultural and natural resources in the developing countries in the recent past have resulted in ecological imbalance and a great threat to the existence of life on earth. Therefore, there is a great need for an institutional efforts as well as people's participation to develop technologies for conservation of natural resources for food security and existence of life on earth. Against this background, the International Conference on "Agricultural and Environmental Science (AAES-2021)" was designed focussing on various scientific tracks covering major areas of Research on Agriculture, Biological and Environmental sciences. In this context, emphasis was pointed on novel tools and technologies in the field of Agricultural and Allied Science, Social Sciences, Biological, and Physical Sciences. The conference also aimed at to bring together the global scientific community, policymakers, administrators, industry representatives and other stakeholders to discuss the practical problems and

challenges encountered and share their views on solutions adopted by the farming community all over the world. It was also a platform to strengthen the friendship and collaboration among the scientists, academia and different Institutes.

ABOUT THE MANGALAYATAN UNIVERSITY, ALIGARH, UTTAR PRADESH, INDIA

To be a leader in education, an institution must be founded on two basic principles: keeping the interest of students as its foremost priority and recognizing that knowledge is ever-growing, omnipresent and requires the opening of minds. The Mangalayatan University take pride in these two qualities being at the core of its existence. Mangalayatan (translated as the germinal bed of positive thought) aims to perpetuate a culture of hard work, embracing change and preparing oneself for the fast progressing world with the latest skills and the highest level of social and personal values. Situated 20 km from the Yamuna Expressway in Uttar Pradesh, the University founded in 2006, has graduated over 10,000+ students. Nearly 3500+ students from India and abroad are currently studying on campus. Mangalayatan University is vision is to give students from all kinds of background, a quality educational experience leading to legitimately rewarding career opportunities.

ABOUT THE DOCTOR'S KRISHI EVAM BAGWANI VIKAS SANSTHA, LUCKNOW, UTTAR PRADESH, INDIA

Doctor's Krishi Evam Bagwani Vikas Sanstha (Doctor's Agriculture and Horticultural Development Society), a Government Registered society was established with and objective of bringing new Agricultural and Horticultural Technology by 'Eco-friendly' approaches in 1999 at Lucknow. The society was established with an idea of Farmers Field program, Kisan Shiqayat' & Organizing conferences at National as well as International levels. Later in 2006, the society started publication of a biannual scientific journal the 'Journal of Eco-friendly Agriculture' devoted to basic and applied aspect of agriculture, horticulture, environmental science and human ecology.

INAUGURATION SESSION

The inauguration function of the international web conference, held on 01 to 02 September, 2021, was jointly inaugurated by Prof. K.V.S.M Krishna, Hon'ble Vice Chancellor, Prof. Ullas Gurudas, Director Research & Academics of Mangalayatan University and Dr. R.P.Srivastava, Ex-Director, CISH, Lucknow. Other dignitaries present on the web dice were Dr. Prashant Kaushik, Vegetable Breeder at Kikugawa Research Station, Nagano University, Yokohama Ueki, Shizuoka, Japan, Dr. Balwant Singh, Professor and Head, Department of Land Resources and Environment, HAC, Eritrea, North East Africa and Dr. Anurag Malik Scientist at Punjab Agriculture University, Punjab. Thereafter, lighting of lamp was done by all guest.

Dr. Syed Danish Yaseen Naqvi, the convenor of the conference on behalf of the organizing committee welcomed the Chief Guest, Hon'ble Vice Chancellor Prof. K.V.S.M Krishna and the eminent scientists, participants and the faculty of Mangalayatan University. He also welcomed the co-organizers of the conference, Dr. R. P. Srivastava, General Secretary, Doctors Krishi Evam Bagwani Vikas Sanstha, Lucknow, Uttar Pradesh and Dr. Sdy. Nagar — — General Secretary, Krishi Rakshak Samiti, Uttar Pradesh and thanked them for their tremendous support to the conference.

Later, the Chief Guest, Hon'ble Vice Chancellor welcomed the large gathering of scientists, researchers, professors, external agencies, entrepreneurs and students attending the conference. He also congratulated the organizers of the events for holding this conference on current scenario.

The anchor, Dr. Akanksha Singh, Assistant Professor, Department of Agriculture, Mangalayatan University, Aligarh started the sessions.

01 September 2021

Session-I: Keynote Lectures

Chairman: Dr. R. P. Srivastava

Rapporteurs: Dr. Syed Danish Yaseen Naqvi and Dr. Yadvendra Singh

Keynote Lectures

- Using Traditional, biotechnological and Genomics Method for Eggplant (*Solanum melongena*). Breeding with a focus on Bioactive Phenolics- Dr.PRASHANT KAUSHIK
- Spatio-temporal trends analysis of seasonal and annual rainfall-DR. ANURAG MALIK
- Integrated Nutrient Management-Dr. Balwant Singh

Dr. Prashant Kaushik,Vegetable Breeder at Kikugawa Research Station, Nagano University,Yokohama Ueki, Shizuoka, Japan spoke about the eggplant (Solanum melongena L.), which is one of the ten vegetables rich in bioactive chemicals. The most significant nutraceutical component in eggplant is chlorogenic acid, a phenolic acid produced from cinnamic acid. The phenolics in eggplant

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fruit are linked to browning and flesh colour. Dr. Kanshik stated that the enhancement of eggplant fruit phenolics can be achieved through traditional, biotechnological, or genomic breeding methods. The genetics of key morphological and biochemical eggplant characteristics was similarly restricted. Agroinfiltration experiments on eggplant fruit were also not standardised. To describe interspecific hybrids of eggplant with wild species, he used 27 morphological and 20 fruit morphometric descriptors based on the phenomics programme Tomato Analyzer. The three groups showed substantial variations, with hybrids having mid 12 values for most characteristics. Overall, his work offered morphological and biochemical information on wild eggplant cousins.

Dr. Anurag Malik, Scientistat Punjab Agriculture University,Punjab spoke about the spatio-temporal patterns of trends on seasonal (pre-monsoon, monsoon, post-monsoon, and winter) and annual rainfall time-series data (1966-2015) of 13 stations located in the central Himalayan region of Uttarakhand State of India. He analyzed using the recently proposed Innovative Trend Analysis (ITA) method with 5 per cent significance level. The results of the ITA method were compared with the traditional Mann-Kendall (MK) test at 5 per cent significance level. The spatial variation of the trends in seasonal and annual rainfall series was interpolated using the Thiessen polygon (TP) method in the ArcGIS 10.2 environment. The results revealed that the trend detected by the MK test (significant positive in 3-time series and significantly negative in 6-time series) can be effectively identified using ITA (significant positive in 19-time series and significant negative in 32-time series). The ITA method could detect some trends that cannot be observed by the MK test.

Dr. Balwant Singh, Professor and Head, Department of Land Resources and Environment, HAC, Eritrea, North East Africa opined that the Green revolution technologies, though have increased the food grain production of India about six times during the last 54 years, it has simultaneously created a problem of nutrient imbalance in soils due to extra mining. This has to be checked in order to maintain the soil health through integrated use of organic manures and fertilizers found promising in maintaining soil health and helping in sustaining crop productivity.

Recommendations

- KVKs, fertilizer industry and NGOs should educate the farmers about the usefulness of INM and balanced use of fertilizers along with manures, crop residues, biofertlizers and green manuring
- The financial agencies should provide easy loans to farmers to adop to INM.
- Information for preparing good quality compost should be transferred to farmers.

Technical Session I (Poster Presentation)

Theme: National Resource Management, Food and Environmental Security

Chairperson: Dr. Prasant Kaushik and Co-chairman Dr. Anurag Malik

Rapporteurs: Dr. Manjri Shukla and Dr. Yadvendra

- **Effect of Domestic Wastewater on the growth parameters of *Lens esculentum* of Srinagar Garhwal Uttarakhand- Anjali Gusain,** Research Scholar Department of Biochemistry, H.N.B. Garhwal University
- **Analysis of different classification algorithms used for the classification on three species of *Iris Limniris* (Tausch) spach dataset-PrannovJamadagni,** Manipal University Jaipur, India

Technical Session II (Oral Presentation)

Theme: Life Sciences, Biomedical Sciences and Biotechnological aspects and AGRI and Animal Husbandry Start-ups

Chairperson: Dr. Prasant Kaushik

Rapporteurs: Dr. Anurag Malik

- **A Study on Constraints Faced by Tribal Women in Crop Based Livelihood Activities-Asha Dagar,** College Of Community And Applied Sciences, Mpuat, Udaipur, Rajasthan)
- **Quality Analysis Of The Extruded Food Prepared With The Green Banana Flour-Dipanshu Ranjan,**Department Of Food Technology, Harcourt Butler Technical University, Kanpur, Up
- **Design Of Solar Tray Tomato Drier-Dr. Jawed Ahmad Rizawi,**Mai-Nefhi College Of Engineering And Technology, Eritrea (East Africa)
- **Formulation and Characterization of Oil Powder using Refractance Window Drying- Nandita,** Harcourt Butler Technical University, Kanpur, Uttar Pradesh-208002, India
- **Effect of Drying Methods on Quality of Amla Powder-Pushpendraverma,**Harcourt Butler Technical University, Kanpur, Up

- **Training an Intervention for Agricultural Mindset – A Case Study on Apple Growers in Ukhrul, Manipur-Rakesh Kumar Singh,** Research Scholar – Mangalayatan University, Beswan – Aligarh, Uttar Pradesh
- **Development of Ultrasonicated Fermented Wheat Starch Edible Film-Shweta Sachan,** Harcourt Butler Technical University, Kanpur, Uttar Pardesh-208002, India
- **Agricultural waste as bio-adsorbent-Sugandha Katiyar,**Harcourt Butler Technical University, Kanpur, Uttar Pardesh-208002, India
- **Study on Development and Characterization of Specialty Extruded Food (Pasta) Incorporated with Jackfruit Flour-Vikas Yadav,** Harcourt Butler Technical University, Kanpur, Uttar Pardesh-208002, India.

02 September 2021

Session-II: Oral Presentation

Chairman: Dr. Syed Danish Yaseen Naqvi

Rapporteurs: Dr. Akanksha Singh and Dr. Manjri

Keynote Lectures

· **History, Origin, Nomenclature and Integrated Insect Pest of Mango- Dr. R. P. Srivastava, Ex. Director, CISH Lucknow (U.P.)**

- **Role of economical agroforestry tree species for improving income of farmers-Dr. Vinita Bisht**
- **Crop responses to abiotic stress and adaptation/ tolerance mechanisms-Dr. Ashutosh Srivastava**

Dr. R. P. Srivastava in his keynote this lecture gave detailed information on nomenclature, history, orgine of mango and many usefuly details of integrated pest management of mango crop.

Dr. Ashutosh Srivastava, College of Agriculture, Rani Lakshmi Bai Central Agricultural University, Jhansi (U.P) spoke about abiotic stresses like high temperature, drought, flooding, salinity. He also awared us about the consequences of abiotic stress. He reported that heavy metal are the major yield-limiting factors for crop plants as these affect plant growth, development, yield, and seed quality. Drought and heat must be examined together because their combined impact is greater than when they were considered separately.Among abiotic stresses, drought and heat stress are two important abiotic factors that limit crop output. However, a great gap exists in knowledge about the level of stress tolerance to developed in crops intended to be grown on a targeted environment. Such kind of knowledge will certainly be helpful in prioritizing traits/selection criteria and developing screening techniques for improved stress tolerance.

Dr. Vinita Bisht, Assistant professor, Department of Silviculture and Agroforestry, College of Forestry, Banda University of Agriculture & Technology,Banda, U.P., spoke about the role of economical agroforestry tree species for improving farmer's income. She also spoke about suitable agroforestry tree species for Bundelkhand and informed their economic value for raising farmer's income.

Technical Session III (Poster Presentation)

Theme: Innovative Approach in Agricultural and Allied Sciences, Life Sciences, Biomedical Sciences and Biotechnological Aspects and AGRI and Animal Husbandry Start-ups

Chairperson: Dr. Prasant Kaushik and Dr. Anurag Malik

Rapporteurs: Dr. Akanksha and Dr. Yadvendra

Use Soil And Water Assessment Tool (Swat) Model For Delineation And Hydrological Response Units (Hrus) Analysis Of The River Basin-Bhagyashri R. Jalgaonkar, Department of Soil and Water Engineering, CTAE (MPUAT), Udaipur, Rajasthan, India.

Hydrological modelling of a Himalayan Watershed Using SWAT-Bhagwat Saran, Research Scholar, College of Technology, Govind Ballabh Pant University of Agricultural and Technology,Pantnagar, Uttarakhand, India**.**

Technical Session IV (Oral Presentation)

Theme: Innovative Approach in Agricultural and Allied Sciencesand Natural Resource Management, Food and Environmental Security

Chairperson: Dr. Prasant Kaushik and Dr. Anurag Malik

Rapporteurs: Dr. Manjri Shukla and Er. Ashish Jain

A Study On Constraints Faced By Tribal Women In Crop Based Livelihood Activities-Asha Dagar, (Research Student) College Of Community And Applied Sciences, Mpuat, Udaipur, Rajasthan.

Development and performance evaluation of tractor mounted shielded sprayer for weed control in wide row crops-Babasaheb S. Gholap, Department of Farm Machinery and Power Engineering, College of Technology and Engineering,,Maharana Pratap University of Agriculture & Technology, Udaipur – 313 001 (Rajasthan).

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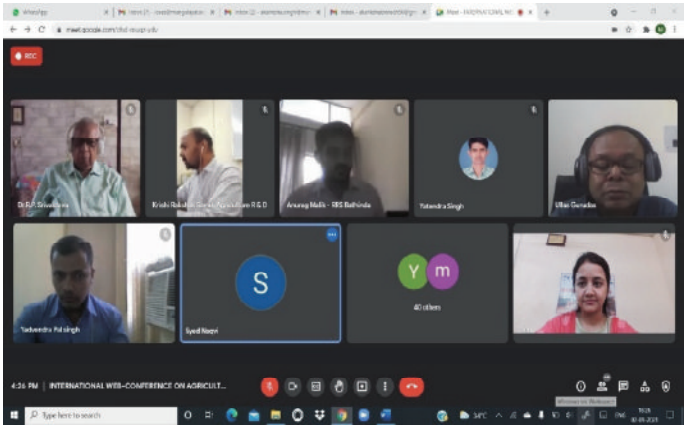
Predication of yield ad biomass for Rice crop through AquaCrop Model- Vikas Sharma, Assistant professor, lovely professional university jaladhar.

Development and Characterization of Ultrasonicated Wine Powder Using Freeze Drying and Refractance Window Drying- Shubham Kumar Rai, Department of Food Technology, Harcourt Butler Technical University, Kanpur, Uttar Pardesh-208002, India.

Development and performance evaluation of pedal operated maize sheller-Harish DigamberNahate, Assistant Professor, college of Agriculture Engineering ,JaLgaonJamod, Maharashtra**.**

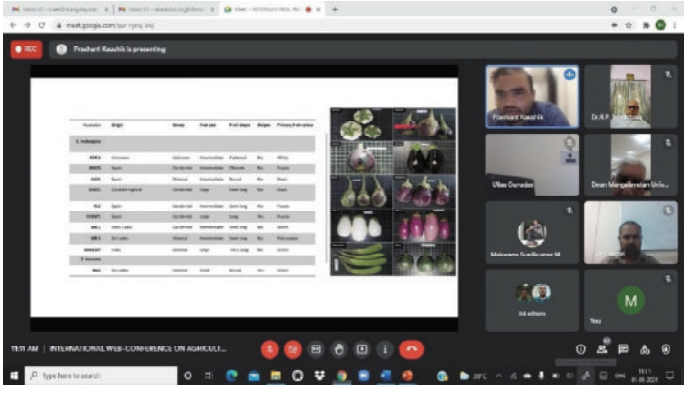
Formulation and Characterization of Protein Enriched Mango Bar Using Refractance Window Drying and Freeze Drying-Shraddha Tripathi, Department of Food Technology, HBTU, Kanpur.

GLIMPSES OF INTERNATIONAL WEB CONFERENCE ON AGRICULTURE AND ENVIRONMENTAL SCIENCES (ICAES-2021)

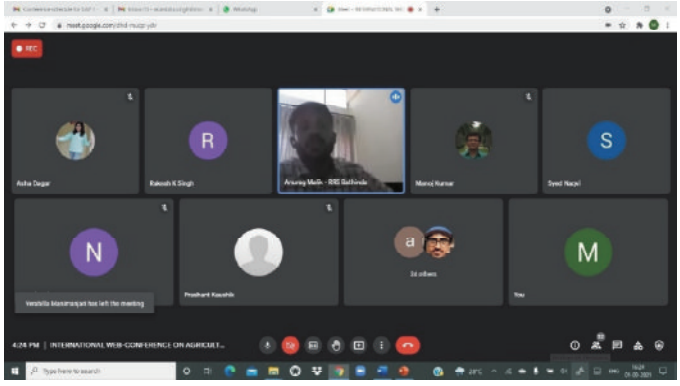


Welcome address by Dr. Syed Danish Yaseen Naqvi, HOD and convenor, Department of Agriculture, Mangalayatan University, Beswan, Aligarh

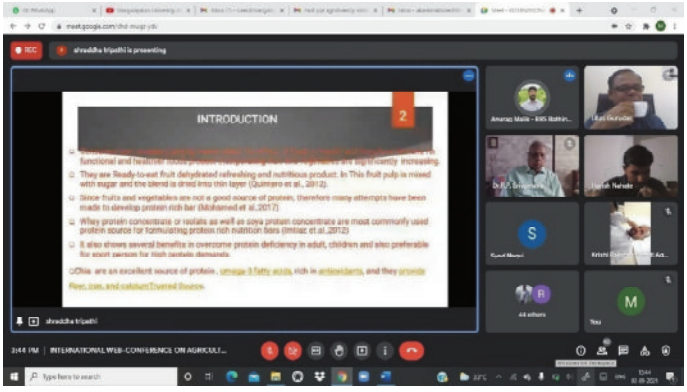
Chief Guest, Hon'ble Vice Chancellor Prof. K.V.S.M Krishna,Mangalayatan University, Beswan, Aligarh



Dr. Prashant Kaushik,Vegetable Breeder at Kikugawa Research Station, Nagano University,Yokohama Ueki, Shizuoka, Japan prenting keynote lecture



Dr. Anurag Malik,Scientist at Punjab Agriculture University, Punjab presenting keynote lecture



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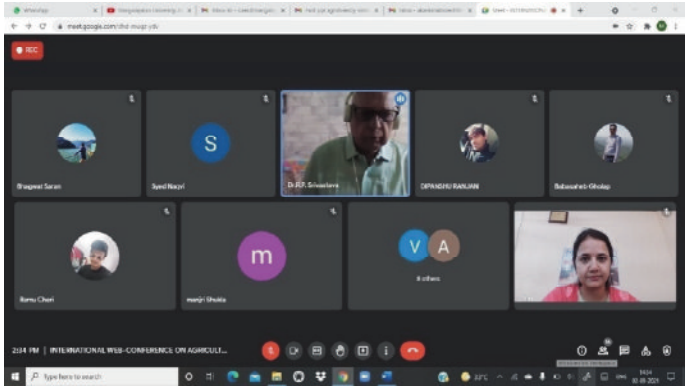
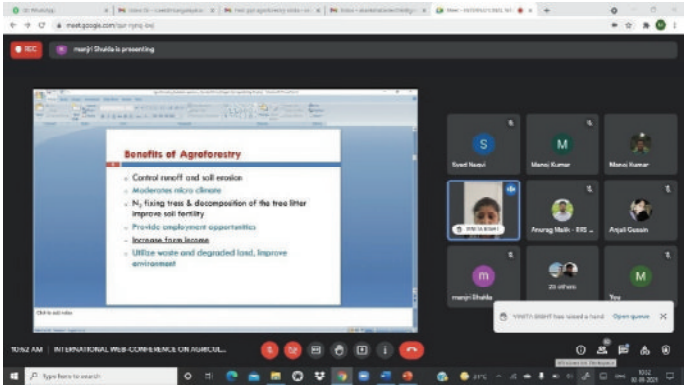


Dr. R. P. Srivastava, Ex-Director, CISH, Lucknow sharing his views on the conference



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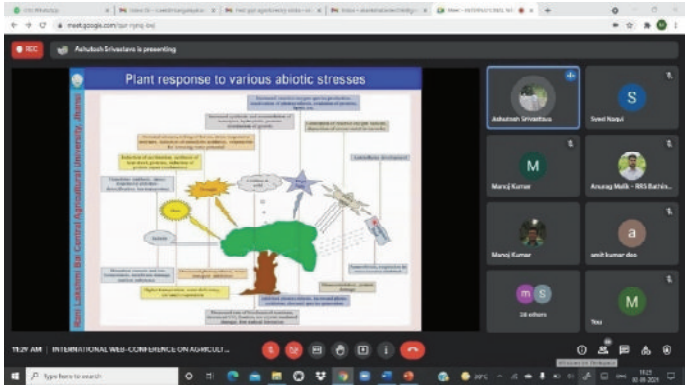

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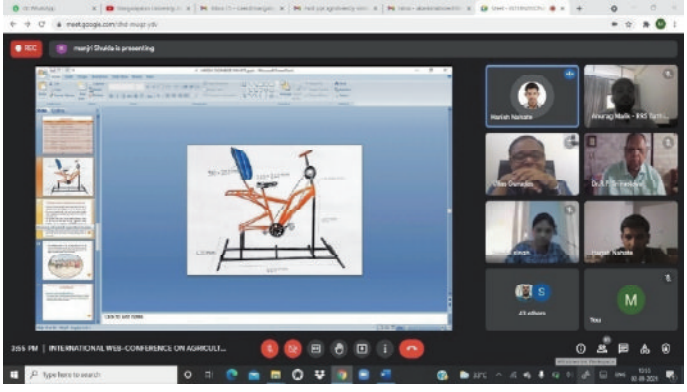
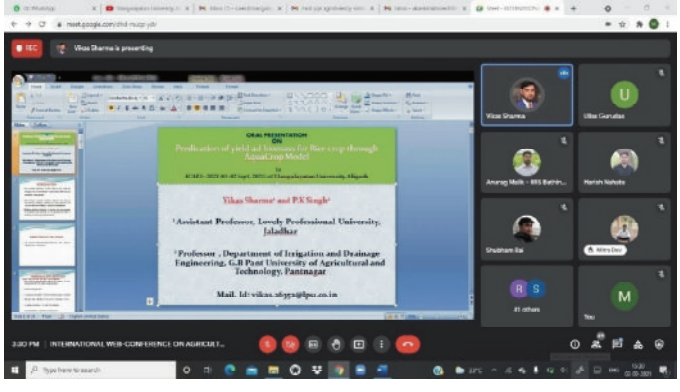


Oral Presentation by Harish Nahate



Oral presentation by Vikas Sharma

Valedictory Function

Dr. Syed Danish Yaseen Naqvi , Convener welcomed Hon'ble Vice Chancellor, Professor KVSM Krishna, Director Research, Registrar, Joint Registrar Dean and HOD, colleagues and all the distinguished guests, delegates, ladies and gentlemen. He, on behalf of Organizing committee of Department of Agriculture MU Association, Doctor Krishi Evam Bagwani Vikas Sanstha and krishi Rakshak Samiti thanked Prof. KVSM Krishna for accepting the invitation for chairing the session in valedictory function. He felt privileged to welcome them to the valedictory function of the Conference ON "AGRICULTURE AND ENVIRONMENTAL SCIENCES (ICAES-2021)" September 01-02, 2021. He expressed a great need to have effective mechanism of coordination and convergence between institutions at the national, regional and global levels. The conference organizing committee members Dr. Akanksha, Dr. Manjri, Dr. Yadvendra, Professor Suresh Tiwari, Dr. R. P Srivastava and others decided to have a way forward in the context of continuity by holding International Conference on Agriculture under the umbrella of Mangalayatan University every year. Dr. Naqvi thanked the key note speakers and speakers for sparing their valuable time and giving presentations on current scenario.

More than 200 participants from different countries attended the conference. The conference covered more than 20 oral and poster presentations.

Following best oral and poster presentation were awarded.

DAY – 1

TECHNICAL SESSION

ORAL PRESENTATION

1.	RAKESH K SINGH	First
2.	NANDITA	Second

POSTER PRESENTATION

1.	PRANNOV JAMADAGNI	First
2.	ANJALI GUSAIN	Second

Day – 2

TECHNICAL SESSION

POSTER PRESENTATION

1.	BHAGYASHRI. R. JALGAONKAR	First
2.	BHAGWAT SARAN	Second

ORAL PRESENTATION

1.	HARSH NAHATE	First
2.	VIKAS SHARMA	Second

The session ended with recitation of national anthem by Dr. Akanksha and Dr. Manjri.

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Effect of liquid and carrier based biofertilizers on flowering attributes, days to harvest and benefit-cost ratio in Tomato

K. LAKSHMIKALA[1], B. RAMESH BABU[2], M. RAVINDRA BABU[3], P. RAMA DEVI[4]

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[4]*P. Rama Devi, ramadevipuvvada@yahoomail.com*

ABSTRACT

The experiment conducted at Horticultural Research Station, Dr. YSR Horticultural University, Venkataramannagudem, West Godavari with nine treatment combinations and three replications showed no significant difference in days to first flowering, days to 50 per cent flowering and days to first harvest in all the treatment combinations. Whereas, the application of 100 per cent RDF along with liquid formulations of NFB, PSB and KSB showed significant difference in days to final harvest as compared to other treatment combinations. The application of 80 per cent RDF along with liquid formulations of NFB, PSB and KSB recorded in significantly higher number of flowers per plant, number of clusters per plant, yield ha^{-1} and the benefit-cost ratio.

Key words: liquid formulations, treatment combinations, flowering attributes, days to harvest, benefit-cost ratio.

Tomato (*Solanum lycopersicum* L.) is an important annual vegetable crop grown throughout the world and ranks second in importance after potato. It belongs to the genus *Solanum* under Solanaceae family. Tomato is a herbaceous sprawling plant growing to a height of 1-3 m with a weak woody stem. The flowers are yellow in colour and the fruits vary in size from cherry tomatoes, about 1–2 cm in size to beefsteak tomatoes, about 10 cm or more in diameter in cultivated varieties. Most cultivars produce red fruits when ripe. The tomato is believed to have been originated in Central Africa and South America (Vavilov, 1951) [13]. In India it is an introduced crop and in being grown on an area of 789.15'000 hectare with an annual production of 19759.32'000 metric tonnes (source NHB database, 2017-18).

The use of N-fertilizer not only spoils the ground water, soil but also have deleterious effects by the emission of harmful gases. The chemical fertilizers should be replaced with the natural and organic fertilizers which can play a key role of the conservation of the environment (Jangral and Lakra, 2014) [4]. Bio fertilizers improve the quantitative and qualitative features of many plants (Yosefi *et al*. 2011) [14]. Biofertilizers used in conjunction with chemical fertilizers improve crop productivity and nutrient use efficiency. It is becoming difficult to meet the nutrient need of farming through chemical fertilizer alone and due to its higher costs, the concept of integrated plant nutrient supply system (IPNS) is gaining ground. Therefore, the investigation was planned and conducted to study the influence of liquid and carrier based biofertilizers on growth, yield and quality of tomato (*Solanum lycopersicum* L.).

MATERIAL AND METHODS

The experiment was conducted during *rabi*, 2018 at Horticultural Research Station, Venkataramannagudem, West Godavari District, Andhra Pradesh a it was laid out in Factorial RBD with three replications. The first factor consisted of three levels of RDF (100 %, 80 % and 60 % of RDF) and the second factor consisted of three levels of different combinations of biofertilizers (NFB + PSB + KSB liquid biofertilizer, NFB + PSB + KSB carrier based biofertilizer and without biofertilizers) comprising nine treatment combinations viz., T_1 (100 % RDF + NFB + PSB + KSB carrier based biofertilizers), T_2 (100 % RDF + NFB + PSB + KSB liquid biofertilizers), T_3 (100 % RDF + NFB + PSB + KSB without biofertilizers), T_4 (80 % RDF + NFB + PSB + KSB carrier based biofertilizers), T_5 (80 % RDF + NFB + PSB + KSB liquid biofertilizers), T_6 (80% RDF + without biofertilizers), T_7 (60 % RDF + NFB + PSB + KSB carrier based biofertilizers), T_8 (60 % RDF + NFB + PSB + KSB liquid biofertilizers), T_9 (60 % RDF + without biofertilizers). The seedlings were transplanted at a spacing of 120 cm x 50 cm in a single row.

RESULTS AND DISCUSSION

Days to first flowering

Different levels of chemical fertilizer application influenced the days taken to first flowering. The time taken for first flowering was 29.11 days in F_1 (100 % RDF) followed by F_2 (80 % RDF) with 29.33 and it was 31.00 days in F_3 (60 % RDF). Days to first flowering ranged from 27.67 to 33.44 with the application of biofertilizers and minimum (27.67) number of days to first flowering was obtained with the application of liquid biofertilizers followed by the application of carrier based biofertilizers (28.33) over control (33.44) days (table 1). It was observed that there was no significant differences among the interactions on days to first flowering.

This may be attributed to the fact that the same variety (Arka Samrat) was used in all the treatments of the experiment and synchronized flowering was observed in all the treatments hence, there was no significant differences observed in days to first flowering and these results are in concurrence with (Direkvandi *et al*., 2008) [2] in tomato.

Days to 50 per cent flowering

Application of different levels of chemical fertilizers influenced the time taken for 50 per cent flowering and it ranged from 33.78 to 36.44. The time taken for 50 per cent flowering (33.78) days was minimum with the application of 80 per cent RDF followed by 100 per cent RDF application (34.00) while, maximum days was recorded in 60 per cent RDF application (36.44) (table 2). The plants treated with NFB, PSB and KSB as liquid biofertilizers recorded minimum number of days to 50 per cent flowering (32.55) followed by the application of mixture of NFB, PSB and KSB as carrier based biofertilizers with 33.33 days when compared to without biofertilizers (control) (38.33 days). Among the interactions, there was no significant differences in days taken to 50 per cent flowering. This may be attributed to the fact that single variety Arka Samrat was used in the experiment and hence, no significant differences observed in time taken for 50 per cent flowering. The results were similar to the findings of (Geetharani and Prathiban, 2014) [3] in tomato.

Table 2. Effect of biofertilizers in combination with different levels of NPK on days to 50% flowering in tomato hybrid Arka Samrat

Recommended dose of fertilizers (200:250:250 kg NPK/ha)	Biofertilizers			
	B_1	**B_2**	**B_3**	**Mean**
F_1	29.67	29.33	29.67	29.56
	(5.54)	(5.51)	(5.54)	(5.53)
F_2	29.67	28.67	29.33	29.22
	(5.54)	(5.47)	(5.51)	(5.50)
F_3	30.33	29.33	30.00	29.89
	(5.60)	(5.51)	(5.57)	(5.56)
Mean	29.89	29.11	29.67	
	(5.56)	(5.49)	(5.54)	
Factors	**F**	**B**	**F × B**	
SE (m) ±	-	-	-	
C.D at 5 %	NS	NS	NS	

(Figures in parenthesis indicates square root transformed values)
F_1- 100 per cent recommended dose of fertilizers; F_2- 80 per cent recommended dose of fertilizers; F_3- 60 per cent recommended dose of fertilizers
B_1- NFB + PSB + KSB (carrier based biofertilizers); B_2- NFB + PSB + KSB (liquid biofertilizers); B_3- without biofertilizer

Table 1. Effect of biofertilizers in combination with different levels of NPK on days to first flowering in tomato hybrid Arka Samrat

Recommended dose of fertilizers (200:250:250 kg NPK/ha)	Biofertilizers			
	B_1	**B_2**	**B_3**	**Mean**
F_1	26.33	26.33	26.33	26.33
	(5.23)	(5.23)	(5.23)	(5.23)
F_2	26.66	26.66	26.66	26.66
	(5.26)	(5.26)	(5.26)	(5.26)
F_3	27.00	27.00	27.00	27.00
	(5.29)	(5.29)	(5.29)	(5.29)
Mean	26.66	26.66	26.66	
	(5.26)	(5.26)	(5.26)	
Factors	**F**	**B**	**F × B**	
SE (m) ±	0.02	-	-	
C.D at 5 %	0.05	NS	NS	

(Figures in parenthesis indicates square root transformed values)
F_1- 100 per cent recommended dose of fertilizers; F_2- 80 per cent recommended dose of fertilizers and F_3- 60 per cent recommended dose of fertilizers
B_1- NFB + PSB + KSB (carrier based biofertilizers); B_2- NFB + PSB + KSB (liquid biofertilizers); B_3- Without biofertilizer

Journal of Eco-friendly Agriculture **17**(1) 2022
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Number of flowers per cluster

Number of flowers per cluster was significantly influenced by the application of chemical fertilizers, biofertilizers and their interactions. Number of flowers/ cluster ranged from 6.82 to 7.88 when chemical fertilizers at different levels were applied. Significantly more number of flowers/cluster (7.88) was recorded with the application of 80 per cent RDF followed by the application of 100 per cent RDF (7.54) when compared to 60 per cent RDF (6.82).

Table 3. Effect of biofertilizers in combination with different levels of NPK on number of flowers per cluster in tomato hybrid Arka Samrat

Recommended dose of fertilizers (200:250:250 kg NPK/ha)	Biofertilizers			
	B_1	B_2	B_3	Mean
F_1	7.90	8.13	6.60	7.54
	(2.98)	(3.02)	(2.76)	(2.92)
F_2	8.36	8.86	6.43	7.88
	(3.06)	(3.14)	(2.73)	(2.98)
F_3	7.03	7.24	6.20	6.82
	(2.83)	(2.87)	(2.68)	(2.80)
Mean	7.76	8.07	6.41	
	(2.96)	(3.01)	(2.72)	
Factors	F	B	F × B	
SE (m) ±	0.02	0.02	0.03	
C.D at 5 %	0.05	0.05	0.08	

(Figures in parenthesis indicates square root transformed values)
F_1- 100 per cent recommended dose of fertilizers; F_2- 80 per cent recommended dose of fertilizers; F_3- 60 per cent recommended dose of fertilizers
B_1- NFB + PSB + KSB (carrier based biofertilizers); B_2- NFB + PSB + KSB (liquid biofertilizers); B_3- without biofertilizer

Significant increase (25.89 %) in number of flowers/ cluster was recorded with the application of liquid biofertilizers (NFB, PSB and KSB) compared to without biofertilizers followed by the application of carrier based biofertilizers (NFB, PSB and KSB) with an increase of 21.06 per cent (table 3). Among the interactions, 80 per cent RDF along with NFB, PSB and KSB as liquid formulation (F_2B_2) recorded the highest (8.86) number of flowers/cluster over the other treatment combinations followed by (F_2B_1) 80 per cent RDF along with NFB, PSB and KSB carrier based biofertilizers (8.36) while, the least (6.20) number of flowers/ cluster was recorded in (F_3B_3) 60 per cent RDF + without biofertilizers and these results may be attributed due to increased supply of major plant nutrients which are required in larger quantities for growth and development of plants. Nitrogen accelerates the development of growth and reproductive phases and protein synthesis, thus promoting yield attributing characters.

Number of clusters plant^{-1}

Significant differences in number of clusters plant^{-1} was recorded with the application of chemical fertilizers, biofertilizers and their interactions. In tomato the application of different levels of chemical fertilizers greatly influenced the number of clusters plant^{-1} which ranged from 6.78 to 8.56 (table 4). Significant higher number of clusters/plant (8.56) was recorded with the application of 80 per cent RDF followed by the application of 100 per cent RDF with (8.07) as against (6.78) in 60 per cent RDF. There was 23.37 per cent significant increase in number of clusters/plant with the application of NFB + PSB + KSB as liquid biofertilizers when compared to control (without biofertilizers) followed by the application of carrier based biofertilizers with 14.57 per cent increase in number of clusters plant^{-1}. Among all the interactions, application of 80 per cent RDF along with NFB, PSB and KSB liquid biofertilizers (F_2B_2) recorded the maximum number of clusters/plant (9.90) when compared to other treatment combinations followed by (F_2B_1) 80 per cent RDF along with NFB, PSB and KSB carrier based biofertilizers (8.72) and the lowest (6.31) was recorded in F_3B_3 60 per cent RDF + without biofertilizers. Application of biofertilizers increases the vegetative growth by production of growth substances and promotes yield characters and the data obtained was similar to the findings of (Meena *et al*., 2014; Prativa and Bhattarai, 2011 and Singh *et al*., 2017) [7,9&11] in tomato.

Table 4. Effect of biofertilizers in combination with different levels of NPK on number of clusters per plant in tomato hybrid Arka Samrat

Recommended dose of fertilizers (200:250:250 kg NPK/ha)	Biofertilizers			
	B_1	B_2	B_3	Mean
F_1	8.30	8.53	7.40	8.07
	(3.05)	(3.09)	(2.90)	(3.01)
F_2	8.72	9.90	7.07	8.56
	(3.12)	(3.30)	(2.84)	(3.09)
F_3	6.80	7.23	6.31	6.78
	(2.79)	(2.87)	(2.70)	(2.79)
Mean	7.94	8.55	6.93	
	(2.99)	(3.09)	(2.82)	
Factors	F	B	F × B	
SE (m) ±	0.03	0.03	0.05	
C.D at 5 %	0.08	0.08	0.15	

(Figures in parenthesis indicates square root transformed values)
F_1- 100 per cent recommended dose of fertilizers; F_2- 80 per cent recommended dose of fertilizers; F_3- 60 per cent recommended dose of fertilizers
B_1- NFB + PSB + KSB (carrier based biofertilizers); B_2- NFB + PSB + KSB (liquid biofertilizers); B_3- without biofertilizer

Days to first harvest

There was no influence on days to first harvest with the application of different levels of chemical fertilizers and their interactions with biofertilizers. Application of liquid biofertilizers (NFB, PSB and KSB) recorded the minimum (68.89) days to first harvest followed by the application of

carrier based biofertilizers (NFB, PSB and KSB) with 69.66 days when compared to without biofertilizers (71.22) (table 5). As the biofertilizers show their effect slowly and same variety was used in the experiment there was no difference regarding days to first harvest and the data regarding days to first harvest was found to be similar with the data obtained by (Kamal *et al.*, 2018) [5] in tomato.

Table 5. Effect of biofertilizers in combination with different levels of NPK on days to first harvest in tomato hybrid Arka Samrat

Recommended dose of fertilizers (200:250:250 kg NPK/ha)	Biofertilizers			
	B_1	B_2	B_3	Mean
F_1	69.33 (8.39)	68.33 (8.33)	70.67 (8.46)	69.44 (8.39)
F_2	69.00 (8.36)	68.00 (8.31)	71.00 (8.49)	69.33 (8.39)
F_3	70.66 (8.47)	70.33 (8.45)	72.00 (8.54)	70.10 (8.48)
Mean	69.66 (8.41)	68.89 (8.36)	71.22 (8.50)	
Factors	F	B	F × B	
SE (m) ±	-	0.03	-	
C.D at 5 %	NS	0.09	NS	

(Figures in parenthesis indicates square root transformed values)
F_1- 100 per cent recommended dose of fertilizers; F_2- 80 per cent recommended dose of fertilizers; F_3- 60 per cent recommended dose of fertilizers
B_1- NFB + PSB + KSB (carrier based biofertilizers); B_2- NFB + PSB + KSB (liquid biofertilizers); B_3- without biofertilizer

Days to final harvest

The data pertaining to days to final harvest as influenced by chemical fertilizers, biofertilizers and their interactions as presented in table 6 and there was significant difference in days to final harvest due to application of chemical fertilizers, biofertilizers and their interactions in tomato. Days to final harvest ranged from 139.11 to 154.67 due to application of different levels of chemical fertilizers and significant more (154.67) number of days to final harvest was obtained with the application of 100 % RDF followed by 80 per cent RDF application with 147.00 days over 60 per cent RDF (139.11).

The application of biofertilizers also showed significant influence on days to final harvest and it varied from 139.33 to 151.78. The maximum (151.78) days to final harvest was obtained in plants treated with B_2 (NFB + PSB + KSB liquid biofertilizers) followed by B_1 (NFB + PSB + KSB carrier based biofertilizers) with 149.67 days when compared to control *i.e.*, without biofertilizers (139.33). The interaction effect of 100 per cent RDF along with NFB, PSB and KSB liquid biofertilizers (F_1B_2) recorded the maximum (160.00) days to final harvest when compared to other interactions followed by 100 per cent RDF and NFB, PSB and KSB carrier based biofertilizers (F_1B_1) with 158.67 days and the minimum (135.67) days to final harvest was recorded in F_3B_3 60 per cent RDF and without biofertilizers. Application of biofertilizers prolongs the growth period which resulted in increased crop duration in tomato crop.

Table 6. Effect of biofertilizers in combination with different levels of NPK on days to final harvest in tomato hybrid Arka Samrat

Recommended dose of fertilizers (200:250:250 kg NPK/ha)	Biofertilizers			
	B_1	B_2	B_3	Mean
F_1	158.67 (12.63)	160.00 (12.69)	145.33 (12.10)	154.67 (12.47)
F_2	150.00 (12.29)	154.00 (12.45)	137.00 (11.75)	147.00 (12.16)
F_3	140.33 (11.89)	141.33 (11.93)	135.67 (11.69)	139.11 (11.84)
Mean	149.67 (12.27)	151.78 (12.36)	139.33 (11.85)	
Factors	F	B	F × B	
SE (m) ±	0.03	0.03	0.04	
C.D at 5 %	0.08	0.08	0.13	

(Figures in parenthesis indicates square root transformed values)
F_1- 100 per cent recommended dose of fertilizers; F_2- 80 per cent recommended dose of fertilizers; F_3- 60 per cent recommended dose of fertilizers
B_1- NFB + PSB + KSB (carrier based biofertilizers); B_2- NFB + PSB + KSB (liquid biofertilizers); B_3- without biofertilizer

Fruit yield per hectare (t ha^{-1})

There was significant difference on yield ha^{-1} in tomato due to application of chemical fertilizers, biofertilizers and their interactions (table 7). Chemical fertilizers with different levels influenced the yield ha^{-1} and it ranged from 96.10 t ha^{-1} to 109.83 t ha^{-1}. The maximum yield ha^{-1} (109.83 t ha^{-1}) was recorded in F_2 (80 % RDF) followed by F_1 (100 % RDF) with 107.19 t ha^{-1} and it was noticed that minimum yield (96.10 t ha^{-1}) was recorded in F_3 (60 % RDF). The tomato plants treated with biofertilizers showed significant influence yield ha^{-1}.

Significant increase in yield ha^{-1} (17.96 %) was recorded with NFB + PSB + KSB liquid biofertilizers (B_2) over control (without biofertilizers) followed by B_1 (NFB + PSB + KSB carrier based biofertilizers) which showed an increase of 11.66 per cent when compared to control. Different levels of chemical fertilizer and biofertilizer interactions showed significant influence on yield ha^{-1}. Among all the interactions the interaction with 80 per cent RDF + NFB + PSB + KSB liquid biofertilizers (F_2B_2) recorded significant higher yield (121.16 t/ha) followed by (F_2B_1) 80 per cent RDF and carrier

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based biofertilizers (113.83 t ha^{-1}) while, it was recorded lowest (92.16 t ha^{-1}) in 60 per cent RDF + without biofertilizers (F_3 B_3). These results might be attributed due to the application of biofertilizers, secretion of ammonia also enhanced in the rhizosphere which enhance the nutrient uptake potential of plant and improved the fruit yield and the experimental data regarding fruit yield was similar with the findings of Chowdhury *et al.* (2015) and Sudhakar and Purushotham, (2008) [1&12] in tomato and Sable *et al*. (2016) [10] in cauliflower.

Table 7. Effect of biofertilizers in combination with different levels of NPK on yield per hectare (t/ha) in tomato hybrid Arka Samrat

Recommended dose of fertilizers	Biofertilizers			
(200:250:250 kg NPK/ha)	B_1	B_2	B_3	Mean
F_1	111.30	114.29	105.38	110.32
F_2	122.00	129.77	101.13	117.63
F_3	102.37	108.91	98.73	103.34
Mean	111.89	117.66	101.75	
Factors	F	B	F × B	
SE (m) ±	0.76	0.76	1.31	
C.D at 5 %	2.29	2.29	3.97	

F_1- 100 per cent recommended dose of fertilizers; F_2- 80 per cent recommended dose of fertilizers; F_3- 60 per cent recommended dose of fertilizers

B_1- NFB + PSB + KSB (carrier based biofertilizers); B_2- NFB + PSB + KSB (liquid biofertilizers); B_3- without biofertilizer

Benefit-cost ratio

The data pertaining to the gross returns, net income and benefit-cost ratio (B:C) as influenced by different levels of chemical fertilizers, biofertilizers and their interactions were presented in table 8. The different treatment combinations showed varying gross returns with respect to their input cost. The highest cost benefit ratio (1:2.96) was obtained in the treatment combination of 80 per cent RDF + NFB + PSB + KSB liquid biofertilizers (F_2B_2) which recorded a gross return of Rs. 4,84,640/- per hectare followed by the combination of 80 per cent RDF + NFB + PSB + KSB carrier based biofertilizers (F_2B_1) (1:2.77) with a gross return of Rs. 4,55,320/- per hectare while, the lowest cost benefit ratio (1:2.37) was recorded with the application of 60 % RDF + without biofertilizers (F_3B_3) which yielded a gross return of Rs. 3,68,640/- per hectare.

The results showed that the higher doses of nutrients provide higher crop yields and improve the benefit cost ratio. The use of 80 per cent RDF + NFB + PSB + KSB liquid biofertilizers (F_2B_2) might have added to the lower cost of cultivation in case of chemical fertilizers, but on the other hand with the use of biofertilizers, it could provide relatively higher yields and also improve the quality characters and also influences the crop growth, yield and reduces the application of 20 per cent chemical fertilizers. The above results were in accordance with Kumar *et al*. (2006) and Narayan *et al*. (2007) [6&8] in tomato.

Table 8. Effect of biofertilizers in combination with different levels of NPK on benefit cost ratio

Treatments	Cost of cultivation (Rs h^{-1})	Gross income (Rs h^{-1})	Net income (Rs h^{-1})	Cost: Benefit ratio
T_1	153667	333900	180233	2.17
T_2	153417	343000	189583	2.23
T_3	152167	316000	163833	2.07
T_4	152333	366000	213667	2.40
T_5	152083	389000	236917	2.56
T_6	150833	303000	152167	2.01
T_7	151050	307000	155950	2.03
T_8	150750	317000	166250	2.10
T_9	149500	296000	146500	1.97

CONCLUSION

It was concluded that application of 80 per cent RDF along with liquid formulations of NFB, PSB and KSB resulted in significantly higher values in flower attributes and days to harvest when compared to other treatment combinations and also resulted in higher cost-benefit ratio.

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Effect of bio-fertilizers on growth, yield and microbial population of Wheat

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ABSTRACT

In an experiment conducted during the *Rabi* season (2014-15), the use of *Azospirillum lipoferum* + 75 per cent RDF of NPK + 25 per cent FYM @ 24 t ha^{-1} recorded significantly higher plant height (86.73 cm), test weight (56.83 g), plant dry weight (26.92 g hill^{-1}), grain yield (6.25 t ha^{-1}) and total microbial count (24.33 × 10^7 CFU g^{-1}) with maximum harvest Index (51.81 %) and a benefit cost ratio (2.00). All the treatments excepting PSB + *A. lipoferum* + 75 per cent NPK + 25 per cent FYM @ 24 t ha^{-1} recorded significantly higher straw yield against the treatment with 100 per cent FYM @ 24 t ha^{-1} Highest straw yield (6.17 t ha-1) was recorded with PSB + *A. chroococcum* + 75 % RDF of NPK + 25 % FYM @ 24 t ha^{-1} which is also at par (5.75 t ha^{-1}) with the treatment comprising *A. lipoferum* + 75 per cent NPK + 25 per cent FYM.

Key-words: Benefit cost ratio, bio-fertilizers, growth, microbial population, wheat (PBW-154), yield.

The first cultivation of wheat occurred about 10,000 years ago, as part of the 'Neolithic Revolution', which saw a transition from hunting and gathering of food to settled agriculture. (Heun *et al*., 1997; Nesbitt, 1998; Dubcovsky and Dvorak, 2007). Cultivation spread to the Near East by about 9000 years ago when hexaploid bread wheat made its ûrst appearance (Feldman, 2001). Wheat cultivation in India started 5000 years ago (Feldman, 2001). It is counted among the 'big three' cereal crops, with over 600 million tonnes being harvested annually. For example, in 2007, the total world harvest was about 607 million tonnes compared with 652 million tonnes of rice and 785 million tonnes of maize (http://faostat.fao.org/).

Soil microbes play an important role in many critical ecosystem processes, including nutrient cycling and homeostasis, decomposition of organic matter, as well as promoting plant health and growth as bio-fertilization (Han et al., 2007). El- Kholy and Gomaa (2000) have succeeded in reducing the recommended doses of chemical fertilizers needed for corn and millet by 50 per cent using bio-fertilizers without loss in the yield. Inoculation of plants with *Azospirillum* could result in significant changes in various growth parameters, such as increase in plant biomass, nutrient uptake, tissue N content, plant height, leaf size and root length of cereals (Bashan et al., 2004). Bio-fertilizers play an important role in sustainable crop production that has been reviewed by several authors (Biswas et al., 1985; Katyalet al., 1994; Wani and Lee, 1995). Keeping in view the importance of environment and human health and how the micro-organisms help in doing so, investigations on understanding the crop growth, yield, economics and also the microbial population of the respected microorganism was aimed at.

MATERIAL AND METHODS

The experiment was conducted during the *Rabi* season 2014-2015 at the Crop Research Farm, Department of Agronomy. It consisted of 9 treatments replicated thrice in a randomized block design *i.e.* T_1: 100 per cent FYM @ 24 t ha^{-1}. T_2: 75 per cent NPK + 25 per cent FYM @ 24 t ha^{-1}. T_3: PSB + 75 per cent NPK + 25 per cent FYM @ 24 t ha^{-1}. T_4: *A. chroococcum* + 75 per cent NPK + 25 per cent FYM @ 24 t ha^{-1}. T_5: *A. lipoferum* + 75 per cent NPK + 25 per cent FYM @ 24 t ha^{-1}. T_6: PSB + *A. chroococcum* + 75 per cent NPK + 25 per cent FYM @ 24 t ha^{-1}. T_7: PSB + *A. lipoferum* + 75 per cent NPK + 25 per cent FYM @ 24 t ha^{-1}. T_8: *A. chroococcum* + *Azospirillum* + 75 per cent NPK + 25 per cent FYM @ 24 t ha^{-1}. T_9: PSB + *A. chroococcum* + *A. lipoferum* + 75 per cent NPK + 25 per cent FYM @ 24 ha^{-1}.

Pre-harvest observations on plant height (cm), number of tillers hill^{-1}, plant dry weight (g hill^{-1}), crop growth rate (g m^{-2} day^{-1}), relative growth rate (g g^{-1} day^{-1}) as well as post-harvest observations on number of effective tillers hill^{-1}, spike length (cm), number of grains spike^{-1}, test weight (g), grain yield (t ha^{-1}), straw yield (t ha^{-1}), harvest index (%) and total bacteria (×10^7 CFU g^{-1}) and also the economic parameters like benefit cost ratio was recorded.

RESULTS AND DISCUSSION

Effect of biofertilizers on growth parameters

The treatment T_5: (*A. lipoferum* + 75 per cent NPK + 25 per cent FYM @ 24 t ha^{-1}) recorded significantly higher plant height (86.73 cm) and plant dry weight (26.92 g hill^{-1}) 24 t ha^{-1}. This indicates that the biofertilizers like *Azospirillum* along with FYM and conventional fertilizers helps in increasing the yields attributes of wheat crop as shown in Table (1). These findings support the finding of Chandrasekar et al. (2005) and Gomaa et al. (2012). *Azospirillum* strains are known to produce phytohormones like indole acetic acid, gibberellins and cytokinins under *in vitro* conditions (Venkateswarlu and Rao, 1983; Tien et al., 1979; Hartmann et al., 1994; Rademacher, 1994).

Maximum significant increase in crop growth rate (24.91 g m^{-2} day^{-1}), relative growth rate (0.073 g g^{-1} day^{-1}) and spike length (10.33 cm) were found with treatment T_4: *A. chroococcum* + 75% NPK + 25% FYM @ 24 t ha^{-1} as shown in Table 1 which are comparable to the findings of Tawfik et al. (2011), Kandil, A. A. (2004) and Malik et al. (2009). Bacterial production of nitrogen, phosphorus and indole-3-acetic acid from biofertilizers in the rhizosphere increases the growth of the crop (Rothballer *et al.*, 2005).

Number of tiller hill^{-1} (6.46) were significantly highest with treatment T_9: (PSB + *A. chroococcum* + *A. lipoferum* + 75% NPK + 25% FYM @ 24 t ha^{-1}) as shown in table 1. Lavakush et al (2014) illustrated this by more availability of NPK in the soil by cumulative effect of plant growth promoting rhizobacteria activities in the rhizosphere of the soil. This also supports the finding of Alam *et al.* (2008) which explained this due to the increased synthesis of hormones (IAA) and solubilization of phosphorus by tested PGPR strains. This is also relevant with the finding of Sasaki *et al.* (2010) which is also due to the ability of PSB to improve solubilization of fixed soil phosphorus easily made available to the crop.

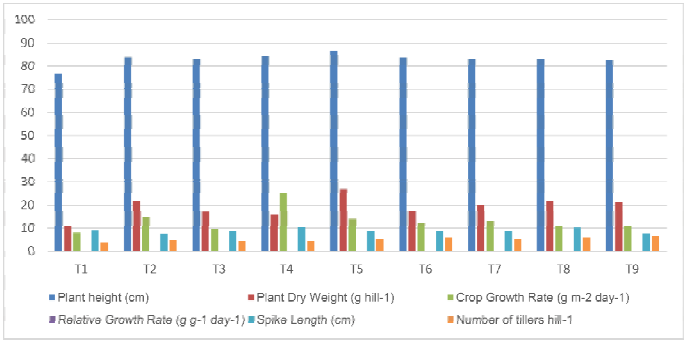

Figure 1: Effect of biofertilizers on growth parameters

Effect of biofertilizers on yield parameters

The data summarized in table 2 shows that the test weight (56.83 g), grain yield (6.25 t ha^{-1}) and harvest index (51.81 %) were significantly highest in treatment T_5 (*A. lipoferum* + 75 per cent NPK + 25 per cent FYM @ 24 t ha^{-1}) which support the findings of Bhaskara Rao and Charyulu (2005) and Ozturk et al. (2003b).This may be due to the increase in physiological activities of the crop and utilized maximum nutrient in the formation of grains.

Straw yield (6.17 t ha^{-1}) was found to be significantly higher in treatment T_6 (PSB + *A. chroococcum* + 75 per cent NPK + 25 per cent FYM @ 24 t ha^{-1}**)** which is relevant with the findings of Devi *et al.* (2011) and Malik *et al.* (2009). Phosphate-solubilizing micro-organisms (PSM) involve different character of micro-organisms which turn insoluble organic compound of phosphorus to soluble form (Raju and Reddy, 1999; Sundara *et al.*, 2002).

Table 1: Effect of biofertilizers on growth parameters

Treatments	Plant height (cm)	Plant dry weight (g hill^{-1})	Crop growth rate (g m^{-2} day^{-1})	Relative growth rate (g g^{-1} day^{-1})	Spike length (cm)	Number of tillers (hill^{-1})
	120 DAS	120 DAS	90 DAS	90 DAS	120 DAS	120 DAS
T_1	76.77c	11.08c	8.03c	0.058ab	9.04abc	4.06b
T_2	83.75ab	21.65ab	14.50b	0.056ab	7.70c	5.20ab
T_3	83.16b	17.25bc	9.70bc	0.040b	8.97abc	4.46b
T_4	84.18ab	15.93bc	24.91a	0.073a	10.33a	4.53b
T_5	86.73a	26.92a	13.97bc	0.050ab	8.75bc	5.26ab
T_6	83.60ab	17.55bc	11.84bc	0.041b	8.99abc	6.26a
T_7	82.78b	20.08ab	13.02bc	0.050ab	8.97abc	5.33ab
T_8	82.99b	21.73ab	11.14bc	0.035b	10.16ab	6.33a
T_9	82.45b	21.48ab	10.92bc	0.037b	7.90c	6.46a
F-test	S	S	S	S	S	S
SEd (±)	1.56	3.699	2.943	0.101	0.679	0.731
CD (P= 0.05%)	3.31	7.842	6.239	0.214	1.44	1.549

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Maximum significant numbers of grains spike-1 (42.20) were recorded in treatment T_8 (*A. chroococcum* + *Azospirillum* + 75 per cent NPK + 25 per cent FYM @ 24 t ha^{-1}). This is in confirmation with the findings of Abd El-Lattief, E. A. (2012) which might be due to the role of bio-fertilizer (*Azotobacter* and *Azospirillum*) in enhancing soil biological activity, which improved nutrient mobilization from organic and chemical sources. Also, the bio-fertilizer plays a significant role in regulating the dynamics of organic matter decomposition and the availability of plant nutrients and in increasing nitrogen fixer.

Number of effective tillers hill^{-1} (6.27) were found to be significantly highest in treatment T_9 (PSB + *A. chroococcum* + *A. lipoferum* + 75 per cent NPK + 25 per cent FYM @ 24 t ha^{-1}), which are relevant with the findings of Malik *et al.* (2009.), Devi *et al.* (2011) and Janaki Rama Suresh Kumar G. (2006) This was due to inoculation with plant growth promoting micro-organism, like *Azotobacter* and *Azospirillum* are attributed mainly to an improvement in root system development, an increase in water and mineral uptake by roots, plant hormone production and biological nitrogen fixation (Okou and Itzigsohn 1995).

Effect of biofertilizers on microbial population.

The highest significant of total bacterial population (24.33 × 10^{7}CFU g^{-1}) was observed in treatment T_5: *A. lipoferum* + 75 per cent NPK + 25 per cent farmyard manure @ 24 t ha^{-1}) as shown in Relevant result was obtained by Roy and Hore (2011). The increase might be due to greater availability of organic carbon and mineralized nutrients for their proliferation and development (Suresh and Surya Prabha, 2005).

Effect of biofertilizers on economics

The maximum benefit cost ratio (2.00) was recorded at T_5 (Azospirillum + 75 per cent NPK + 25 per cent farm yard manures @ 24 t ha^{-1}). This in conformation with the findings of Golada *et al.* (2012) who reported that the treatment with 10 t FYM + 100 kg N ha-1 + *Azospirillum* inoculation recorded significantly higher green forage yield, net return and BCR. This increase may be due to the increase in grain yield.

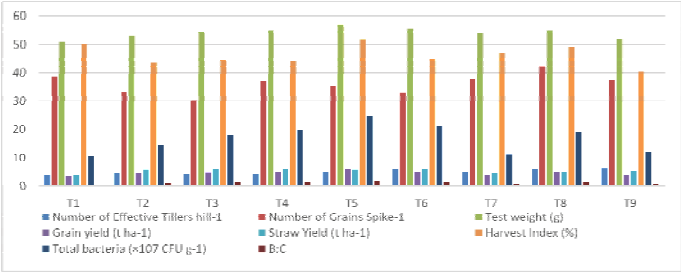

Figure 2: Effect of biofertilizers on Yield parameters, Economics and Microbial Population

CONCLUSION

It was concluded that the wheat crop treated with *Azospirillum lipoferum* + 75 per cent NPK + 25 per cent FYM @ 24 t ha^{-1} showed positive effects on growth, yield and economics of wheat and also showed maximum response to microbial population within the rhizosphere of the crop.

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Table 2: Effect of biofertilizers on yield parameters, economics and microbial population

Treatments	Number of effective tillers hill^{-1}	Number of grains spike^{-1}	Test weight (g)	Grain yield (t ha^{-1})	Straw yield (t ha^{-1})	Harvest index (%)	Total bacteria (×10^7 CFU g^{-1})	B:C
T_1	3.73c	38.8ab	50.97d	3.42c	3.58c	50.23	10.66e	0.04
T_2	4.53bc	33.27bc	53.13bcd	4.50bc	5.67ab	43.83	14.66d	1.19
T_3	4.07c	30.33c	54.47abc	4.67bc	5.92ab	44.73	18.00c	1.26
T_4	4.20c	37.13ab	54.93abc	4.83abc	6.08a	44.13	19.66bc	1.34
T_5	4.93abc	35.33bc	56.83a	6.25a	5.75ab	51.81	24.33a	2.00
T_6	5.80ab	33.07bc	55.50ab	5.07ab	6.17a	44.93	21.00b	1.45
T_7	5.13abc	37.80ab	54.23abc	4.00bc	4.50bc	46.74	11.33e	0.92
T_8	5.87ab	42.20a	54.96abc	4.83abc	5.08ab	49.11	19.00bc	1.31
T_9	6.27a	37.73ab	51.86cd	3.67bc	5.42ab	40.44	12.00e	0.79
SEd(±)	0.692	3.065	1.523	0.723	0.692	5.095	1.204	-
CD (P= 0.05%)	1.467	6.497	3.229	1.531	1.468	NS	2.553	-

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Microbial validation of organic preparations used in natural farming

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ABSTRACT

In this study, the scientific validation of different organic preparations like amritpani, panchagavya, vermiwash and Jeevamrita in terms of microbial population and their activity has been performed. Microbes isolated from these preparations were tested for plant growth promoting (PGP) properties and finally characterized by biochemical and molecular tools. Strains of *Bacillus pumilus* and other bacillus sp. isolated form panchagavya showed positive results for HCN and ammonia production while bacteria isolated from vermiwash (*Bacillus pumilus* and *Brevibacterium laterosporus*) showed positive results for HCN production and P-solubilization. Plant growth promoting strain *Brevibacillus borstelensis* (isolated from amritpani) showed positive results for all PGP properties (including HCN, siderophore, ammonia, catalase and IAA production) except

P-solubilisation. Based on these PGPR properties, such strains can be utilised for development of microbial consortia and other bio-formulations for sustainable agriculture production.

Key words: Panchagavya, Amritpani, Vermiwash, Jeevamrita, PGPRs, Rhizosphere

Rhizospheric soil of plants characterized by microbial activity and rhizospheric microbes have the potential to stimulate plant growth and manage soil and plant health. Nowadays management of the rhizosphere bacterial population has advanced toward the concept of plant growth promoting rhizobacteria (PGPR) owing to emergence of fungicide-resistant pathogens and health concerns for the growers and consumers. Using PGPR as bio-fertilizers is an efficient approach to replace agro-chemicals for sustainable production. In this connection use of organic liquid preparations is an age old practice in India (Hagedorn and Holt; 1954). Organic liquid preparations are obtained by active fermentation of animal and plant residues over specific duration. *Panchagavya, amritpani* and *jeevamrit* are rich source of microbial consortia, macro and micronutrients and plant growth promoting substances (Goud *et al*., 1985).They are used for seeds/seedlings treatment and rapid decomposition of organic wastes in composting process. *Panchagavya* is the most effective bio-enhancer demonstrated by many researchers (Pikovskaya *et al*., 1948). Availability of quality organic inputs viz. organic manures, bio-enhancers, bio-pesticides etc. for organic production is one of the major constrain. Therefore, on farm quality input production is cost effective aspect of organic production.

PGPR inoculants can facilitate beneficial interactions in plants leading to significant solutions for sustainable and environment-friendly agriculture. The applications of rhizospheric soil of agricultural crops with desirable bacterial populations have established considerable promises in both the laboratory and greenhouse experiment. These rhizospheric microbes have the potential to stimulate plant vigour (Fred *et al*., 1932). Plant growth promoting bacteria are linked with many plants and are commonly present in diverse environments. PGPRs can be classiûed as biofertilizers, phytostimulators and bio-pesticides with certain bacteria having overlapping applications. The most widely studied group of PGPB are plant growth-promoting rhizobacteria colonizing the root surfaces and the closely adhering soil interface (Jensen *et al*., 1954). *Rhizobacteria* have been analysed as promising replacements for chemical fertilizers due to the huge deterioration in terms of physicochemical properties of cultivated land. Using PGPR as biofertilizers is an efficient approach to replace chemical fertilizers and pesticides for sustainable cultivation (Norris and Chapman; 1968). Considering the multiple applications of rhizobacteria, it is essential to study their diversity, which could be useful in designing strategies to use these native strains as bio-inoculants for sustainable and organic agriculture without causing harm to the environment and farmers. With the currently available tools, the microbial community structure can be examined at several levels. The simplest analysis is based on DNA profiles, generated by PCR followed by restriction digestion of PCR product, to identify differences in the community composition. Furthermore, sequencing (16S rRNA gene) provides greater discrimination over functional diversity studies and better

characterization of an isolate. The objective of the present study was to isolate and characterise PGPRs and their community structure associated with *Amritpani, Panchagavya, Vermiwash, and Jeevamrita* by *in vitro* assays and gene amplification techniques.

MATERIAL AND METHODS

Organic preparations were made using locally available materials. *Panchagavya* was prepared by mixing of five products of cow, *i.e.* dung (5 kg), urine (3 l), milk (2 l), curd (2 l) and ghee (1 l). To this, sugar cane juice (3 l), jaggery (0.5 kg), tender coconut water (3 l), toddy (coconut wine) (2 l) and ripe bananas (12) were added in a wide mouthed plastic container. Another preparation, *Amritpani* was prepared by incubating 10 kg cow dung along with 250 g cow ghee, 500g honey and 150 liters of water in a plastic container (Okon *et al*., 1977) while *Jeevamrita* was prepared by fermenting 10 kg cow dung, 5 liters urine, 2 kg jaggery, 2 kg pulse flour and 250 g virgin soil and 150 liters water in a plastic container. Vermi wash is a liquid leachate obtained by excess water to saturate the vermi composting substrates. It was prepared in plastic drum of 200 liters capacity provided with tap in the bottom placed in shade. Five cm each of concrete and red sand was laid in the bottom of the drum for effective drainage. Soften kitchen wastes (30-40cm) or one-week-old dung was filled in the drum. 200-300 earth worms were released in the waste/dung. An earthen pot with minute hole in the bottom from where water pours drop wise was hanged over the drum after one week of worm's inoculation. After 2-3 days, extract collected in earthen pot from the tap placed at the bottom of drum is called as Vermiwash (Bakker & Schippers; 1987).

Amritpani, panchagavya, vermiwash and jeevamrita are enriched with diversity of microorganisms and its evident by analyzing total microbial count (CFU ml^{-1}) by using serial dilution or pour plate techniques in different media types like: nutrient agar (specific for bacteria), rose bengal agar (specific for fungi), actinomycetes agar (specific for actinomycetes), kings B agar (specific for pseudomonas), crystal violet agar (specific for G^+ bacteria), methyl red agar (specific for G^- bacteria), pikovaskya agar (specific for P-solubilizing micro-organisms), CRYEMA (specific for rhizobium), N-free malate medium (specific for Azospirillum) and jensen agar medium (specific for Azotobacter).

Enumeration of different beneficial microbial populations viz. bacteria, fungi, actinomycetes, *Pseudomonas*, gram positive bacteria, gram negative bacteria, p-solubilizing bacteria, *Rhizobium, Azotobacter and Azospirillum* were carried out by using dilution plate count method using selective media *viz*. nutrient agar, rose bengal chloramphenicol agar (RBCA), actinomycetes isolation agar, King's B, methyl red agar. crystal violate agar, pikovskaya's agar, Yeast extract mannitol agar with congo red (CRYEMA) modified Jenson's agar and N-free malate medium respectively. Petri dishes were made by pouring each specific solid medium. Then 10 ml of each bio-enhancers sample were diluted to 90 ml sterile water and that was considered being 10^{-1} dilution factor. Transferring of 1 ml of 10^{-1} dilution to 9 ml sterilized water with the help of a sterilized pipettes yielded 10^{-2} dilution. In this way, a series of up to 10^{-8} dilutions were prepared under aseptic condition. 0.1ml of the suspension from required dilution (e.g. 10^{-8}) was taken and poured into the respective agar media on petri dish and spread with L-spreader with the help of Plate Master (Hi-Media). Then plates were incubated at 28 ± 2°C for 3-5 days. The numbers of visible colonies were counted and total count was obtained by multiplying number of visible colonies in the plate by the dilution factor.

All the isolates were morphologically and phenotypically characterized on the basis of gram's staining, motility test and utilization of 10 different substrates (all from Hi-Media). The reactions tested were HCN production (Ahmad *et al*., 2012), Siderophore production (Israr *et al*., 2014), NH_3 production using peptone water, PO_4-solubilization (Ram *et al*., 2018; Ram *et al*., 2019), IAA production (Ram *et al*., 2019), catalase test *etc*.

The cultures were maintained on nutrient agar (NA, Hi Media) at 25^0C. For DNA extraction pure culture of 16 isolates were grown in liquid culture of nutrient broth (Hi Media) at 25^oC for 24 to 48 hours. Pellets were collected by centrifugation at 13,000 rpm. Total genomic DNA was extracted by using the Hi Media kit. Pure DNA was dissolve in 50µl elution buffer. Purity of DNA was quantified by UV spectrophotometry and ethidium bromide fluorescence. Primer PA (5'AGAGTTTGATCCTGGCTCAG3') and PH ($^{5'}$AAGGAGGTGATCCAG CCGCA$^{3'}$) were used to amplify a fragment of 16s rDNA (Ross *et al*., 2000). PCR amplification were performed in a total volume of 100 µl by mixing 100 ng of genomic DNA, 0.5 µm conc. of each primer, 2.5 mM dNTPs and 2 unit of *Taq* DNA polymerase in 1x PCR buffer. The reaction were subjected to initial denaturation of 94^0C for 5.0 min followed by 35 cycle of 94^0C for 40 sec., 48^0 C for 30 sec, 72^0C for 1.0 min. with a final extension of 72^0 C for 5 min. in a Biored thermal cycler. PCR amplified products were analysed by running in 1.2% Agarose gel prepared in 1xTBE buffer and containing 0.5 µg ethidium bromide and photographed over a transilluminator. 1.5 kb PCR products were sent to commercial gene sequencing laboratory of Xcelrise, India for sequencing. Sequence analysis of these isolates was performed using BLAST (blastn) 2.0 search tool

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(http://www.ncbi.nlm.nih.gov) available on the NCBI homepage. The nucleotide sequences of 16s rRNA were deposited in Gene Bank. The accession numbers of the 16S rRNA nucleotide sequences of some of the isolates are presented table 2.

RESULT AND DISCUSSION

Microbial population dynamics in *panchagavya*

Bacterial population on nutrient agar media gradually increased during the fermentation period and highest number was recorded at 21st days after preparation (12.5 x 10^8cfu ml^{-1}) (table 2). Population of fungi was initially increased up to 4th days after preparation (4.0 x 10^5cfu ml^{-1}), but then gradually decreased at maturity stage. Interestingly, actinomycetes population was increased (8.0 x 10^6 cfu ml^{-1}) on 20th day (table 2). Gram positive bacterial population decreased towards maturity stage in *Panchagavya*. Gram negative bacteria increased over time and reached at a very high level (35.8 x 10^6 cfu ml^{-1}) on 20th day. Similar phenomenon was also observed with *Pseudomonas*. Population of *Rhizobium* was almost stable at the initial period but, slightly increased and highest was recorded at 3rd day (67.4 x 10^5 cfu ml^{-1}). The numbers of p-solubilizing microbes were also increased gradually from initial number of 21.3 x 10^5 (25 day). Two bacterial strains were identified as *Bacillus sp*. and *B. Pumilus* with the accession number KY523253 and KY523254 respectively in *Panchagavya* (table 1).

The maximum number of all type of PGP isolates existed in *Panchagavya* formulation due to presence of cow dung (Okon *et al*., 1977) (fig 1). Cow dung is an active ingredient of *Panchagavya* and is a rich source of beneficial microbes as reported earlier by many workers (Sangeetha and Thevanatha; 2010). Cow milk also contains beneficial microbes (Crielly *et al*., 1994). Cow curd is a rich source of *Lactobacillus* spp. The highest number of bacteria in *Panchagavya* may be due to nutrient richness of the mixture obtained from ingredients of cow origin viz. cow milk, curd and milk *etc.* Amalraj *et al*., (2013) had reported highest population of total bacteria (22 x 10^9 cfu ml^{-1}), actinomycetes (60 x 10^4 cfu ml^{-1}), p-solubilizers (103 x 10^6 cfu ml^{-1}), fluorescent pseudomonas (151 x 10^5 cfu ml^{-1}). Chadha *et al*., (2012) also reported highest load of viable bacterial populations, *Azotobacter* spp., actinomycetes as well as p-solubilizers in *Panchagavya*. Microbial and biochemical analysis of *Panchagavya* were also worked out by Radha and Rao, (2014).

Microbial population dynamics in *Amritpani*

Bacterial population increased in the preparation from an initial value of 24 x 10^7 cfu ml^{-1} at 0 day but then gradually decreased on nutrient agar medium similarly fungi population showed maximum value (1.2 x 10^5) at 0 DAP (table 2). Actinomycetes population was decreased initial days but increased maximum at 25th day, showed 85.4 x 10^7 CFU ml^{-1}. Population of gram positive bacteria was high (17 x10^7) at 3 DAP but decreased with further time intervals. Gram negative bacteria had increased at 9th day. Interestingly, the *Pseudomonas* population was gradually increased from an initial value of 48 x 10^6 cfu ml^{-1} at 14th day. The populations of *azotobacter* and *azospirillum* showed maximum count at 0 DAP (table 2). Based on PGP properties 2 bacterial strains were identified as *B. subtilis* (KY523245) and uncultured bacterium (KY523248) table 1.

Microbial population dynamics in *Vermiwash*

Vermiwash enriched with diversity of microorganisms and it's evident by analyzing total microbial count (CFU ml^{-1}) by using spread plate technique in different specific medium. The maximum number of bacterial count (4 x 10^7 CFU ml^{-1} and 36.5 x 10^6) observed on nutrient agar and crystal agar medium respectively after 15 DAP (table 2). In vermiwash the considerable number of all PGP bacteria, fungi, azotobacter and actinomycetes were observed (fig 1). Finally from vermiwash 2 bacterial strains were identified as *Bacillus pumilus, Brevibacterium laterosporus,* and *Brevibacterium laterosporus* with accession no. KY523251, KY523246 and KY523247, respectively (table 1).

Table 1: Identification of bacteria based on maximum similarity with 16sRNA sequences and PGP properties

Organic preparation	Strain	NCBI Accession no.	Isolate code	HCN production	Siderophore production	NH_3 production	P solubilization	IAA Production	Catalase
Amritpani	*Bacillus subtilis*	KY523245	CISH-PSB10	+	-	-	-	+	+
	Uncultured bacterium	KY523248	CISH-PSB13	++	-	-	-	+	-
Jeevamrita	*Brevibacillus borstelensis*	KY523252	CISH-PGPR67	++	+	+	-	+	+
Panchgavya	*Bacillus sp.*	KY523253	CISH-PGPR92	+	-	+	-	-	-
	B. pumilus	KY523254	CISH-PGPR96	+	-	+	+	-	-
Vermiwash	*Brevibacillus laterosporus*	KY523246	CISH-PSB11	+	-	+	-	-	-
	B. pumilus	KY523251	CISH-PSB23	+	-	+	-	-	-

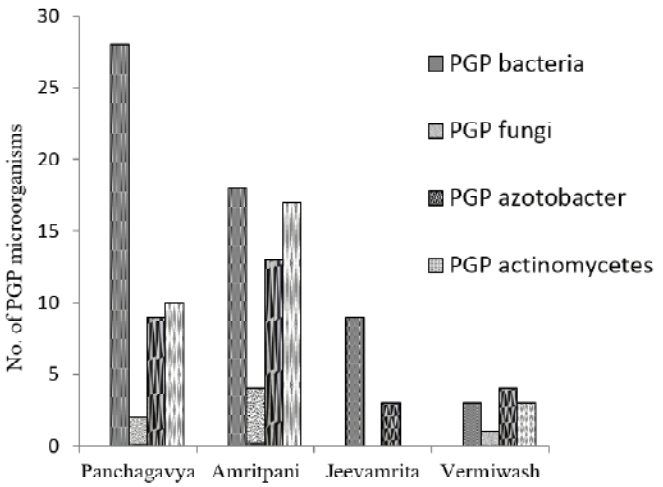

Figure 1: Number of plant growth promoting (PGP) microorganisms in different organic preparations

Microbial population dynamics in *Jeevamrita*

The highest bacterial population was observed on 6 DAP i.e. 28.8 x 10^8 CFU ml^{-1} (table 2). The maximum PGP *rhizobium* (35 x 10^8) was observed maximum in jeevamrita as compared to other organic preparations. Maximum population of bacteria was recorded 288 x 10^7 cfu ml^{-1} at 6^{th} day on nutrient agar medium, similarly fungi population showed maximum value (44.2 x10^6) at 14^{th} DAP (table 2). Actinomycetes population was decreased during initial days but increased maximum at 25^{th} day, showed 65 x 10^6 CFU ml^{-1}. Population of gram positive bacteria was high (47 x10^7) at after 25 DAP but decreased with further time intervals. Gram negative bacteria had increased at 9^{th} day. Interestingly, the *Pseudomonas* population was gradually increased up to 12.6 x 10^7 cfu ml^{-1} at 3^{rd} day. The populations of *azotobacter* and *azospirillum* showed maximum count at 0 DAP (table 2). Based on PGP properties one bacterial strain, *Brevibacillus borstelensis* (KY523252) identified (table 1).

Based on the microbial populations in different organic preparations (table 2), selected microorganisms were tested for PGP properties in different media types. Results showed that *panchagavya* enriched with maximum number of PGP bacteria while *amritpani* proved to be best in terms of number of PGP fungi, azotobacter and actinomycetes among all organic preparations (fig. 1). The search for PGPRs and their mode of action are increasing at a rapid rate in order to use the best PGPR strain as commercial biofertilizer. Investigations into the mechanisms of plant growth promotion by PGPR strains indicated that the effective PGPRs increased plant growth basically by changing the whole microbial community structure in rhizosphere. The general mechanisms of plant growth promotion by PGPR includes associative nitrogen fixation, lowering of ethylene levels, production of siderophore and phytohormones, induction of pathogen resistance, solubilization of nutrients, promotion of mycorrhizal functioning, decreasing pollutant toxicity (Van Loon, L.C 2007; Castro *et al*., 2009) etc. PGPR can significantly changes the root architecture and stimulate plant development with the production of different phytohormones. PGPR appeared as potential rhizobacteria in getting established in soil ecosystem due to their high adaptability in a diverse environments, faster growth rate and metabolic versatility for a wide range of natural and xenobiotic compounds. PGPR can promote growth and yield of crop plants by direct and indirect mechanisms. Potential role of PGPRs in conferring resistance to water stress in different crops has been investigated (Mayak *et al*., 2004; Jensen, H.L. 1954). Fluorescent *Pseudomonas* and *Bacillus* species were reported with very high efficiency in host root colonization and production of growth metabolites resulting in improved strategic crop yield (Khalid *et al*., 2004). The various modes of action of *Bacillus subtilis* against phytopathogens, suggested the role of the bacterium in plant vitality.

Mechanisms of plant growth promotion by PGPR strains indicated that the effective PGPRs increased plant growth basically by changing the whole microbial community structure in rhizosphere. PGPR can alter root architecture and promote plant development with the production of different phytohormones. PGPR seemed as successful rhizobacteria in getting established in soil ecosystem due to their high adaptability in a wide variety of environments, faster growth rate and biochemical versatility to metabolize a wide range of natural and xenobiotic compounds. PGPR can promote growth and yield of crop plants by direct and indirect mechanisms. The various modes of action of a *Bacillus subtilis* against phytopathogens, suggests the role of the bacterium in plant health management. The bacteria isolated from *Amritpani, Panchagavya, Vermiwash, and Jeevamrita* were tested for their biochemical characters and molecular confirmation using molecular tools. The substrate utilization pattern showed that the characteristics of the majority of the tested strains share a similar pattern. However, few isolates displayed varied patterns. Identification of these isolates showed that most of them belong to genus *Bacillus* and *Alcaligenes* spp. Sequence analysis of these isolates was performed using BLAST (blastn) 2.0 search tools (http://www.ncbi.nlm.nih.gov) available on the NCBI homepage. The nucleotide sequences of 16S rRNA were deposited in Gene Bank (table 1).

Table 2: Microbial population in different organic preparations at different time intervals

Microbial groups	Amritpani (cfu ml^{-1})	Jeevamrita (cfu ml^{-1})	Panchagavya (cfu ml^{-1})	Vermiwash (cfu ml^{-1})
Bacteria (on Nutrient agar)	24x10^{7} (0 DAP)	288 x10^{7} (6 DAP)	125x10^{7} (21 DAP)	4x10^{7} (15 DAP)
Fungi	1.2x10^{5} (0 DAP)	44.2 x10^{6} (14 DAP)	4 x 10^{5} (4 DAP)	nil
Actinomycetes	85.4 x 10^{7} (> 25 DAP)	65 x10^{6} (>25 DAP)	8 x10^{6} (20 DAP)	10.4 x10^{5} (15 DAP)
G^{+} bacteria	17x10^{7} (3 DAP)	47 x 10^{7} (>25 DAP)	12 x 10^{7}(25 DAP)	36.5 x 10^{6} (15 DAP)
G^{-} bacteria	50 x10^{7} (9 DAP)	11.9 x10^{7} (6 DAP)	35.8 x10^{6} (20DAP)	12 x10^{5} (15 DAP)
Pseudomonas	48x10^{6} (14 DAP)	12.6 x 10^{7} (3 DAP)	57 x10^{6} (20 DAP)	10 x10^{4} (15 DAP)
Rhizobium	16.4 x10^{5} (20 DAP)	35 x10^{8} (3 DAP)	67.4 x10^{5} (3 DAP)	7 x 10^{3} (15 DAP)
P-solubilizing	68x 10^{7} (9 DAP)	78 x 10^{5} (3 DAP)	21.3 x 10^{5} (25 DAP)	60 x10^{3} (15 DAP)
Azotobacter	29.7 x10^{6} (0 DAP)	51 x 10^{5} (>25 DAP)	45 x 10^{5} (0 DAP)	14 x 10^{4} (15 DAP)
Azospirillum	2.01 x10^{6} (0 DAP)	9 x10^{5} (0 DAP)	16 x10^{4} (20 DAP)	7 x 10^{3} (15 DAP)

*DAP-Days after Preparation

Phosphorus is one of the essential nutrients for plant growth and development. Ironically, soils may have large reservoir of total phosphorous (P) but the amounts available to plants are usually a tiny proportion of this total. Several phosphate solubilizing micro-organisms (PSMs) are now recorded to convert the insoluble form of phosphorus to soluble form through acidification, secretion of organic acids or protons (Richardson *et al*., 2009) and chelation and exchange reactions (Hameeda *et al*., 2008). The ability of PGPRs to solubilize mineral phosphate, therefore, has been of immense interest to agricultural microbiologists since it can enhance the availability of phosphorus for effective plant growth. PGPRs have been recorded to solubilize precipitated phosphates to plants, representing a possible mechanism of plant growth promotion under field conditions (Verma *et al*., 2001).The present study has resulted in the isolation of PGPRs *Bacillus subtilis, Brevibacillus laterosporus, Bacillus pumilus, Brevibacillus borstelensis,* that can be utilised in microbial consortia preparation and bio-formulation for organic production of various crops.

CONCLUSION

The study concludes that PGPB are potential source for sustainable agricultural production because they improve plant growth and yield under adverse climatic conditions. Identified PGPB strains can directly facilitate the plant vigour by production of phytohormones like indole-3-acetic acid (IAA) produced by test strains. These strains also showed phosphorus solubilization, siderophores and HCN production. All the bacterial strains identified in this study can be used efficiently to combat abiotic stress and synergistically play role in natural farming practices.

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Performance of chrysanthemum at dark exposure under polytunnel condition

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ABSTRACT

The field experiment carried out in chrysanthemum (cv. Chandrika) nursery 10 at College of Horticulture, Dapoli, Dist. Ratnagiri 2020-21 under polytunnel conditions during 2020-21 revealed that exposing chrysanthemum plants to darkness for 180 minutes using 90 per cent black shed net, showed promising results in increasing plant spread, number of branches, average leaf area, leaf area index, days required for 50 per cent flowering, flower diameter, length of flower stalk and the flower yield parameters, respectively. The shelf life of chrysanthemum flowers was extended by 24.84 days at 2°C temperature level.

Keywords: Chrysanthemum, levels of light interruption, Chandrika, shelf life, temperature levels.

Chrysanthemums, called mums in short in the U.S.A. or chrysanths, and commonly known as 'Glory of the east', are flowering plants of the genus Chrysanthemum in the family Asteraceae (Compositae). The genus includes over 200 species of annuals, herbaceous perennials and the sub shrubs. It is widely distributed over Asia, Africa and Europe. Many authorities claim that it has been originated from China. (Cumming, 1964). They are native to East Asia and north eastern Europe, America and Africa; Vernacular names of chrysanthemum are Guldaudi, Guldawri (in Hindi), Chandramallika (Bengali), Bagauri (Punjabi), Guldaudi (Gujarati), Gulsevati, shevanti (Marathi), Gavanthi (Tamil), Savantige (Kannada), Chamanti (Telugu). Most species originate from East Asia and the center of diversity is in China. Countless horticultural varieties and cultivars exist. There is hardly any other garden flower which has such diverse and beautiful range of colour shades, widely different flower shapes, and height range as chrysanthemum.

Chrysanthemum is not cultivated on commercial platform in Konkan region of Maharashtra till today. There is a huge demand of these flowers throughout the year in metro cities like Mumbai, Thane and even in developing towns in Ratnagiri, Raigad and Sindhudurg district due to rise in tourism industry. Accordingly, an effort to manipulate reproductive growth and to have at least three cycles of crop in a year by interrupting light for early flower induction using 90 per cent black shade net was made. This will ultimately help in fulfilling the demand of end users by providing flowers throughout the year and making chrysanthemum a lucrative crop for this region

MATERIAL AND METHODS

The field experiment was conducted using chrysanthemum (cv. Chandrika) in nursery at College of Horticulture, Dr. Balasaheb Sawant Konkan Krishi Vidyapeeth, Dapoli, Dist. Ratnagiri (M.S.) India during *rabi* season from 24th October, 2020 to 24th February, 2021. The selected polytunnel was uniformly levelled. The soil was red lateritic with uniform sub plot (1.25m×1m) having good drainage and pH 6.0. The experiment was laid out in Randomized Block Design with nine treatments replicated thrice. The treatments comprised of interruption of light/ dark exposure for 30 minutes (T_1), 60 minutes (T_2), 90 minutes (T_3), 120 minutes (T_4), 150 minutes (T_5), 180 minutes (T_6), 210 minutes (T_7), 240 minutes (T_8) and the control (T_9). The laboratory experiment was conducted in Control Randomized Design (CRD) at Post Harvest Technology Laboratory. The shelf life of chrysanthemum flowers (cv. Chandrika) was studied at three temperature levels viz., 2°C (T_1), 4°C (T_2), 12°C (T_3) and control at room temperature (T_4). Statistical analysis was carried out by standard method of analysis of variance described by Panse and Sukhatme (1995).

RESULT AND DISCUSSION

Vegetative parameters

Data presented in table 1 showed that the average plant spread was significant between various treatments with different duration of dark exposure except at 30 minutes of exposure. Maximum average plant spread (38.64 cm) was noticed at 180 minutes (T_6), which was significantly superior over other treatments. The minimum plant spread (30.26 cm)

was observed at 120 minutes in the control (T_9). Increase in plant spread might be due to production of a greater number of branches and by the genetic nature of the plant. Variation in plant spread may also be due to additive gene effect. (Viidalie *et al.,* 1992). It might also be due to bigger size leaves of the cultivar.

The treatment (T6) also recorded the highest number of primary (9.47), secondary (18.73), and tertiary branches per plant (23.95), average leaf area (14.41 cm^2) and the leaf area index (5.22) as against of lowest 7.30, 10.27, 15.15, 7.09 and 2.74, respectively) in T_9 (control). Year-round production can be achieved in greenhouses by controlling the climate conditions. During winter, high energy inputs are required to maintain good plant quality and production levels (Van Der Ploeg and Heuvelink, 2006). The vegetative vigour of the plant was enhanced in treatment T_6 i.e., 180 minutes interruption of light. This may be contributed to photoperiod as well as optimum carbon dioxide fixation. Which ultimately resulted in maximum average spread of plant.

Greater leaf area may lead to more dry matter accumulation, which resulted in the accumulation of maximum photosynthates that contributed to produce bigger sized flower or a greater number of flowers. Similar variation in leaf area among cultivars was found in carnation (Gharge *et al.,* 2009; Shiragur *et al.,* 2004).

Flowering parameters

Data on various flowering parameters are summarized in table 2. This showed that T6 (180 minutes) recorded the lowest average days (103.9) for 50 per cent flowering and maximum flower diameter (4.59 cm), length of flower stalk (9.13 cm) and fresh weight of 100 flowers (193.6 g) as against 132.20 days, 3.8 cm, 6.72 cm and 120.15 g, respectively recorded in control (T_9). The variation in the days taken for 50% flowering among the different levels of light interruption may be due to genetic make-up of particular plant, light and genotype environment interactions leading to varied rates (Kumar *et al.,* 2018). Change in photoperiod levels for early commencement of days for 50% flowering in chrysanthemum have been noted by Dhiman *et al.,* (2017). Generally being ethylene non-sensitive flower the difference in days taken to flower senescence may be due to varietal characteristics of chrysanthemum cultivar. Similar findings were found in rose (Tabassum *et al.,* 2002) and in gerbera (Nair and Mehedi, 2004).

The findings on flower diameter are similar to those reported by Siddiqua *et al.,* (2018) that greater leaf area may lead to more dry matter accumulation, which resulted in the accumulation of more photosynthates that contributed in producing higher number of big sized flowers.

Yield parameters

The average data on various yield parameter viz., fresh weight of flowers in a plant, fresh and dry weight of single flower are given in Table B. The treatment T_6 (180 minutes) in this case also was significantly superior recording the maximum average fresh flower weight per plant (81.92 g), fresh weight of single flower (1.93 g) and dry weight of single flower (0.71 g) against the minimum of 39.46 g, 1.20 g and 0.28 g recorded in control (T_9). Jiang *et al.,* (2010) reported improvement in flower quality with increase in photoperiod.

Shelf life of chrysanthemum flowers at different temperature

A maximum shelf life of 24.84 days was recorded at (T_1) 2°C temp. This was statistically at par at (14.78 days) at 4°C temp. (T_2) and 8.67 days at 12°C temp (T_3). Minimum shelf life (3.51 days) was recorded at room temperature control.

Table 1: Effect of different duration of dark exposure on vegetative parameters in Chrysanthemum cv. Chandrika

Duration of Treatments (Light Interruption)		Average plant spread (cm)	Average no. of branches			Average leaf area (cm^2)	Leaf area index
			Primary	Secondary	Tertiary		
T_1	30 minutes	30.90	7.70	11.57	16.01	7.90	2.97
T_2	60 minutes	31.64	8.00	12.37	16.17	9.04	3.37
T_3	90 minutes	33.46	8.47	13.70	18.60	9.77	3.69
T_4	120 minutes	34.68	8.97	17.30	20.47	10.28	4.31
T_5	150 minutes	36.70	9.20	17.93	23.12	11.32	4.83
T_6	180 minutes	38.64	9.47	18.73	23.95	14.41	5.22
T_7	210 minutes	33.98	8.70	15.83	19.93	9.87	3.91
T_8	240 minutes	32.66	8.03	13.07	17.71	9.25	3.59
T_9	Control	30.26	7.30	10.27	15.15	7.09	2.74
	Range	30.26-38.64	7.30-9.47	10.27-18.73	15.15-23.95	7.09-14.41	2.74- 5.22
	Mean	33.65	8.42	14.49	19.01	9.88	3.84
	S. Em. ±	0.32	0.13	0.32	0.47	0.52	0.16
	C. D. at 5%	0.97	0.40	0.96	1.40	1.55	0.47

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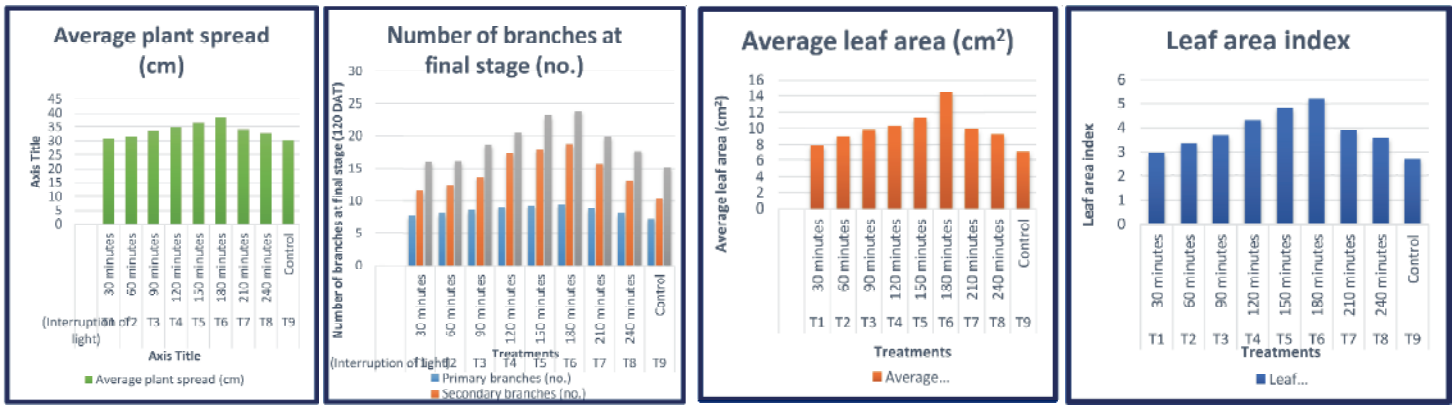
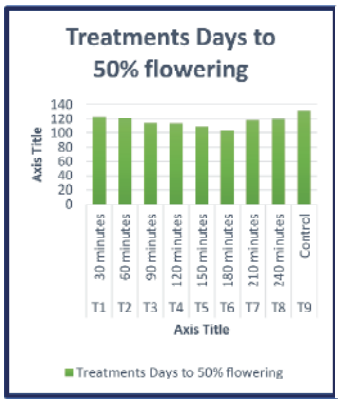
Table 2: Effect of different duration of dark exposure on flowering parameters in chrysanthemum cv. Chandrika

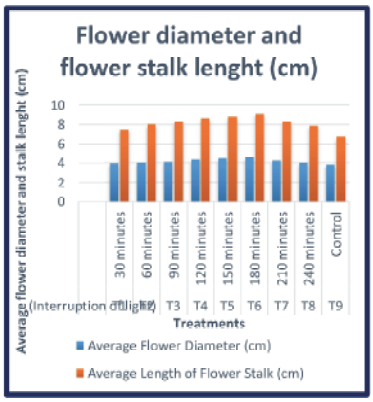
	Duration of Treatments (Light interruption)	Days to 50 per cent flowering	Flower diameter (cm)	Average flower stalk length (cm)	Fresh weight of 100 flowers (g)
T_1	30 minutes	123.93	3.92	7.50	123.10
T_2	60 minutes	120.83	4.05	8.02	133.99
T_3	90 minutes	114.60	4.16	8.31	145.31
T_4	120 minutes	113.87	4.35	8.58	165.73
T_5	150 minutes	109.00	4.46	8.76	188.92
T_6	180 minutes	103.90	4.59	9.13	193.60
T_7	210 minutes	117.87	4.26	8.30	157.59
T_8	240 minutes	119.13	4.03	7.87	124.27
T_9	Control	132.20	3.8	6.72	120.15
	Range	103.90-132.20	3.80-4.59	6.72-9.13	120.15-193.60
	Mean	117.25	4.18	8.13	150.29
	S. Em. ±	1.89	0.06	0.15	14.57
	C.D. at 5%	5.68	0.18	0.46	43.68

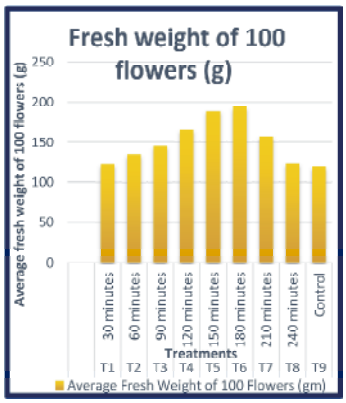
Table 3: Effect of different duration of dark exposure on yield parameters in chrysanthemum cv. Chandrika

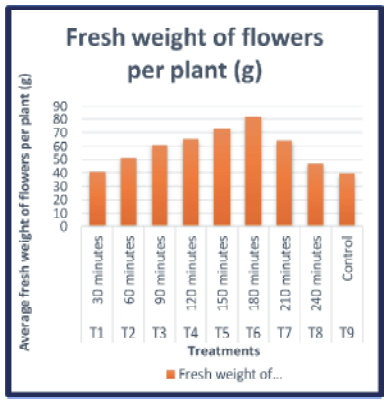
	Duration of treatments (Light Interruption)	Fresh weight of flowers		Dry weight of single flower (g)
		Plant^{-1} (g)	Single flower (g)	
T_1	30 minutes	40.88	1.22	0.37
T_2	60 minutes	51.40	1.33	0.39
T_3	90 minutes	61.27	1.45	0.44
T_4	120 minutes	65.61	1.66	0.54
T_5	150 minutes	73.34	1.88	0.64
T_6	180 minutes	81.92	1.93	0.71
T_7	210 minutes	64.26	1.57	0.49
T_8	240 minutes	47.04	1.24	0.38
T_9	Control	39.46	1.20	0.28
	Range	39.46- 81.92	1.20- 1.93	0.28- 0.71
	Mean	58.35	1.49	0.47
	S. Em. ±	1.97	0.01	0.01
	C. D. at 5%	5.91	0.03	0.03

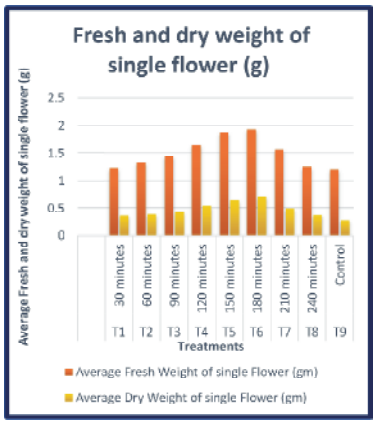


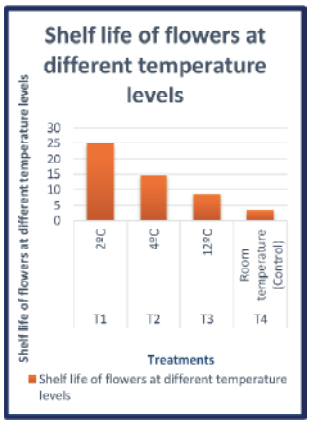












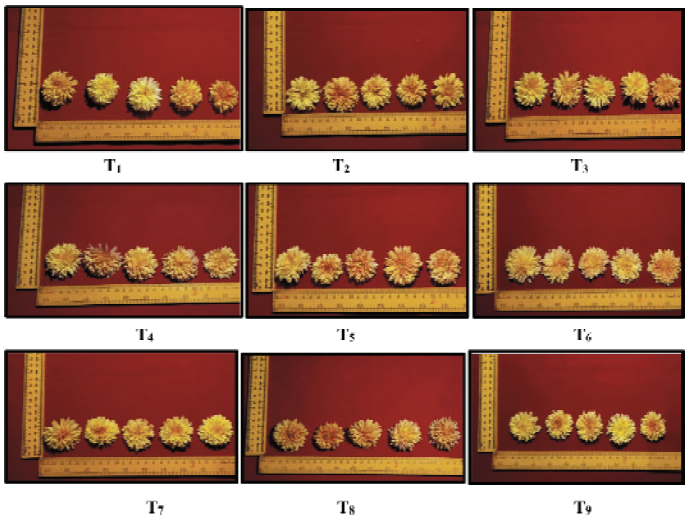


Plate I : Flower diameter (cm) in different treatments of light interruption/dark exposure

Table 4: Effect of temperature levels on shelf life of chrysanthemum flowers

	Temperature levels (°C)	Shelf life of flowers (days)
T_1	2°C	24.84
T_2	4°C	14.78
T_3	12°C	8.66
T_4	Room temperature (Control)	3.30
	Range	3.30-24.84
	Mean	12.90
	S. Em. ±	6.09
	C.D. at 5%	18.35

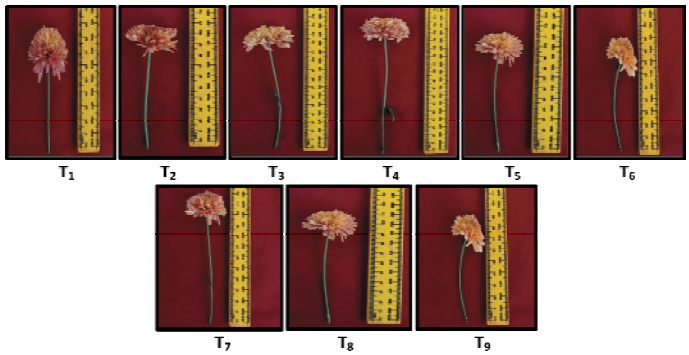


Plate II : Effect of different duration of dark exposure on stalk length in Chrysanthemum cv. Chandrika

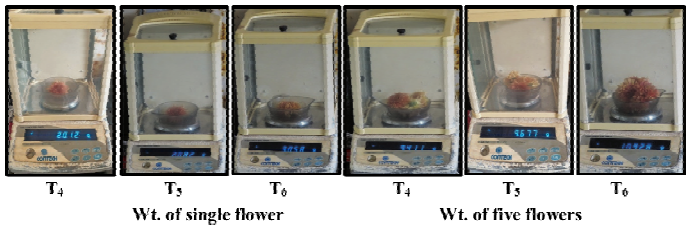


Plate III : Effect of different temperature levels on weight of flowers in Chrysanthemum cv. Chandrika

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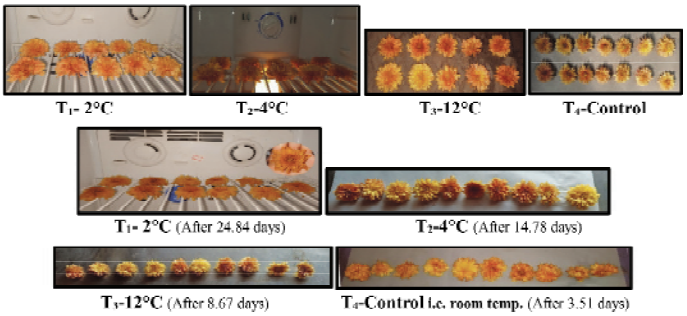

Plate IV: Effect of different temperature levels on shelf life of flowers of Chrysanthemum cv. Chandrika

Variation in shelf life may be attributed to differential accumulation of carbohydrates from varied leaf production, sensitivity of cultivars to ethylene, genetical framework and environmental factors of the plant (Dewan *et al.*, 2016). While evaluating the data on senescent flowers of chrysanthemum, it was observed that for evaluation at 3-, 8-, 12- and 25-days flowers that remained in temperature of 2°C, the process of development was slowly compared to those stored at 4 and 12°C. The stems to come out of cold storage had a rate of 5 to 7 per cent of senescing flowers, but to those stored for 12 days at room temperature, the index remained below 50 per cent at 2°C, while index in those stored at 4 and 8°C remained at 49 and 56 per cent, respectively (Thakur *et al.*, 2018).

These results are explained by the retardation of physiological processes (Taiz and Zeiger, 2004). Brackmann *et al.*, (2000) noted that the percentage of senescent flowers in chrysanthemum 'Red refocus' was lower in stems stored at low temperature. Metabolic activity observed in flowers during the period demonstrated that the sensitivity grows at low temperatures, which requires the use of temperatures less than 5.0°C during storage. These results are comparable with the data reported by Vieira and Souza (2009), who observed greater symptoms of senescence in chrysanthemum stored at 1.5°C. The above investigation indicate that shelf life of chrysanthemum flowers was maximum at low temperature i.e., 2°C which may have contributed to low generation of ethylene causing slow senescence of flowers and ultimately prolonging that's shelf life.

CONCLUSION

The maximum average plant spread, number of branches, average leaf area, leaf area index, days required for 50 per cent flowering, flower diameter, length of flower stalk, fresh weight of 100 flowers, fresh weight of flowers per plant, fresh weight of single flower and dry weight of single flower were recorded in plants treated with interruption of light for 180 minutes (T_6). The maximum shelf life of 24.84 days was noticed in treatment T_1 (2°C).

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Effects of pruning levels and growth regulators on Jasmine (*Jasminum sambac* L.) under Konkan condition

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ABSTRACT

An investigation carried out to study the effect of two levels of prunings (40 and 60 cm above ground) and two concentrations each of the growth regulators, the gibberellic acid (at 50 & 100 ppm) and cycoal (at 750 and 1000 ppm) on Jasmine (*Jasminum sambac* L.) under Konkan conditions revealed that pruning done at 60 cm above ground coupled with application of gibberellic acid at 100 ppm resulted in significant increase in the plant height, number of primary and secondary branches, girth of primary branch at base, number of leaves, average plant spread, leaf area, flower, bud length and width, number of flower buds plant^{-1}, number of days required for flower initiation, duration of flowering and flowers, yield plant^{-1} and kg ha^{-1} of jasmine flowers under Konkan conditions of Maharashtra.

Keywords: Jasmine, pruning, growth regulators.

The term jasmine is derived from an Arabic word Jessamine (Bailey, 1947) and also an Old Persian name Yasmyn meaning fragrance. In English, it is known as Arabian jasmine whereas in Marathi it is known as Mogra, which is fragrant evergreen shrub. Arabian jasmine (*Jasminum sambac* L.) is a tropical and subtropical flowering plant belonging to family Oleaceae. The genus has 13 basic chromosome and most of the species under this are diploid (2n=26). It is native to south East Asia.

In India about 40 species are distributed in Sikkim, West Bengal, Assam, Khasia and Jaintia hills, tropical North West Himalayas, Kashmir, Deccan peninsula, from Konkan to Travancore, Malabar Coast, Western Ghat, Niligiris, Palani hills, Coonor and South Andaman in the tropical forests (Bhattacharjee, 1980). About 43 species are found in Indian subcontinent, but commercially only four species namely, *Jasminum sambac* Ait., *Jasminum auriculatum* Vahl., *Jasminum grandiflorum* Linn. and *Jasminum multiflorum* Roth. locally called as Mogra, Jai, Jui and Kunda respectively (Priya Devi *et al*., 2013) important.

Man has been cultivating this flower for its fragrant properties and has been using it for ceremonial purpose since ancient times. In south India, a large quantity of jasmine flowers are used by women for adorning their hair. Its flowers are also being used for production of perfumed hair oil, attars, soap, cosmetics, syrups, aerated water, confectionary perfumes, disinfectants, ointments and even for medicinal purpose. The fragrance of jasmine flower cannot be imitated by any of the known synthetic aromatic chemicals. The Chinese have been using this flower to flavor the tea for many centuries. The flowers having a sweet taste are edible also.

Keeping in view its scope and the increasing demand throughout the year, the present investigation was carried out.

MATERIAL AND METHODS

The research was conducted at High-Tech nursery of College of Horticulture, Dr. Balasaheb Sawant Konkan Krishi Vidyapeeth, Dapoli, Ratnagiri (M.S.) in split plot design with three replication and two factors *viz*. levels of pruning ie., pruning at 40 cm above ground level (P_1), pruning at 60 cm above ground (P_2) and two levels each of two growth regulators G_1 - gibberellic acid (GA_3) 50 ppm, G_2 - gibberellic acid (GA_3) 100 ppm, G_3 - cycocel (CCC) 750 ppm, G_4 cycocel (CCC) 1000 ppm and G_5 - control. The vegetative parameters were recorded 90 days after pruning and application of the growth regulators. Flowering and yield parameters were recorded immediately after plucking. Statistical analysis of the data collected during the course studies was done using standard method of analysis of variance as described by Panse and Sukhatme (1995).

RESULT AND DISCUSSION

Vegetative parameters

Data in table 1 showed that the treatment $P_2 G_2$ (pruning at 60 cm above ground level) with gibberellic acid at $A_3$100 ppm (GA_3) was significantly promising in parameters like maximum plant height (147.80 cm), number of primary branches (5.87), secondary branches (53.53), girth of primary branch at base (24.28 mm), number of leaves (1751.00), average plant spread (135.20 cm) and average leaf area (66.30 cm^2) as against minimum plant height (125.40 cm), number of primary branches (3.97), secondary branches (38.27), girth of primary branch at base (10.89 mm), number of leaves (840.33), average plant spread (115.00 cm) and average leaf area and (39.46 cm^2) observed in P_1G_5 treatment with Pruning at 40 cm above ground level and control.

Table 1: Effect of pruning levels and growth regulators on vegetative parameters in Jasmine

Treatments	Plant height (cm)	No. of primary branches	No. of secondary branches	Girth of primary branch at basal stem (mm)	No. of leaves	Avg. plant spread (cm)	Avg. leaf area (cm^2)
P_1G_1	135.40	4.80	42.53	15.98	1336.33	124.93	48.28
P_1G_2	145.20	5.53	50.43	22.13	1692.00	133.33	62.39
P_1G_3	128.00	4.20	38.70	11.27	918.33	117.07	40.35
P_1G_4	138.27	4.80	44.67	16.60	1415.33	127.13	51.48
P_1G_5	125.40	3.97	38.27	10.89	840.33	115.00	39.46
P_2G_1	143.80	5.30	48.30	20.44	1603.00	130.47	59.08
P_2G_2	147.80	5.87	53.53	24.28	1751.00	135.20	66.30
P_2G_3	130.93	4.33	39.53	12.59	1063.33	120.73	42.88
P_2G_4	140.33	4.90	46.67	18.40	1505.67	129.27	55.70
P_2G_5	132.13	4.40	40.60	14.66	1200.00	122.73	46.35
Mean	136.73	4.81	44.32	16.72	1332.53	125.59	51.23
S.Em ±	0.38	0.11	0.67	0.47	22.62	0.41	0.51
CD@ 5%	1.14	0.32	2.02	1.42	67.80	1.24	1.54

Table 2: Effect of pruning levels and growth regulators on flowering parameters in Jasmine

Treatments	Avg. bud length (mm)	Avg. bud width (mm)	No. of flower buds $plant^{-1}$	Days required for flower initiation	Duration of flowering (days)
P_1G_1	23.50	9.93	581.67	70.74	174.82
P_1G_2	27.32	11.12	604.00	67.80	179.75
P_1G_3	21.16	8.90	568.00	73.54	172.13
P_1G_4	26.09	10.75	595.33	68.04	177.92
P_1G_5	20.17	8.12	555.00	75.75	170.30
P_2G_1	24.57	10.48	589.67	70.26	176.76
P_2G_2	30.24	11.87	625.33	64.53	182.42
P_2G_3	21.74	9.63	574.00	72.17	174.07
P_2G_4	29.86	11.76	611.00	66.33	181.43
P_2G_5	20.49	8.31	559.00	75.40	171.36
Mean	24.51	10.09	586.30	70.46	176.10
S.Em ±	0.26	0.11	1.13	0.30	0.30
CD@ 5%	0.78	0.34	3.39	0.90	0.91

Table 3: Effect of pruning levels and growth regulators on yield parameters in Jasmine

Treatments	Weight of 100 flowers (g)	Flower yield (g $plant^{-1}$)	Flower yield (kg ha^{-1})
P_1G_1	42.57	427.02	2965.48
P_1G_2	46.78	435.61	3025.02
P_1G_3	41.82	421.16	2924.73
P_1G_4	44.39	432.92	3006.35
P_1G_5	40.09	380.83	2644.65
P_2G_1	43.42	429.10	2979.84
P_2G_2	48.00	467.91	3249.37
P_2G_3	42.09	424.29	2946.43
P_2G_4	47.19	437.25	3036.49
P_2G_5	40.69	384.66	2671.20
Mean	43.70	424.08	2944.96
S.Em ±	0.21	4.95	29.77
CD@ 5%	0.63	14.83	89.24

These results are similar to those reported by the Pawar *et al.* (2019) with respect to vegetative parameters like plant height, which might be due to initial higher height and higher polysaccharide content in plants. The treatment GA_3 caused rapid cell elongation and improved source sink relation resulting in enhanced vegetative vigour in Jasmine. Similar pattern was observed by Kumar and Ughreja (1998) with respect to primary and secondary branches where pruning helped in suppression of apical dominance producing higher number of lateral branches. Further, the number of leaves, average plant spread and average leaf area were promising due to sufficient light intensity received by the plant due to adequate pruning levels which might have caused due to synthesis of food. This is in agreement with Lokhande *et al.* (2015) with respect to leaf area and maximum number of leaves as also reported by Dhanasekaran (2018).

Flowering parameters

Maximum average bud length (30.24 mm), average bud width (11.87 mm), number of flower buds/plant (625.33), duration flowering (182.42 days), minimum number of days required for flower initiation (64.53 days) was perceived under pruning at 60 cm above ground level with GA_3 100 ppm (P_2G_2). The minimum average bud length (20.17 mm), average bud width (8.12 mm), number of flower buds/plant (555.00), duration flowering (170.30 days), maximum number of days required for flower initiation (75.75 days) was noted in treatment Pruning at 40 cm above ground level with control (P_1G_5).

The levels of pruning exhibited significant variation in average bud length and the maximum length of flower. This could be due to increase in photosynthates due to enhance vegetative growth of plant which might have been utilized

for the production of better quality flower. The results are in close agreement with findings of Ghulam *et al.* (2004) in Rose. The study conducted by Lokhande *et al.* (2015) in *Jasminum sambac* (L.), recorded the maximum diameter of flower bud (10.25 mm), maximum number of flower buds per plant (593.30), minimum days (64.00 days) for emergence of first flower in plants pruned at 30 cm above ground level. This might be due to increased availability of photosynthesis due to enhanced vegetative growth of plant which might have been utilised for the production of better quality flower of jasmine. The minimum number of days required for flower initiation could be due to fact that pruning helps to broaden C: N ratio, thus stimulating flowering and increasing vigour of plant as a result of adequate pruning level. These results are close agreement with the findings of Ghulam *et al.* (2004) in rose. The application of GA_3 found promising in minimum days required for flower initiation (26.38 DAP) and maximum flowering duration (171.00 days) in plant sprayed with GA_3 150 ppm in Jasmine, Dhanasekaran (2018). This might be attributed to enhance vegetative growth in early phase due to exogenous application of GA_3 which may have favoured the increased photosynthesis and CO_2 fixation. Further it would have favoured convenience of factor influencing floral initiation i.e, carbohydrates pathway and photo periodic pathway with GA_3 pathway.

Yield parameters

Maximum weight of 100 flower (48.00 g), flower yield plant^{-1} (467.91 g), flower yield (3249.37 kg ha^{-1}) was found in treatment with pruning at 60 cm above ground level with GA_3 100 ppm (P_2G_2). Minimum weight of 100 flower (40.09 g), flower yield (plant^{-1}) (380.83 g), flower yield (2644.65 kg ha^{-1}) was found in treatment Pruning at 60 cm above ground level with GA_3 100 ppm (P_2G_2).

The pruning at 60 cm above ground left significant effect on weight of 100 flowers. Above mentioned results are in close confirmatory with Mundhe *et al.* (2018), who reported that pruning done at 50 cm above ground level produced superior weight of 100 flowers (21.00 g). Dhanasekaran (2018) noted that maximum mean hundred flower weight (600.76 g) was noticed with GA_3 150 ppm which is followed by the GA_3 100 ppm (595.66 g). During the period of research the pruning at 60 cm above ground level shows significant variation in flower yield plant^{-1} and kg ha^{-1}. The result of present research work is in close confirmatory with Mundhe *et al.* (2018). They observed that the increase in number of flowers plant^{-1} (3838.85) and (55.97 q ha^{-1}) observed in 50 cm height of pruning may be due to accumulation of cytokinin in the producing shoot and that could have caused increased number of flower per plant. The data pertaining to the flower yield per plant are in close confirmatory with Pawar *et al.* (2019) on *Jasminum sambac* var. Baramasi under south Gujarat condition. He concluded that significantly higher flower per plant (1116.28 g plant^{-1} and 6.69t ha^{-1}) was received in plants pruned at 50 cm from the ground level. The increase in yield was due to vigorous growth of plant and maximum number of productive shoots per plant.

CONCLUSION

It was concluded that the treatment combination P_2G_2 i.e. pruning at 60 cm above ground level coupled with application of gibberellic acid at 100 ppm was found promising with respect to vegetative, flowering and yield parameters in Jasmine under Konkan conditions.

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Biochemical basis of plant resistance in brinjal genotypes against shoot and fruit borer (*Leucinodes orbonalis*, Guenee)

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ABSTRACT

The biochemical factors responsible for resistance in brinjal genotypes against shoot and fruit borer was investigated during kharif 2017-18 at college of Horticulture, Venkataramannagudem using sixty genotypes and three check cultivars. Among all the genotypes evaluated, IC 136061 registered lowest shoot infestation (9.01%) and fruit infestation (12.73%) due to the presence of high quantities of phenol, peroxidase, PAL in shoot (4.60 mg g^{-1}, 6.62 units $^{-1}min^{-1}g^{-1}$, 7.96 µmol $^{-1}$ min $^{-1}$ mg^{-1}) and in fruit (5.31 mg g^{-1}, 7.56 units $^{-1}min^{-1}g^{-1}$, 8.26 µmol $^{-1}$ min $^{-1}$ mg^{-1}) compared to Dommeru Local (Susceptible check-1) which received highest shoot infestation (34.99%), fruit infestation (46.77%) due to low phenol, peroxidase, PAL content in shoot (0.54 mg g^{-1}, 1.62 units $^{-1}min^{-1}g^{-1}$, 1.21 µmol $^{-1}$ min $^{-1}$ mg^{-1}) and in fruit (0.61 mg g^{-1}, 1.77 units $^{-1}min^{-1}g^{-1}$, 1.43 µmol $^{-1}$ min $^{-1}$ mg^{-1}). The brinjal genotypes IC 136061 with high phenol, peroxidase and PAL content in shoot-fruit could be used in breeding programmes for developing resistant cultivars against shoot and fruit borer.

Keywords: Biochemical basis, Resistance, Brinjal genotypes, *Leucinodes orbonalis*

Brinjal (*Solanum melongena* L.) is widely grown vegetable of tropical and subtropical parts of the world. Due to its low calorific value (24 kcal 100 g^{-1}) and high potassium content (200 mg 100 g^{-1}), it is suitable for diabetes, hypertensive and obese patients. Among the major pests infesting the crop, shoot and fruit borer *Leucinodes orbonalis* (Guenee) was considered as main constraint in production all over the India, causing heavy yield losses up to 70 per cent (Jat and Pareek, 2003; Kalawate and Dethe, 2012).

Chemical control is widely used in managing insect pests in brinjal. Repeated uses of broad spectrum synthetic chemicals results in environmental contamination, bioaccumulation and biomagnifications of toxic residues and disturbance in ecological balance (Dadmal et al. 2004). Hence, there is an urgent need to look for an alternate and safer method.

The most important and effective way to manage an insect pest is the use of host plant resistance mechanism. IPM system along with host plant resistance is yielding promising and encouraging results and hence, development of insect resistant varieties is a potential objective of the crop scientists.

Biochemical factors of the host plant have been reported to play a vital role on resistance to various insect and disease pests (Panda and Khush, 1995) and relatively resistant varieties contained higher amount of secondary metabolites inherently (Dhaliwal and Dilawari, 1993). The biochemical defense mechanism would certainly be helpful in the selection of plants as a source of resistance. Exploiting host plant resistance through breeding approaches will be highly beneficial to develop superior high yielding genotypes with resistance to the shoot and fruit borer in brinjal. Identification of resistance is possible through quantifying the biochemical components present in the genotype. The biochemical constituents like glycoalkaloid (solasodine), phenols, phenolic oxidase enzymes namely poly phenol oxidase, peroxidase and Phenyl alanaine ammonialyase are available in brinjal and these biochemical constituents possess insect resistance properties (Kalloo 1988).

An understanding of different biochemical components of resistance is essential for developing strategies to breed for resistance to insect pests. Hence, the present investigation was carried out to identify the brinjal genotypes for resistance against shoot and fruit borer and to understand the biochemical basis of resistance.

MATERIAL AND METHODS

The present experiment was conducted at college of Horticulture, venkataramannagudem during kharif 2017-18. Sixty genotypes and three check cultivars of brinjal were screened against shoot and fruit borer in augmented block design. The seedlings were transplanted in the main field at 35-40 DAS in a single row of 5m length with a spacing of 70 cm x 60 cm. The checks were planted in a randomized manner after every eight test genotypes in each block. Recommended agronomic package of practices were adopted for raising the crop excluding the plant protection measures. Five plants were tagged in each genotype and checks at random.

Shoot and fruit samples from the apical shoots of selected plants of each genotype were collected. The leaves were clipped off and remaining shoot portion was taken. Similarly samples of edible size healthy brinjal fruits of the same physiological age were picked when the plants were at 90 to 120 days age. Total phenols from brinjal shoot and fruit was estimated by following Bray and Thorpe (1954) method. Peroxidase activity was determined according to the method suggested by Schimidt *et al.* (1982) whereas phenylalanine ammonialyase was estimated as per the procedure laid down by Dickerson *et al.* (1984).

The shoot infestation was recorded by counting the healthy as well as infested shoots (withered tender shoots) from randomly tagged plants of each genotype and checks at fortnightly intervals from 15 days after transplantation to final harvest. Mean per cent shoot infestation of each genotype was calculated. Data on fruit infestation was recorded from the randomly tagged plants of each genotype at each harvest. The per cent fruit infestation was worked out on number basis. Yield on weight basis from tagged plants of each genotype was recorded starting from first harvest to final harvest. Only healthy fruits were considered for recording the yield data.

Statistical analysis after appropriate transformation of data was undertaken (Gomez and Gomez, 1976). Data was analysed through statistical analysis software for augmented design (Rathore *et al*. 2004) using the mean values of all chemical constituents in shoot and fruit. Augmented design analysis was carried out according to the statistical procedure developed (Rathore *et al.* 2004). The following definitions and relations hold:

c = Number of check cultivars = 3

v = Number of tested genotypes = 60

b = Number of blocks = 8

n = v / b = Number of tested genotypes per block = 8

p = c + n = Number of plots per block = 11

N = bc + v = b (c + n) = total number of plots in the experiment = 88

The total number of blocks is determined by the need to have at least 10 degrees of freedom for error in the analysis of data on various parameters. This, in turn, is determined by the number of check varieties (c) used in the trial. In the analysis of variance of check varieties, the experimental error has (b-1) (c-1) degrees of freedom.

The first step of the analysis is to construct a two- way ANOVA using the data of check varieties across blocks, consequently, the resulted mean square error is used to adjust the tested genotypes mean for the block effect. Also, the resulted mean square error is used to estimate four orders of least significant differences LSD as follows:

1. LSD to compare between two check variety means = t 0.05 (2 MSE / b) 0.5 (ci-cj)
2. LSD to compare between adjusted mean of two tested genotypes in the same block = t 0.05 (2 MSE) 0.5 (BiVi-BiVj).
3. LSD to compare between adjusted mean of two tested genotypes in different blocks = t 0.05 (2 MSE (c + 1)/c) 0.5 (Vi-Vj).
4. LSD to compare between adjusted mean of tested genotype and a check variety mean = t 0.05 (MSE (b+1) (c+1)/ bc) 0.5 (Vi-Vj)

   Where, for all LSD values, tabulated t value has (b1) (c-1) degrees of freedom (df)

RESULTS AND DISCUSSION

Phenol

The observations on phenol content in shoot and fruit of the test genotypes were presented in Table 1. The data revealed that the phenol content in shoot of brinjal genotypes varied from 0.54 to 4.60 mg g^{-1}. The highest phenol content in shoot was recorded in IC 136061 (4.60 mg $g^{-1)}$ whereas the lowest (0.54 mg g^{-1}) in Dommeru Local. In genotypes, IC 136148 (4.20), IC 132912 (4.16), IC 136299 (4.08), IC 136041 (4.08), IC 136296 (3.96), IC 154517(3.88), A.Nidhi (3.88), IC 203589 (3.84), IC 213564 (3.84), IC 154517(3.79), Bhagymathi (3.78), S.Pratibh (3.74), JB-07 (3.70), IC 136189 (3.59), BLR - 24 (3.61), Aryana (3.58) and Jawere brinjal (3.12) the phenol content in shoot ranged between 3.00 to 4.20 mg g^{-1}. The phenol content in remaining genotypes ranged from 0.55 to 3.00 mg g^{-1}.

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Table 1. Biochemical factors of shoot and fruit in different brinjal genotypes screened against *L. orbonalis* during 2017-18

Block number	Genotype	Shoot			Fruit			Per cent shoot infestation	Per cent fruit infestation
		Phenol (mg gm^{-1})	Peroxidase (units $min^{-1}gm^{-1}$)	Phenylalanine ammonialyase (µmol min^{-1} mg^{-1})	Phenol (mg gm^{-1})	Peroxidase (units min^{-1} gm^{-1})	Phenylalanine ammonialyase (µmol min^{-1} mg^{-1})		
1	IC 136148	4.20	6.07	6.51	7.01	7.43	7.15	11.97 (20.24)	14.79 (22.61)
	IC 135912	4.16	5.92	6.46	6.94	7.35	7.20	13.02 (21.15)	15.83 (23.44)
	IC 136299	4.08	5.83	6.38	6.85	7.31	7.11	15.36 (23.07)	15.83 (23.44)
	Pb. Shree	3.09	3.61	4.73	4.39	4.92	5.25	21.53 (27.64)	26.04 (30.68)
	IC 136096	3.39	4.29	5.06	5.01	5.59	5.61	20.43 (26.87)	24.16 (29.44)
	IC 136017	3.31	4.42	5.26	5.23	5.80	5.80	20.87 (27.18)	22.29 (28.17)
	IC 089888	3.61	4.40	5.64	5.51	5.93	5.96	18.55 (25.51)	21.04 (27.3)
	IC 144515	3.53	4.48	5.52	5.56	6.01	6.02	19.71 (26.36)	20.41 (26.86)
2	IC 136231	3.57	4.93	5.59	5.98	6.24	6.36	18.19 (25.24)	19.37 (26.11)
	IC 136451	3.43	4.29	5.44	6.30	6.45	6.15	19.77 (26.40)	32.08 (34.5)
	IC 144525	3.40	5.00	5.75	6.06	6.73	6.41	17.95 (25.06)	19.58 (26.26)
	Swarnamani	3.06	3.74	4.81	4.47	5.01	5.32	21.87 (27.88)	25.41 (30.27)
	IC 136455	1.64	1.73	3.48	3.05	3.71	3.37	26.78 (31.16)	33.33 (35.26)
	IC 136308	2.44	3.06	3.87	4.03	4.27	4.92	23.46 (28.97)	27.25 (31.46)
	IC 136296	3.96	5.31	6.31	6.75	7.20	7.03	15.36 (23.08)	16.83 (24.22)
	IC 136041	4.08	5.69	6.39	6.79	7.26	7.15	15.00 (22.78)	15.83 (23.44)
	IC 136290	1.90	2.93	4.18	3.70	5.41	4.75	23.66 (29.10)	29.58 (32.94)
	Anamalika	1.93	1.68	3.59	2.95	3.69	3.23	23.31 (28.87)	33.75 (35.51)
3	DBR-08	1.39	1.90	3.62	3.18	3.80	3.95	26.35 (30.88)	32.5 (34.75)
	BVB-71-1	1.41	1.79	3.54	3.12	3.71	3.63	26.61 (31.05)	33.33 (35.26)
	P.Bindu	1.49	1.62	3.38	2.86	3.50	3.18	25.86 (30.56)	33.54 (35.39)
	JB-02	2.14	3.30	4.47	4.18	4.67	5.20	23.07 (28.70)	26.87 (31.22)
	AB-02	1.03	1.07	2.94	2.18	3.20	2.44	27.59 (31.69)	38.95 (38.62)
	A.Kurmakar	1.20	1.31	3.20	2.38	3.37	2.83	27.04 (31.33)	37.08 (37.51)
	KS 331	2.23	2.83	4.00	3.68	5.38	4.61	23.56 (29.04)	30.00 (33.21)
	Aryana	3.58	4.64	5.32	5.31	5.83	6.00	19.55 (26.24)	21.41 (27.56)
	DRNKV-104-43	0.98	1.20	3.12	2.23	3.25	2.51	26.28 (30.84)	38.75 (38.49)
4	Green long	0.63	0.77	1.79	1.77	1.91	2.21	29.27 (32.75)	42.5 (40.68)
	IVBL-116-131	1.06	1.27	3.14	2.31	3.25	2.70	26.91 (31.25)	37.91 (38.00)
	VR-02	0.70	0.84	1.89	1.88	2.00	2.21	27.83 (31.84)	42.08 (40.44)
	JB-03-06	3.32	4.38	5.21	5.11	5.68	5.86	19.55 (26.24)	23.12 (28.74)
	IC 136260	1.34	1.47	3.30	2.59	3.40	3.05	24.37 (29.58)	35.00 (36.27)
5	JB-64	1.99	2.21	3.72	3.40	3.93	4.20	24.97 (29.98)	30.83 (33.73)
	IC 136309	2.98	3.50	4.51	4.23	4.81	5.20	22.40 (28.25)	26.66 (31.09)
	BH-02	1.94	2.16	3.75	3.35	3.80	4.14	23.40 (28.93)	31.66 (34.24)
	IC 136306	3.20	3.95	4.86	4.60	5.17	5.50	20.58 (26.98)	25.62 (30.41)
	IC 203589	3.84	5.47	6.10	6.49	7.13	7.08	15.57 (23.24)	16.66 (24.09)
	IC 215021	0.86	0.95	2.90	2.15	2.00	2.32	25.48 (30.32)	39.33 (38.84)
	IC 137751	3.47	5.04	5.80	6.13	6.74	6.83	17.01 (24.36)	18.95 (25.81)
	IC 154517	3.79	5.14	6.25	6.72	7.12	7.03	15.83 (23.44)	16.66 (24.09)
6	IC 136292	3.19	4.19	4.93	4.93	5.43	5.65	19.81 (26.43)	25.41 (30.27)
	IC 213564	2.90	3.02	3.88	3.94	4.29	4.92	22.52 (28.33)	27.91 (31.89)
	JB-15	2.18	2.28	3.86	3.49	4.17	4.38	23.14 (28.75)	30.38 (33.44)
	IC 136258	2.12	2.49	3.55	3.41	4.12	4.38	24.16 (29.44)	30.41 (33.47)
	IC 136222	2.03	1.85	3.58	3.15	4.81	3.85	25.64 (30.42)	32.5 (34.75)
	IC 136189	3.59	3.75	5.41	5.38	5.91	5.97	18.84 (25.72)	21.66 (27.74)
	IC 136249	2.98	3.14	3.98	4.11	4.33	5.09	21.2 (27.42)	27.5 (31.62)
	IC136293	0.79	0.91	1.92	2.01	1.92	2.32	27.06 (31.34)	41.25 (39.96)

Block number	Genotype	Shoot			Fruit			Per cent shoot infestation	Per cent fruit infestation
		Phenol (mg g m^{-1})	Peroxidase (units $min^{-1}gm^{-1}$)	Phenylalanine ammonialyase (µmol min^{-1} mg^{-1})	Phenol (mg gm^{-1})	Peroxidase (units min^{-1} gm^{-1})	Phenylalanine ammonialyase (µmol min^{-1} mg^{-1})		
7	IC 136251	2.80	2.83	3.81	3.51	4.28	4.53	21.41 (27.56)	29.54 (32.92)
	A.Nidhi	3.88	5.10	6.25	6.73	7.04	6.85	14.09 (22.05)	17.29 (24.57)
	Jaware Brinjal	3.12	4.05	4.90	4.84	5.22	5.45	20.76 (27.1)	26.25 (30.82)
	IC 136307	2.70	3.50	4.64	4.28	4.89	5.21	21.01 (27.28)	26.24 (30.81)
	BLR-24	3.61	4.91	5.64	6.00	6.70	5.90	16.41 (23.9)	20.83 (27.15)
	S.Pratibh	3.62	5.01	5.88	6.18	6.81	5.68	16.65 (24.08)	18 (25.1)
	JB-07	3.70	5.07	5.99	6.31	6.92	6.99	16.15 (23.7)	17.25 (24.54)
	IC 136061	4.60	6.52	6.62	7.96	7.56	8.26	10.81 (19.19)	13.55 (21.6)
8	DRNKV-02-104	2.55	3.22	4.14	4.16	4.44	5.15	22.69 (28.45)	26.95 (31.27)
	IC 136589	1.93	2.12	3.68	3.23	4.92	4.07	25.14 (30.09)	32.08 (34.5)
	A.Abhilamb	1.42	1.56	3.32	2.67	3.42	3.14	27.1 (31.37)	34.16 (35.76)
	IC 136311	1.27	1.37	3.25	2.44	3.35	2.97	26.18 (30.77)	35.83 (36.77)
	Checks								
	Dommeru Local-SC	0.54	0.49	1.62	1.21	1.77	1.43	33.53 (35.38)	45.83 (42.61)
	Tadepalligudem Local-SC	0.60	0.70	1.88	1.60	1.88	1.99	31.79 (34.32)	43.41 (41.21)
	Bhagyamathi-RC	3.58	4.22	5.92	7.08	6.85	7.54	16.81 (24.2)	18.25 (25.29)
	Mean	2.36	2.97		4.13	4.56	4.59	28.47	32.77
CD (P=0.05)	0.54	0.88	0.58	0.85	0.88	0.71	0.77	1.60	2.10
SEM±	0.18	0.29	0.19	0.28	0.29	0.23	0.25	0.54	0.70
CD (P=0.05)	1.52	2.48	1.64	2.41	2.48	2.02	2.18	4.6	6.0
SEM±	0.49	0.81	0.54	0.79	0.81	0.66	0.71	1.54	2.05
CD (P=0.05)	1.86	3.03	2.01	2.95	3.03	2.47	2.67	5.7	7.0
SEM±	0.61	0.99	0.66	0.97	0.99	0.81	0.87	1.88	2.33
CD (P=0.05)	1.34	2.19	1.45	2.13	2.19	1.79	1.92	4.1	5.1
SEM±	0.44	0.71	0.47	0.70	0.71	0.59	0.63	1.35	1.69

Ci – Cj (Critical difference between two control treatments),BiVi – BiVj (Critical difference between two augmented treatments in the same block), Ci – Vj (Critical difference between control treatment and augmented treatment) Vi – Vj (Critical difference between two augmented treatments in different blocks).

Phenol content in fruit was in the range of 1.21 to 7.56 mg g^{-1}. IC 136061 registered high phenol content (7.56 mg g^{-1}) while the lowest was recorded in Dommeru Local (1.21 mg g^{-1}). The phenol content in Bhagymathi (7.08), IC 136148 (7.01), IC 132912 (6.94), IC 136299 (6.85), IC 136041 (6.79), IC 136296 (6.75), IC 154517(6.72), A.Nidhi (6.73), IC 203589 (6.49), JB 07 (6.31), S.Pratibh (6.18), IC 137751 (6.13), IC 144525 (6.06), BLR-24 (6.00) ranged from 6.00 to 7.08 mg g-1. The remaining test genotypes recorded phenol content in the range of 5.99 to 1.20 mg g-1.

The data on phenol content in shoot and fruit of 60 tested genotypes and three check cultivars for Kharif 2017-18 is presented in Table 1. In augmented block design there were four LSD values to compare the significant differences among tested genotypes and three check cultivars allowing all possible comparisons to be made to select the elite genotypes.

1. Comparison among the three check cultivars

Comparison among the three check cultivars for phenol content in shoot and fruit indicated that bhagyamathi with high phenol content (3.78 mg g^{-1}, 7.08 mg g^{-1}) differed strongly with the other two cultivars dommeru local (0.54 mg g^{-1}, 1.21 mg g^{-1}) and tadepalligudem local (0.60 mg g^{-1}, 1.60 mg g^{-1}) whereas non-significant difference was observed between dommeru local and tadepalligudem local.

2. Comparison among the tested genotypes in the same blocks

Pb.Shree with low phenol content in shoot (2.05 mg g^{-1}) and fruit (2.90 mg g-1) differed greatly with moderately resistant genotypes IC 136148(4.42 mg g^{-1}, 5.26 mg g^{-1}), IC 135912 (4.36 mg g^{-1}, 5.24 mg g^{-1}), IC 136299 (4.20 mg g^{-1}, 5.21 mg g^{-1}) and IC 136096 (3.96 mg g^{-1}, 5.01 mg g^{-1}) in the first block. The results of second block genotypes indicated that IC 136455 with low Phenol content in shoot and fruit (1.64

mg g^{-1}, 1.73 mg g^{-1}) showed highly significant difference with IC 136231(3.57 mg g^{-1}, 4.64 mg g^{-1}), IC 136451 (3.43 mg g^{-1}, 6.30 mg g^{-1}), IC 144525 (3.65 mg g^{-1}, 6.06 mg g^{-1}), IC 136296 (3.96 mg g^{-1}, 6.75 mg g^{-1}) and IC 136041 (4.08 mg g^{-1}, 6.79 mg g^{-1}). Pair wise comparison among the genotypes in the third block for phenol content in shoot revealed that IC 136290(2. mg g^{-1}), JB-02 (2.60 mg g^{-1}) differed with AB -02 (0.94 mg g^{-1}) as it contained with low phenol while pair wise comparison among the genotypes for phenol content in fruit revealed that all the genotypes are at par. DRNKV -104-43 (0.98 mg g^{-1}, 2.23 mg g^{-1}), green long (0.63 mg g^{-1}, 1.77 mg g^{-1}), VR-02 (0.70 mg g^{-1}, 1.88 mg g^{-1}) with low phenol content in shoot and fruit showed difference with Aryana (3.58 mg g^{-1}, 5.31 mg g^{-1}) and JB-03-06 (3.32 mg g^{-1}, 5.11 mg g^{-1}) in fourth block. Among the genotypes in the fifth block, IC 215021 with low phenol content in shoot (0.86 mg g^{-1}, 2.15 mg g^{-1}) differed significantly with IC 203589 (3.84 mg g^{-1}, 6.49 mg g^{-1}), IC 137751 (3.70 mg g^{-1}, 6.13 mg g^{-1}) and IC 154517 (3.90 mg g^{-1}, 6.72 mg g^{-1}). IC 136293 (0.79 mg g^{-1}, 2.01 mg g^{-1}) showed differences with IC 213564 (2.90 mg g^{-1}), IC 213564 (2.40 mg g^{-1}), IC 136189 (3.38 mg g^{-1}) and IC 136249 (2.50 mg g^{-1}) for phenol content in shoot and with IC 136189 (5.38 mg g^{-1}) for fruit in sixth block. Moderately resistant genotypes IC 136061 (4.60 mg g^{-1}, 7.96 mg g^{-1}), JB-07 (3.80 mg g^{-1}, 6.31 mg g^{-1}), S.Pratibh (3.74 mg g^{-1}, 6.18 mg g^{-1}) and A.Nidhi (3.88 mg g^{-1}, 6.73 mg g^{-1}) differed greatly with IC 136251 (2.24 mg g^{-1}, 3,51 mg g^{-1}) as it pocessed low phenol content in shoot and fruit in seventh block. No difference in phenol content of shoot and fruit was observed among eighth block genotypes in pair wise comparison.

3. Comparison among the tested genotypes in different blocks

In pair wise comparison among genotypes of different blocks showed significant difference in phenol content of shoot and fruit between IC 136061(4.60 mg g^{-1}, 7.96 mg g^{-1}) – IC 36455 (1.64 mg g^{-1}, 3.05 mg g^{-1}), IC 136148 (4.20 mg g^{-1}, 7.01 mg g^{-1}) – IC 136293 (0.79 mg g^{-1}, 2.01 mg g^{-1}), S.Pratibh (3.74 mg g^{-1}, 6.18 mg g^{-1}) - VR-02 (0.70 mg g^{-1}, 1.88 mg g^{-1}), IC 136041(4.08 mg g^{-1}, 6.79 mg g^{-1}) – AB-02 (0.94 mg g^{-1}, 2.18 mg g^{-1}), Aryana (3.27 mg g^{-1},5.31 mg g^{-1}) - Green long (0.63 mg g^{-1}, 1.77 mg g^{-1}), IC 203589 (3.84 mg g^{-1}, 6.49 mg g^{-1}) - IC 215021 (0.86 mg g^{-1}, 2.15 mg g^{-1}) and IC 154517 (3.90 mg g^{-1}, 6.72 mg g^{-1})-IC 136293 (0.79 mg g^{-1}, 2.01 mg g^{-1}). Non-significant difference in pair wise comparison among the genotypes was observed between IC 136148 (4.32 mg g^{-1}, 7.01 mg g^{-1}) - IC 144525 (3.65 mg g^{-1}, 6.06 mg g^{-1}), BVB-71-1 (1.69 mg g^{-1}, 3.12 mg g^{-1}) - IC 136260 (1.34 mg g^{-1}, 2.59 mg g^{-1}), DRNKV-104-43 (0.98 mg g^{-1}, 2.23 mg g^{-1}) -VR-02 (0.70 mg g^{-1}, 1.88 mg g^{-1}), IC 154517 (3.79 mg g^{-1}, 6.72 mg g^{-1}) - IC 136296 (3.96 mg g^{-1}, 6.75 mg g^{-1}) and IC 144515 (3.53 mg g^{-1}, 5.56 mg g^{-1}) - IC 144525 (3.65 mg g^{-1}, 6.06 mg g^{-1}) for phenol content in shoot and fruit.

4. Comparison among the tested genotypes and three check cultivars

The tested genotypes showed more phenol content in shoot and fruit compared to the check cultivars. Among the check cultivars, Bhagyamathi (3.58 mg g^{-1}, 7.08 mg g^{-1}) was on par with moderately resistant and tolerant genotypes. Dommeru local (0.54 mg g^{-1}, 1.21 mg g^{-1}) and Tadepalligudem local (0.60 mg g^{-1}, 1.60 mg g^{-1}) were identified as genotypes with very less phenol content in shoot and fruit recorded with significant difference with IC 136148 (4.20 mg g^{-1}, 7.01 mg g^{-1}),IC 135912 (4.16 mg g^{-1}, 6.94 mg g^{-1}), IC 136299 (4.08 mg g^{-1}, 6.85 mg g^{-1}), Pb.Shree (3.09 mg g^{-1}, 4.39 mg g^{-1}), IC 136017 (3.31 mg g^{-1}, 5.23 mg g^{-1}), IC 089888 (3.61 mg g^{-1}, 5.51 mg g^{-1}), IC 144525 (4.40 mg g^{-1}, 6.06 mg g^{-1}), IC 136292 (3.19 mg g^{-1}, 4.93 mg g^{-1}), IC 136296 (3.96 mg g^{-1}, 6.75 mg g^{-1}), IC 136041 (4.08 mg g^{-1}, 6.79 mg g^{-1}), JB-02 (2.14 mg g^{-1}, 4.18 mg g^{-1}), Aryana (3.58 mg g^{-1}, 5.31 mg g^{-1}), IC136309 (2.98 mg g^{-1}, 4.23 mg g^{-1}), IC 136306 (3.20 mg g^{-1}, 4.60 mg g^{-1}), IC 203589 (3.84 mg g^{-1}, 6.89 mg g^{-1}), IC 137751 (3.47 mg g^{-1}, 6.13 mg g^{-1}), IC 154517 (3.79 mg g^{-1}, 6.72 mg g^{-1}), IC 136292 (3.19 mg g^{-1}, 4.93 mg g^{-1}), IC 136189 (3.59 mg g^{-1}, 5.38 mg g^{-1}), A.Nidhi (3.88 mg g^{-1}, 6.73 mg g^{-1}), Jaware Brinjal (3.12 mg g^{-1}, 4.84 mg g^{-1}), IC 136307 (2.70 mg g^{-1}, 4.28 mg g^{-1}), BLR-249(3.61 mg g^{-1}, 6.00 mg g^{-1}), S.Pratibh (3.62 mg g^{-1}, 6.18 mg g^{-1}), JB-07 (3.70 mg g^{-1}, 6.31 mg g^{-1}), IC 136061(4.60 mg g^{-1}, 7.96 mg g^{-1}) and DRNKV-02-104 (2.55 mg g^{-1}, 4.16 mg g^{-1}) whereas non-significant difference was observed with other genotypes.

Phenol is the most abundant plant allelochemical found associated with feeding deterrent or growth inhibition of herbivores. Hence, genotypes with high phenol content in the shoot and fruit showed low level of shoot and fruit infestation while the susceptible and highly susceptible genotypes with low phenol content exhibited high shoot and fruit infestation. It clearly indicated that the phenol in shoot and fruit plays an important role in imparting resistance against shoot and fruit borer.

Peroxidase

Research finding presented in Table.1 on peroxidase content in shoot of brinjal genotypes revealed that the maximum and minimum content of peroxidase ranged from 1.62 to 6.62 units $min^{-1}g^{-1}$. Peroxidase activity was high in IC 136061 (6.62 units $min^{-1}g^{-1}$) whereas the lowest activity of Peroxidase (1.62 units $min^{-1}g^{-1}$) was recorded in Dommeru Local. Besides IC 136061, other genotypes viz., IC 136148 (6.51), IC 132912 (6.46), IC 136299 (6.38), IC 136296 (6.31), IC 136041 (6.39), IC 154517 (6.25), A.Nidhi (6.25), IC 203589

(6.10),JB 07 (5.99), S.Pratibh (5.88), IC 144525 (5.75), IC 136231 (5.59), IC 089888 (5.64), IC 144515 (5.52), Aryana (5.32) , IC 136017 (5.26), IC 136096 (5.06), IC 136292 (4.93), IC 136306 (4.86), Pb Shree (4.73), IC 136309 (4.51), (IC 136290 (4.18), JB-02 (4.47) and KS 331 were recorded with peroxidase activity in shoot in the range of 4.00 to 6.51 units $min^{-1}g^{-1}$. The remaining test genotypes were recorded with peroxidase activity in shoot in the range of 1.61 to 3.99 units $min^{-1}g^{-1}$.

Data pertaining to peroxidase content in fruits of brinjal genotypes ranged from 1.77 to 7.56 units min-1g-1. Peroxidase activity was high in IC 136061 (7.56 units $min^{-1}g^{-1}$) whereas the lowest activity of peroxidase (1.77 units min-1g-1) was recorded in Dommeru Local. Other genotypes viz., IC 136148 (7.43), IC 132912 (7.35), IC 136299 (7.31), IC 135041 (7.26), IC 136296 (7.20), IC 203589(7.13), IC 154517 (7.12), A.Nidhi (7.04), JB 07 (6.92), Bhagyamathi (6.85), S.Pratibh (6.81), IC 144525 (6.73), BLR 24 (6.70), IC 144515 (6.01), IC 136189 (5.91), IC 136017 (5.80), IC 089888 (5.93), IC 136093 (5.59), IC 136242 (5.43), Jaware Brinjal (5.22), IC 136306 (5.17) and IC 136292 (5.01) were recorded with peroxidase activity in fruit in the range of 5.00 to 7.43 units $min^{-1}g^{-1}$. The remaining test genotypes were recorded with peroxidase activity in fruit in the range of 1.76 to 5.00 units min-1g-1.

The data on peroxidase content in shoot of 60 tested genotypes and three check cultivars for 2017-18 season is presented in Table 1. In Augmented block design, there are four LSD values to compare the significant differences among tested genotypes and three check cultivars allowing all possible comparisons to be made to select the elite genotypes.

1. Comparison among three check cultivars

Comparison among the three check cultivars for peroxidase content in shoot and fruit indicated that Bhagyamathi with high peroxidase content (4.22 units $min^{-1}g^{-1}$, 6.85 units $min^{-1}g^{-1}$) differed strongly with the other two cultivars Dommeru Local (0.49 units $min^{-1}g^{-1}$, 1.77 units min-1g-1) and Tadepalligudem Local (0.70 units $min^{-1}g^{-1}$, 1.88 units $min^{-1}g^{-1}$) whereas non-significant difference was observed between Dommeru Local and Tadepalligudem Local.

2. Comparison among the tested genotypes in the same block

Pb.Shree with low peroxidase content in shoot and fruit (3.61 units $min^{-1}g^{-1}$, 4.92 units $min^{-1}g^{-1}$) differed with moderately resistant genotypes IC 136148 (6.07 units $min^{-1}g^{-1}$, 7.43 units $min^{-1}g^{-1}$), IC 135912 (5.92 units $min^{-1}g^{-1}$, 7.35 units $min^{-1}g^{-1}$) and IC 136299 (5.83 units $min^{-1}g^{-1}$, 7.31 units $min^{-1}g^{-1}$) in the first block. Low peroxidase content in shoot and fruit of IC 136455 (1.73 units $min^{-1}g^{-1}$, 3.71 units $min^{-1}g^{-1}$) showed significant difference with IC 136231 (4.93 units $min^{-1}g^{-1}$, 6.24 units $min^{-1}g^{-1}$), IC 136451(4.29 units $min^{-1}g^{-1}$, 6.45 units $min^{-1}g^{-1}$), IC 144525 (5.00 units $min^{-1}g^{-1}$, 6.73 units $min^{-1}g^{-1}$), IC 136296 (5.31 units $min^{-1}g^{-1}$, 7.20 units $min^{-1}g^{-1}$) and IC 136041 (5.69 units $min^{-1}g^{-1}$, 7.26 units $min^{-1}g^{-1}$) in the second block. Pair wise comparison among the genotypes in the third block for peroxidase content in shoot and fruit, AB 02 with low peroxidase content (1.07 units $min^{-1}g^{-1}$, 3.20 units $min^{-1}g^{-1}$) differed significantly with JB 02 (3.30 units $min^{-1}g^{-1}$) and IC 136290 (5.41 units $min^{-1}g^{-1}$). Green long (0.77 units $min^{-1}g^{-1}$, 1.91 units $min^{-1}g^{-1}$), VR-02 0.84 units $min^{-1}g^{-1}$, 2.00 units $min^{-1}g^{-1}$) with low peroxidise in shoot and fruit showed significant difference with Aryana (4.64 units $min^{-1}g^{-1}$, 5.83 units $min^{-1}g^{-1}$), JB-03-06 (4.38 units $min^{-1}g^{-1}$, 5.68 units $min^{-1}g^{-1}$) in fourth block. Among the genotypes in the fifth block, IC 215021 (0.95 units $min^{-1}g^{-1}$, 2.00 units $min^{-1}g^{-1}$) was significantly different in peroxidase content with IC 136309 (3.50 units $min^{-1}g^{-1}$, 4.81 units $min^{-1}g^{-1}$), IC 136306 (3.95 units $min^{-1}g^{-1}$, 5.17 units $min^{-1}g^{-1}$), IC 203589 (5.47 units $min^{-1}g^{-1}$, 7.13 units $min^{-1}g^{-1}$), IC 137751 (5.04 units $min^{-1}g^{-1}$, 6.74 units $min^{-1}g^{-1}$) and IC 154517 (5.14 units $min^{-1}g^{-1}$, 7.12 units $min^{-1}g^{-1}$). IC 136293 (0.91 units $min^{-1}g^{-1}$, 1.92 units $min^{-1}g^{-1}$) showed difference in peroxidase content with IC 136292 (4.19 units $min^{-1}g^{-1}$, 5.43 units $min^{-1}g^{-1}$), IC 213564 (3.20 units $min^{-1}g^{-1}$, 4.29 units $min^{-1}g^{-1}$), IC 136189(3.75 units $min^{-1}g^{-1}$, 5.91 units $min^{-1}g^{-1}$) in sixth block. The genotypes IC 136061 (6.52 units $min^{-1}g^{-1}$, 7.56 units $min^{-1}g^{-1}$), JB-07 (5.079 units $min^{-1}g^{-1}$, 6.92 units $min^{-1}g^{-1}$), S.Pratibh (5.01 units $min^{-1}g^{-1}$, 6.81 units $min^{-1}g^{-1}$) A.Nidhi (5.10 units $min^{-1}g^{-1}$, 7.04 units $min^{-1}g^{-1}$) due to high peroxidase content in shoot and fruit differed greatly with IC 136251 (2.83 units $min^{-1}g^{-1}$, 4.28 units $min^{-1}g^{-1}$) in seventh block.

No difference in peroxidase content of shoot and fruit was observed among eighth block genotypes in pair wise comparison.

3. Comparison among the tested genotypes in different blocks

In pair wise comparison among genotypes belonging to different blocks showed significant difference between IC 136061 (6.52 units $min^{-1}g^{-1}$, 7.56 units $min^{-1}g^{-1}$) – IC 136293 (0.91 units $min^{-1}g^{-1}$,1.92 units $min^{-1}g^{-1}$), IC 136148 (6.07 units $min^{-1}g^{-1}$, 7.43 units $min^{-1}g^{-1}$) – IC 136455 (1.73 units $min^{-1}g^{-1}$, 3.71 units $min^{-1}g^{-1}$), IC 136296 (5.31 units $min^{-1}g^{-1}$, 7.20 units $min^{-1}g^{-1}$) - AB-02 (1.07 units $min^{-1}g^{-1}$, 3.20 units $min^{-1}g^{-1}$), Aryana (4.64 units $min^{-1}g^{-1}$, 5.83 units $min^{-1}g^{-1}$) -VR-02 (0.84 units $min^{-1}g^{-1}$, 2.00 units $min^{-1}g^{-1}$), JB-03-06 (4.38 units $min^{-1}g^{-1}$, 5.68 units $min^{-1}g^{-1}$) - IC136293 (0.91 units $min^{-1}g^{-1}$, 1.92units $min^{-1}g^{-1}$), A.Nidhi (5.01 units $min^{-1}g^{-1}$, 7.04 units

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$min^{-1}g^{-1}$) - IC215021 (0.95 units $min^{-1}g^{-1}$, 2.00 units $min^{-1}g^{-1}$), IC 136041 (5.69 units $min^{-1}g^{-1}$, 7.26 units $min^{-1}g^{-1}$) - Green long (0.77 units $min^{-1}g^{-1}$, 1.91 units $min^{-1}g^{-1}$).

Non significant difference in pair wise comparison among the genotypes was observed between IC 136148 (6.07 units $min^{-1}g^{-1}$, 7.43 units $min^{-1}g^{-1}$)-IC 136231 (4.93 units $min^{-1}g^{-1}$, 6.24 units $min^{-1}g^{-1}$), IC 136296 (5.31 units $min^{-1}g^{-1}$, 7.20 units $min^{-1}g^{-1}$) – Aryana (4.64 units $min^{-1}g^{-1}$, 5.83 units $min^{-1}g^{-1}$), JB -03-06 (4.38 units $min^{-1}g^{-1}$, 5.68 units $min^{-1}g^{-1}$)- IC 137751 (5.04 units $min^{-1}g^{-1}$, 6.74 units $min^{-1}g^{-1}$), IC 213564 (3.02 units $min^{-1}g^{-1}$, 4.29 units $min^{-1}g^{-1}$) -IC 136260 (1.47 units $min^{-1}g^{-1}$, 3.40 units $min^{-1}g^{-1}$) and IC 136249 (3.14 units $min^{-1}g^{-1}$, 4.33 units $min^{-1}g^{-1}$) - IC 136307 (3.50 units $min^{-1}g^{-1}$, 4.89 units $min^{-1}g^{-1}$).

4. Comparison among the tested genotypes and three check cultivars

The tested genotypes showed more peroxidase content in shoot and fruit compared to the check cultivars. Among the check cultivars, Bhagyamathi (resistant check) recorded with (4.22 units $min^{-1}g^{-1}$, 6.85 units $min^{-1}g^{-1}$) which was found on par with moderately resistant and tolerant genotypes. Dommeru Local (0.49 units $min^{-1}g^{-1}$, 1.77 units $min^{-1}g^{-1}$) and Tadepalligudem Local (0.70 units $min^{-1}g^{-1}$, 1.88 units $min^{-1}g^{-1}$) were identified as genotypes with very low peroxidase content in shoot and fruit showed high differences with moderately resistant and tolerant genotypes which contained peroxidase content in shoot and fruit in the range of 4.46 to 6.51 units $min^{-1}g^{-1}$ and 5.00 to 7.43 units $min^{-1}g^{-1}$.

Many biochemical factors are known to be associated with insect resistance in crop plants. It is obvious in many cases that the biochemical factors are more important than morphological and physiological factors in conferring non-preference and antibiosis. Some biochemical constituents may act as feeding stimuli for insects. Occurrence at lower concentration or total absence of such biochemicals leads to insect resistance (Singh 1983).

Phenylalanine ammonialyase

Phenylalaninine ammonialyase (PAL) activity ranged between 1.62 to 6.62 $\mu mol^{-1} min^{-1} mg^{-1}$, in the shoot of the tested genotypes. IC 136061 recorded highest phenylalanine ammonialyase activity (6.62 $\mu mol^{-1} min^{-1} mg^{-1}$). Lowest was recorded in dommeru local (1.62 $\mu mol^{-1} min^{-1} mg^{-1}$) whereas PAL activity in the fruits of the sixty genotypes and three checks was in the range of 1.43 to 8.26 $\mu mol^{-1} min^{-1} mg^{-1}$. The highest PAL activity was observed in IC 136061 (8.26 $\mu mol^{-1} min^{-1} mg^{-1}$) and the lowest was observed in Dommeru Local (1.43 $\mu mol^{-1} min^{-1} mg^{-1}$). In augmented block design, there are four LSD values to compare the significant differences among tested genotypes and three check cultivars allowing all possible comparisons to be made to select the elite genotypes.

1. Comparison among three check cultivars

Comparison among the three check cultivars for PAL content in shoot and fruit indicated that bhagyamathi with high PAL content (5.92 $\mu mol^{-1} min^{-1} mg^{-1}$, 7.54 $\mu mol^{-1} min^{-1} mg^{-1}$) differed strongly with the other two cultivars dommeru local (1.62 $\mu mol^{-1} min^{-1} mg^{-1}$, 1.43 $\mu mol^{-1} min^{-1} mg^{-1}$) and tadepalligudem local (1.88 $\mu mol^{-1} min^{-1} mg^{-1}$, 1.99 $\mu mol^{-1} min^{-1} mg^{-1}$). There was no significant difference between Dommeru Local and Tadepalligudem Local.

2. Comparison among the tested genotypes in the same block

Pb.Shree with low PAL content in shoot and fruit (4.73 $\mu mol^{-1} min^{-1} mg^{-1}$, 5.25 $\mu mol^{-1} min^{-1} mg^{-1}$) differed with IC 136148 (6.51 $\mu mol^{-1} min^{-1} mg^{-1}$, 7.15 $\mu mol^{-1} min^{-1} mg^{-1}$), IC 135912 (6.46 $\mu mol^{-1} min^{-1} mg^{-1}$, 7.20 $\mu mol^{-1} min^{-1} mg^{-1}$) and IC 136299 (6.38 $\mu mol^{-1} min^{-1} mg^{-1}$, 7.11 $\mu mol^{-1} min^{-1} mg^{-1}$) in the first block. Low PAL content in IC 136455 (3.47 $\mu mol^{-1} min^{-1} mg^{-1}$, 3.37 $\mu mol^{-1} min^{-1} mg^{-1}$) showed significant difference with IC 136231(5.59 $\mu mol^{-1} min^{-1} mg^{-1}$, 6.36 $\mu mol^{-1} min^{-1} mg^{-1}$), IC 136451(5.44 $\mu mol^{-1} min^{-1} mg^{-1}$, 6.15 $\mu mol^{-1} min^{-1} mg^{-1}$), IC 144525 (5.75 $\mu mol^{-1} min^{-1} mg^{-1}$, 6.41 $\mu mol^{-1} min^{-1} mg^{-1}$), IC 136296 (6.31 $\mu mol^{-1} min^{-1} mg^{-1}$, 7.20 $\mu mol^{-1} min^{-1} mg^{-1}$) and IC 136041 6.39 $\mu mol^{-1} min^{-1} mg^{-1}$, 7.26 $\mu mol^{-1} min^{-1} mg^{-1}$) in the second block. Pair wise comparison among the genotypes in the third block for Phenylalanine Ammonialyase content in shoot and fruit revealed that all the genotypes were on par to each other. Green long (1.79 $\mu mol^{-1} min^{-1} mg^{-1}$, 2.21 $\mu mol^{-1} min^{-1} mg^{-1}$), VR-02 (1.89 $\mu mol^{-1} min^{-1} mg^{-1}$, 2.21 $\mu mol^{-1} min^{-1} mg^{-1}$), DRNKV-104-43 (1.20 $\mu mol^{-1} min^{-1} mg^{-1}$, 3.35 $\mu mol^{-1} min^{-1} mg^{-1}$) with low PAL content in shoot and fruit showed significant difference with Aryana (4.64 $\mu mol^{-1} min^{-1} mg^{-1}$, 5.83 $\mu mol^{-1} min^{-1} mg^{-1}$) and JB-03-06 (4.68 $\mu mol^{-1} min^{-1} mg^{-1}$, 5.68 $\mu mol^{-1} min^{-1} mg^{-1}$) in the fourth block. Among the genotypes in the fifth block, IC 215021 (2.90 $\mu mol^{-1} min^{-1} mg^{-1}$, 2.32 $\mu mol^{-1} min^{-1} mg^{-1}$) showed significant difference in PAL content with IC 136306 (4.86 $\mu mol^{-1} min^{-1} mg^{-1}$, 5.17 $\mu mol^{-1} min^{-1} mg^{-1}$), IC 203589 (5.47 $\mu mol^{-1} min^{-1} mg^{-1}$, 7.13 $\mu mol^{-1} min^{-1} mg^{-1}$), IC 137751 (5.04 $\mu mol^{-1} min^{-1} mg^{-1}$, 6.74 $\mu mol^{-1} min^{-1} mg^{-1}$) and IC 154517 (5.14 $\mu mol^{-1} min^{-1} mg^{-1}$, 7.12 $\mu mol^{-1} min^{-1} mg^{-1}$). IC 136293 (1.92 $\mu mol^{-1} min^{-1} mg^{-1}$, 2.32 $\mu mol^{-1} min^{-1} mg^{-1}$) showed significant difference in PAL content with IC 136292 (4.19 $\mu mol^{-1} min^{-1} mg^{-1}$, 5.43 $\mu mol^{-1} min^{-1} mg^{-1}$), IC 136189 (5.41 $\mu mol^{-1} min^{-1} mg^{-1}$, 5.97 $\mu mol^{-1} min^{-1} mg^{-1}$) in sixth block. The genotypes IC 136061 (6.62 $\mu mol^{-1} min^{-1} mg^{-1}$, 8.26 $\mu mol^{-1} min^{-1} mg^{-1}$), JB-07 (5.99 $\mu mol^{-1} min^{-1}$

^{1} mg^{-1}, 6.99 µmol^{-1} min^{-1} mg^{-1}), S. Pratibh (5.88 µmol^{-1} min^{-1} mg^{-1}, 5.68 µmol^{-1} min^{-1} mg^{-1}), BLR 24 (5.64 µmol^{-1} min^{-1} mg^{-1}, 5.90 µmol^{-1} min^{-1} mg^{-1}), A.Nidhi (6.25 µmol^{-1} min^{-1} mg^{-1}, 6.85 µmol^{-1} min^{-1} mg^{-1}) differed significantly with IC 136251 (3.81 µmol^{-1} min^{-1} mg^{-1}, 4.53 µmol^{-1} min^{-1} mg^{-1}) due to high PAL content in shoot and fruit.

No difference in PAL content of shoot and fruit was observed among eighth block genotypes in pair wise comparison.

3. Comparison among the tested genotypes in different blocks

Pair wise comparison among genotypes belongs to different blocks showed significant difference between IC 136148 (6.51 µmol^{-1} min^{-1} mg^{-1}, 7.15 µmol^{-1} min^{-1} mg^{-1}) – Green long (1.79 µmol^{-1} min^{-1} mg^{-1}, 2.21 µmol^{-1} min^{-1} mg^{-1}), IC 136061 (6.62 µmol^{-1} min^{-1} mg-1, 8.26 µmol^{-1} min^{-1} mg^{-1})-IC 215021 (2.20 µmol^{-1} min^{-1} mg^{-1}, 2.32 µmol^{-1} min^{-1} mg^{-1}), IC 136041(6.39 µmol^{-1} min^{-1} mg^{-1}, 7.15 µmol^{-1} min^{-1} mg^{-1})- VR 02 (1.89 µmol^{-1} min^{-1} mg^{-1}, 2.21 µmol^{-1} min^{-1} mg^{-1}), JB-03-06 (5.21 µmol^{-1} min^{-1} mg^{-1}, 5.81 µmol^{-1} min^{-1} mg^{-1})-IC 136293 (1.92 µmol^{-1} min^{-1} mg^{-1}, 2.32 µmol^{-1} min^{-1} mg^{-1}), IC 203589 (6.10 µmol^{-1} min^{-1} mg$^{-1, 7.13}$ µmol^{-1} min^{-1} mg^{-1}) –AB 02 (2.94 µmol^{-1} min^{-1} mg^{-1}, 2.44 µmol^{-1} min^{-1} mg^{-1}), IC 136299 (6.38 µmol^{-1} min^{-1} mg^{-1}, 7.11 µmol^{-1} min^{-1} mg^{-1}) – A. Abhilamb (3.32 µmol^{-1} min^{-1} mg^{-1}, 3.14 µmol^{-1} min^{-1} mg^{-1}) for PAL content in shoot and fruit.

In pair wise comparison among genotypes belongs to different blocks showed non-significant difference between IC 136061(6.62 µmol^{-1} min^{-1} mg^{-1}, 8.26 µmol^{-1} min^{-1} mg^{-1}) – IC 136299 (6.38 µmol^{-1} min^{-1} mg^{-1}, 7.11 µmol^{-1} min^{-1} mg^{-1}), IC 136231 (5.59 µmol^{-1} min^{-1} mg^{-1}, 6.36 µmol^{-1} min^{-1} mg^{-1}) – IC 136189 (5.41 µmol^{-1} min^{-1} mg^{-1}, 5.97 µmol^{-1} min^{-1} mg^{-1}), IC 136148 (6.51 µmol^{-1} min^{-1} mg^{-1}, 7.51 µmol^{-1} min^{-1} mg^{-1}) –IC 136296 (6.31 µmol^{-1} min^{-1} mg^{-1}, 7.03 µmol^{-1} min^{-1} mg^{-1}), Aryana (4.64 µmol^{-1} min^{-1} mg^{-1}, 5.83 µmol^{-1} min^{-1} mg^{-1}) – IC 137751 (4.04 µmol^{-1} min^{-1} mg^{-1}, 6.74 µmol^{-1} min^{-1} mg^{-1}), JB 07 (5.99 µmol^{-1} min^{-1} mg^{-1}, 6.99 µmol^{-1} min^{-1} mg^{-1}) - IC136189 (5.41 µmol^{-1} min^{-1} mg^{-1}, 5.97 µmol^{-1} min^{-1} mg^{-1}), Jaware Brinjal (4.90 µmol^{-1} min^{-1} mg^{-1} , 5.45 µmol^{-1} min^{-1} mg^{-1})- Aryana (4.64 µmol^{-1} min^{-1} mg^{-1}, 5.83 µmol^{-1} min^{-1} mg^{-1})- S. Pratibh (5.58, µmol^{-1} min^{-1} mg^{-1}, 5.68 µmol^{-1} min^{-1} mg^{-1})- JB 3-06 (5.21 µmol^{-1} min^{-1} mg^{-1}, 5.86 µmol^{-1} min^{-1} mg^{-1}) for PAL content in shoot and fruit.

4. Comparison among the tested genotypes and three check cultivars

The tested genotypes showed more PAL content in shoot and fruit compared to the check cultivars. Among the check cultivars, bhagyamathi recorded with 5.92 µmol^{-1} min^{-1} mg-1, 7.54 µmol^{-1} min^{-1} mg-1of PAL in shoot and fruit which was found on par with moderately resistant and tolerant genotypes. Dommeru local (1.62 µmol^{-1} min^{-1} mg^{-1}, 1.43 µmol^{-1} min^{-1} mg^{-1}) and Tadepalligudem Local (1.88 µmol^{-1} min^{-1} mg^{-1}, 1.99 µ mol^{-1} min^{-1} mg^{-1}) were identified as genotypes with very low PAL content in shoot and fruit which showed high significant differences with the moderately resistant and tolerant genotypes as they possessed the PAL in shoot and fruit in the range of 1.21 to 7.96 µ mol^{-1} min^{-1} mg^{-1}, 1.43 to 8.26 µ mol^{-1} min^{-1} mg^{-1} and non-significant difference was observed with susceptible and highly susceptible genotypes.

Many biochemical factors are known to be associated with insect resistance in crop plants. It is obvious in many cases that the biochemical factors are more important than morphological and physiological factors in conferring non-preference and antibiosis. Some biochemical constituents may act as feeding stimuli for insects. Occurrence at lower concentration or total absence of such biochemicals leads to insect resistance (Singh 1983). The biochemical constituents like glycoalkaloid (solasodine), phenols, phenolic oxidase enzymes namely polyphenol oxidase and peroxidase are available in brinjal and these biochemical constituents possess insect resistant properties (Kalloo 1988).

In present investigations, among the tested genotypes, IC 136061 (9.18%, 11.04 %), IC 136148 (11.97%, 14.79 %), IC 135912 (13.02 %, 15.83 %), A.Nidhi (14.09%, 17.29%), IC 136041(15.00%, 15.83%), IC 136296 (15.36%, 16.83%), IC 203589 (15.57%, 16.66%), 154517 (15.83%, 16.66%), JB 07 (16.15%, 17.35%), S.Pratibh (16.65%, 18.00%), IC 137751(17.01%, 18.95%), IC 144525 (17.95%, 19.58%), IC 136231 (18.19%, 19.37%), registered with shoot infestation in the range of 9.18 to 18.19 per cent and fruit infestation in the range of 11.04 to 19.58 per cent (Categorized as moderately resistant). Only five genotypes *viz*., IC 136293 (27.06%, 41.25%), VR-02 (27.83%, 42.08%), Green long (29.27%, 42.50%), Tadepalligudem Local (31.79%, 43.41%) and Dommeru Local (36.27%, 45.83%) were grouped under highly susceptible category as the incidence of shoot infestation was in the range of 27.00 to 45.83 per cent and fruit infestation was above 40.00 per cent.

Among the moderately resistant genotypes, IC 36061 has recorded with highest phenol (4.60, 7.96 mg gm^{-1}), peroxidase (6.52, 7.56 units min^{-1}gm^{-1}) and PAL (6.62, 8.26 µmol min^{-1} mg^{-1}) content in shoot and fruit resulted in lowest shoot and fruit infestation. Hence this genotype was selected and incorporated as a module in management of the pest. These results are in conformity with the findings of Panda and Das (1975) for presence of chemical constituents in brinjal genotypes as factors of resistance against shoot and fruit borer; Raju *et al.* (1987) identified that high total phenol

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content in brinjal genotypes conferred resistance against shoot and fruit borer. Jat and Pareek (2003) reported that the biochemical characters such as total sugars, free amino acids and crude protein were positively correlated with fruit borer infestation, while total phenols had negative correlation. Praneetha (2002) studied the biochemical basis of borer resistance and reported that the variety EP 65 had the highest level of peroxidase, polyphenol oxidase and total phenol contents in both shoot and fruit. The cross EP 65 x Pusa Uttam registered the highest level of peroxidase and polyphenol oxidase, while the cross EP 104 x APAU Bagmathi had more of total phenol content. Prabhu *et al*. (2009) observed that total phenol content and its activity is higher in shoots as compared to fruits at all stages of growth and the phenol content is the one of the most important character to reduce the shoot and fruit borer incidence. If the phenol content is high borer infestation will be less. According to Prasad *et al*. (2014) low phenol content in accession IC 090093 led to high susceptibility to EFSB whereas high phenol content in Pusa Shyamala rendered resistance against fruit borer. Other workers also claimed that higher phenol content in fruit was responsible for resistance against shoot and fruit borer. Some of the notable findings are Dadmal *et al*. 2004 b; Patil, 2014; Showket *et al*. 2017; Nirmala and Vethomani, 2016. The results pertaining to phenol content in fruit in the present investigation are also in similar trend *i.e* higher content in moderately resistant genotypes and lower quantities in susceptible genotypes. Hence the present findings are in accordance with the above reports.

According to Khorsheduzzaman *et al*. (2010) resistant lines have comparatively more PAL activity in fruit than the susceptible lines and it is responsible for resistance in brinjal genotypes. Several investigators made the similar type of observations in their studies. In an investigation, Martin (2004) obtained higher PAL activity in the wild relatives of brinjal, which showed higher resistance against BSFB in India. Similar findings were also reported by Engelberth (2000) in lima bean pod borer, Feltaon *et al*. (1999) in tobacco with *Heliothis virescens*, McConn *et al*. (1997) in *Manduca sexta*. The findings revealed that genotypes having higher PAL activity in shoot/fruit showed the lowest shoot and fruit bores infestation.

CONCLUSION

The biochemical analysis of genotypes revealed presence of higher phenol, peroxidise and phenylalanine ammonialyase (PAL) contents in the shoot and fruit of brinjal genotype IC 136061. Hence, IC 136061 could be used in breeding brinjal cultivars resistant to fruit and shoot borer.

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Principal component analysis and variability in coriander genotypes

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ABSTRACT

The investigation carried out with twenty four diverse genotypes for ten characters revealed high heritability coupled with high genetic advance as percentage of mean for seed yield per plant, umbels per plant, harvest index and seeds per umbellate indicating high additive gene effects and are least influenced by the environment. Hence, the selection based on these characters will be more effective and reliable for crop improvement. The principal component analysis revealed that three components possessed eigen value more than 1. The PC1, PC2 and PC3 contributed 24.03, 20.82 and 17.56 per cent of variability. Together, they accounted for 62.41 per cent of the variability of the genotypes.

Keywords: Coriander, heritability, genetic advance, Principal Component Analysis, yield.

Coriander (*Coriandrum sativum* L.), an annual herbaceous plant in the Apiaceae family, is one of the most essential seed spices used to add taste, flavour, and pungency to a variety of foods around the entire globe. It is also a frequent ingredient in the preparation of ayurvedic medicines and is a traditional home therapy for different ailments viz., rheumatism, joint pain, gastrointestinal complaints, flatulence (Said et al., 1996), indigestion, insomnia, convulsions, anxiety, loss of appetite (Emamghoreishi et al., 2005). It's grown for both seed and foliage. Coriander is a cool season (*Rabi*) crop in India when it is grown for seed.

India is recognized as the "Home of Spices," and a wide variety of spice producing species are produced throughout the country. The states of Rajasthan and Gujarat are the most important coriander-growing locations. In India, coriander is cultivated in an area of about 5.32 lakh hectares with total production of 7.09 lakh tones and productivity of 1333 kg hectare^{-1} (Aonoymous, 2018). Presently in Rajasthan, coriander is cultivated in an area of about 1.81 lakh hectares with a total production of 2.06 lakh tonnes and productivity of 1139 kg ha^{-1} (Anonymous, 2017).

Seed yield is the result of multiple morphological characters acting together. Component qualities are those characteristics that have an impact on the final character, the seed yield. The component traits are expected to be highly correlated to the seed yield, positively or negatively (Rymuza, 2012, Trnka, 2014). Evaluation of different coriander genotypes to identify of high yielder for a particular agro-climatic region will be more beneficial for realizing high yield and income. The principal component analysis as an important tools can be used in identifying the plant characters that categorize the distinctiveness among the promising genotypes. PCA helps in eradicating redundancy in data sets due to regular variation occurring regularly in the crop species (Maji and Saibu, 2012 and Ramakrishnan *et al.*, 2014). Hence, the considering the importance of PCA the present investigation was carried out in the rice germplasm.

MATERIAL AND METHODS

Twenty four genotypes namely, RCr-20, RCr-41, RCr-435, RCr-436, RCr-446, RCr-475, RCr-480, RCr-684, RCr-728, Hissar Anand, UD-565, UD-705, UD-706, UD-717, NS-2017-1, NS-2017-2, NS-2017-3, UD-50, UD-123, UD-182, UD-220, UD-246, UD-431 and UD-433 were evaluated in the Randomized Complete Block Design with three replications at Horticulture Farm, S.K.N. College of Agriculture, Jobner (Jaipur) during *Rabi season* 2018-19. This area in Rajasthan is classified as agro-climatic zone IIIA (Semi-Arid Eastern Plains). The seeds were planted on the 29th of October 2018 during the *rabi* season, with a row to row and plant to plant spacing of 30 x 10 cm. To develop a healthy crop, the entire agronomic package of practices were adapted. Five plants were randomly tagged and marked for observation in each replication.

Observations were recorded for 10 characters viz., plant height (cm), number of branches per plant, days to 50 percent flowering, number of umbels per plant, number of umbellets per umbel, number of seeds per umbellate, test weight (g), seed yield per plant (g), days taken to maturity and harvest index (%). Heritability in broad sense was calculated by the formula proposed by Hanson *et al*. (1956) and expressed in

percentage. Genotypic and phenotypic coefficient of variations was estimated according to Burton and Devana (1953) based on estimate of genotypic and phenotypic variance. The genetic advance (GA) for each character was calculated using the Johnson et al., 1955 formula. PCA analysis was carried out on 24 genotypes and 10 characters using SPSS package. The first three PCs were plotted in three dimensional and bi-plot in various combinations. Only the bi plots of the first three most informative components (PCAs) are presented.

RESULT AND DISCUSSION

Variability, heritability and genetic advance

Analysis of variance revealed significant differences among the accessions for all the quantitative traits studied indicating the presence of adequate variability which can be exploited through selection (Table 1). Table 2 shows estimates of genetic parameters such as genotypic and phenotypic coefficients of variation, heritability in the broad sense, and genetic progress, as well as the mean and range of various features. For all of the traits studied, there was a wide range of variation. Generally, phenotypic coefficient of variation revealed relatively high values in comparison to corresponding genotypic coefficients of variation for all the characters, indicating the effects of environment in expression of traits.

Table 1. Mean sum of square for growth and yield attributes in coriander genotypes

Characters	Mean sum of squares		
	Replications (d.f. = 2)	Genotypes (d.f. = 23)	Error (d.f. = 46)
Plant height (cm)	143.76	711.67**	24.29
Branches per plant	5.52	3.16**	0.22
Days to 50% flowering	49.94	114.17**	5.12
Umbels per plant	174.44	890.59**	11.56
Umbellates per umbel	9.32	2.14**	0.091
Seeds per umbellate	32.50	7.53**	0.042
Days to maturity	362.28	119.90**	37.33
Seed yield per plant (g)	41.51	123.40**	2.58
Test weight (g)	25.31	8.61**	0.33
Harvest index (%)	1027.72	438.78**	1.42

**- Significance at 1%.

The range of PCV was observed from 7.13 to 39.09 per cent for the traits under study which provides a picture of the extent of phenotypic variability in the population. The PCV was noted high for seed yield per plant, umbels per plant, harvest index and seeds per umbellate. Plant height, days to 50 per cent flowering, and umbellates per umbel had moderate PCV levels. PCV values were low for the rest of the characters. Sharma et al. (2016) and Kumar et al. (2016) found similar results (2018). Genetic coefficient of variation along with heritability estimates would provide clear picture on the efficiency of the selection (Burton, 1952). The estimate of broad sense heritability was the highest for all the characters except days to maturity (Table 2). The values were high for plant height, branches per plant, days to 50 per cent flowering, umbels per plant, umbellates per umbel, seeds per umbellate, seed yield per plant, test weight and harvest index. Similar, estimates of high heritability have been reported by Verma *et al.* (2018). All the traits under the study showed high magnitude of heritability in broad sense (70%), suggesting that the highly heritable characters were least affected by environmental variations and selection for these characters based on phenotypic performance may be more effective for coriander improvement.

Heritability alone does not provide clue for genetic gain resulting from the best selected individuals. Burton (1952) suggested that heritability, in combination with genetic advance (GA), was more reliable in predicting the effect of selection. In the present experiment, high genetic advance as percentage of mean was observed for seed yield per plant,

Table 2. Mean, range, variance, genotypic and phenotypic coefficients of variation, heritability (broad sense) and genetic advance for yield and other attributes of coriander

Characters	Mean	Range		Variance		Coefficients of variation		Heritability % (BS)	Genetic Advance	GA as (%) of mean
		Min.	Max.	Phenotypic	Genotypic	PCV	GCV			
Plant height (cm)	102.42	69.31	126.50	237.21	229.12	15.54	14.77	90.41	29.60	28.94
Branches per plant	7.20	5.23	8.73	1.05	0.98	15.25	13.74	81.16	1.83	25.50
Days to 50% flowering	57.54	44.00	70.00	38.05	36.35	11.91	10.47	87.65	11.62	20.20
Umbels per plant	47.64	21.20	96.74	296.86	293.01	36.63	35.92	96.20	34.58	72.59
Umbellates per umbel	5.75	4.03	7.33	0.71	0.68	15.32	14.39	88.16	1.60	27.83
Seeds per umbellate	6.55	4.15	10.22	2.50	2.49	24.31	24.10	98.33	3.22	49.24
Days to maturity	112.85	101.00	130.33	39.96	27.52	7.13	4.64	42.44	7.04	6.23
Seed yield per plant (g)	16.75	6.25	33.60	41.14	40.27	39.09	37.89	93.96	12.67	75.67
Test weight (g)	11.76	9.54	15.21	2.87	2.76	14.95	14.12	89.16	3.23	27.46
Harvest index (%)	42.46	27.33	75.38	146.25	145.78	28.57	28.43	99.02	24.75	58.29

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umbels per plant, harvest index and seeds per umbellate. High genetic advance as recorded in the present investigation was earlier reported for umbels per plant and seed yield per plant by Meena *et al.* (2013), for seeds per umbellate by Farooq *et al.* (2017); for harvest index by Kumar *et al.* (2018). The moderate estimate of genetic advance as percentage of mean was observed in characters like plant height, umbellates per umbel, test weight, branches per plant and days to 50 per cent flowering. Moderate genetic advance as recorded in the present investigation was earlier reported by Sharma *et al.* (2016) and Farooq *et al.* (2017).

High heritability with high genetic advance as percentage of mean was observed for seed yield per plant, umbels per plant, harvest index and seeds per umbellate indicated heritable nature of variation and scope for selection for these traits among the coriander genotypes. The high heritability combined with high genetic advance as per cent mean is indicative of additive gene action and hence, selection based on these parameters would be more reliable (Johnson *et al.* 1955).

Principal component analysis

PCA is a well-known dimension reduction method that may be used to condense a big collection of variables into a small set that retains the majority of the information from the larger set (Jolliffie, 2002). Only the first 3 PCs showed eigen values more than one, and they cumulatively explained 62.41 per cent variability (Table 3). The first PC explained 24.03 per cent of the total variation, and the remaining 2 PCs explained 20.82 and 17.56 per cent variation, respectively. The first absorbed and accounted for the greatest share of total variation in the set of all PCs, whereas the others accounted for progressively less and less variation. Similar result has been reported earlier by Dyulgerov and Dyulgerova (2013) in coriander. Eigen values of 10 principal components are shown within the scree plot (Fig. 1).

Table 3 : Eigen value and percent of total variation for various principal components in coriander

	Eigen values	Variance explained (%)	Cumulative explained (%)
PC1	2.403	24.03	24.03
PC2	2.082	20.82	44.85
PC3	1.756	17.56	62.41
PC4	0.977	9.77	72.18
PC5	0.789	7.89	80.07
PC6	0.713	7.13	87.2
PC7	0.527	5.27	92.47
PC8	0.471	4.71	97.18
PC9	0.234	2.34	99.52
PC10	0.048	0.48	100.00

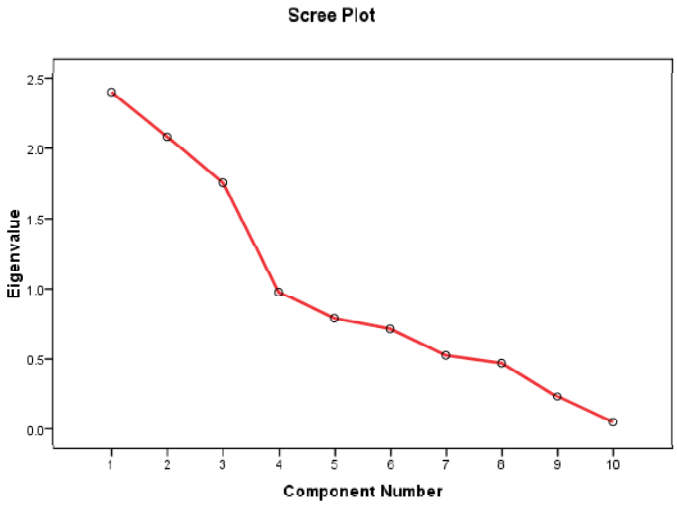

Fig.1 Scree plot showing the Eigen value variation for ten quantitative traits of coriander

Without rotating the axes, the analysis was unable to load all of the variables, implying that it could not provide any insight into the idea of correlation between the variables and the principal components. Varimax rotation was applied and this resulted in loading of all the variables on different principal components. Factors' loadings of different variables presented in Table 4 revealed that the first principal component (PC1) which accounted for the highest variation (24.03%) was mostly related traits for seed yield per plant, umbels per plant, seeds per umbellets. The second principal component (PC2) was dominated by traits *viz.*, plant height, number of primary branches per plant, umbellets per umbel and seeds per umbellets. A similar result was reported by Dyulgerov and Dyulgerova (2013) and Sastry *et al.* (2016) in coriander.

Principal factor scores (PF scores) for all the 21 genotypes were estimated for all the three PCs. These scores may be utilized to construct precise selection indices based on variability exhibited by each of the principal factor. Fig. 2 displays a biplot in the dimension of the first and second

Table 4 : Factor loadings of nine characters with respect to different PC's (principal components)

Characters	PC1	PC2	PC3
Plant height	-0.061	0.732	0.009
No. of primary branches per plant	0.266	0.520	0.649
Days to 50% flowering	-0.260	-0.119	0.775
Umbels per plant	0.815	0.193	-0.257
Umbellates per umbel	-0.429	0.479	-0.497
Seeds per umbellate	0.393	0.533	-0.084
Days to maturity	0.265	0.329	0.474
Seed yield per plant (g)	0.910	-0.061	-0.278
Test weight (g)	-0.556	0.278	-0.333
Harvest index (%)	0.226	-0.722	0.00

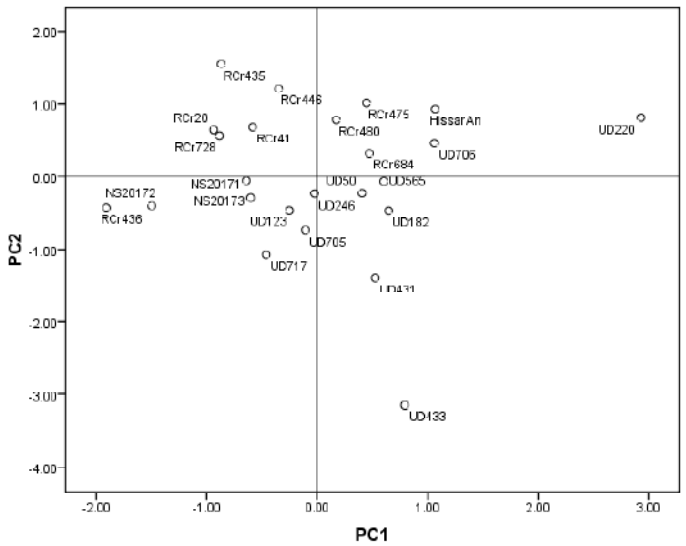

Fig. 2 Scatter diagram for PC1 and PC2 in 24 coriander accessions

PCs which together explained 62.41 per cent variability and included the major seed yield characters. Positive side of PC1 indicated that the genotypes giving high seed yield per plant, umbels per plant, and seeds per umbellets were indicative as UD-50, UD-565, UD-182, UD-431 and UD-433. Similarly, positive values of PC 2 were RCr-435, RCr-446, RCr-20, RCr-728 and RCr-41 and these genotypes were combining for plant height, number of primary branches per plant, umbellets per umbel and seeds per umbellets. The genotypes RCr-475, RCr-480, RCr-684, UD-706, Hissar Anand and UD-220 scored high and positively both for PC1 and PC2 i.e. the genotypes giving number of primary branches per plant, umbels per plant, seeds per umbelletes and days to maturity contributing characters.

CONCLUSION

The study indicates the possibility of reducing large number of variables into only three principal factors and identify different lines better for different combinations of characters. Hence, indirect selection for seed yield based on component traits may lead to create better genetic recombinants for improving yield and yield attributing characters.

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Evaluation of orange-fleshed sweet potato genotypes (*Ipomoea batatas* L.) for yield and quality parameters

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ABSTRACT

The experiment conducted with eight orange-fleshed sweet potato entries during *Kharif* seasons of 2018 to 2020 at the Regional Horticultural Research Station and extension center, Dharwad (Karnataka) revealed that the Sree Bhadra genotype yielded the highest total and marketable tuber yield of 26.75 and 23.53 t ha^{-1}, respectively. TSp 16-10 was found on par with Sree Bhadra producing total and marketable tuber yield of 26.67 and 23.17 t ha^{-1}, respectively. Higher dry matter and starch content of 22.17 and 17.68 per cent were recorded in TSp 16-3, respectively. While ST-14 recorded the higher ß –carotene, 12.99 mg 100g dry^{-1} weight, the TSp 16-10 and TSp 16-5, recorded significantly higher and lower sugar content of 4.64 and 2.32 per cent, respectively.

Key words: *Ipomoea batatas*, orange flesh sweet potato and ß –carotene

Sweet potato (*Ipomoea batatas* L. Lam), an important root crop of Asia, is grown all over the world in tropical and subtropical countries. Asia as a whole, accounts for about 78 per cent of the world area and about 92 per cent of the world production. India is one of the leading producers of this crop along with China, America, Brazil, Peru, Mexico and Thailand. China is the largest producer and consumer of sweet potato in the world accounting for about 67 per cent of global area and about 86 per cent of the global production. India accounts for about 68 per cent of the total production of South Asia followed by 27 per cent in Bangladesh and about 5 per cent in Sri Lanka (Yooyongech *et al.*, 2014). In India, Sweet potato is cultivated mainly in Orissa, Uttar Pradesh, West Bengal, Bihar, Karnataka, Tamil Nadu and Kerala. In India, sweet potato is grown in an area of 1.31 m ha with a production of 15.0 m t and productivity of 11.50 t ha-^{1}. In Karnataka it is cultivated on area of 2.71 thousand hectare and production 34.80 thousand tones (Anon., 2018). Sweet potato tubers are rich in starch, sugars, minerals and vitamins. Being rich in â-carotene, the orange- fleshed sweet potato is gaining importance as the cheapest source of antioxidant having several physiological attributes like anti-oxidation, anti-cancer and protection against liver injury and is most suiting as biofortified crop to combat malnutrition in small and marginal farming community. Orange fleshed sweet potato has considerable potential to contribute to a food based approach to tackle the problem of vitamin A deficiency, a major public health concern of the poorer sections. Thus, there is a great possibility of this subsistence crop for being adopted as regular diet of the consumer food chain to supplement as an alternative staple food source for the resource poor farmers. Hence, efforts were made to identify promising orange-fleshed sweet potato genotypes for commercial cultivation in northern part of Karnataka.

MATERIAL AND METHODS

The experiment was conducted during *Kharif* seasons of 2018 to 2020 at the Regional Horticultural Research Station and extension center, Dharwad (Karnataka), under the All India Co-ordinate Research Project (AICRP) on Tuber Crops. Eight genotypes of orange-fleshed sweet potato viz., TSp16-3, TSp16-5, TSp16-6, TSp16-7, TSp16-9, TSp16-10, Bhu Sona and Sree Bhadra (local check), were evaluated for yield, dry matter, starch, sugar, β-carotene and weevil infestation. Cuttings of these varieties were planted at a spacing of 60 cm x 20 cm in plots of size 3.0 m x 2.4 m. The plots were arranged in a randomized block design with three replications. Planting was done during May. The crop was grown under irrigated condition. Recommended package of practices (Anon., 2013) was followed and mature tubers were harvested at 120 days after planting. The data on various characters studied during the course of investigation were statistically analyzed as per the procedure outlined by (Panse and Sukhatme, 1985).

RESULTS AND DISCUSSION

The pooled data across three consecutive years revealed that the variety Sree Bhadra, among 08 entries tested, documented highest total and marketable tuber yield of 26.75 and 23.53 t ha^{-1}, respectively. TSp 16-10 was found on par with Sree Bhadra recording highest total and marketable

Table 1. Performance of different orange flesh Sweet potato entries for yield during 2018, 2019 and 2020

	Total tuber yield (t ha^{-1})				Marketable tuber yield (t ha^{-1})			
Entries	**2018**	**2019**	**2020**	**Pooled**	**2018**	**2019**	**2020**	**Pooled**
TSp16-3	22.70	23.50	21.20	22.47	18.88	20.81	19.25	19.65
TSp16-5	13.73	15.35	14.50	14.53	10.03	12.66	12.33	11.67
TSp16-6	16.78	18.65	17.21	17.55	12.53	15.67	14.56	14.25
TSp16-7	17.46	19.10	18.22	18.26	13.38	15.40	15.66	14.81
TSp16-9	18.62	20.85	19.25	19.57	14.82	16.89	18.20	16.64
TSp 16-10	26.25	28.45	25.30	26.67	22.50	24.47	22.55	23.17
Bhu Sona (ST-14)	25.47	26.33	23.45	25.08	21.40	22.50	21.00	21.64
Local (Sree Bhadra)	28.55	27.50	24.20	26.75	24.23	23.69	22.66	23.53
SE(m)	1.73	1.56	1.30	1.80	1.71	1.55	1.10	1.45
CD@ 5%	5.27	4.73	3.94	5.44	5.19	4.70	3.35	4.40
CV(%)	14.20	12.02	11.02	14.56	17.21	14.71	10.46	13.85

Table 2. Weevil infestation on different entries of orange flesh sweet potato during 2018, 2019 and 2020

Entries	**2018**	**2019**	**2020**	**Pooled**
TSp16-3	5.95	6.17	7.82	6.71
TSp16-5	5.95	6.50	7.12	6.59
TSp16-6	11.90	12.33	8.22	10.92
TSp16-7	5.95	12.33	15.53	11.38
TSp16-9	5.95	6.17	8.05	6.79
TSp 16-10	11.90	6.17	8.43	8.92
Bhu Sona (ST-14)	11.90	12.33	7.22	10.59
Local (SreeB hadra)	11.90	12.67	15.53	13.50
SE(m)	0.622	0.49	0.80	0.41
CD@ 5%	1.89	1.51	2.45	1.23
CV(%)	12.06	9.25	14.39	7.45

Table 3. Qualitative parameters of different orange flesh sweet potato entries during 2020-21

Entries	**Dry matter (%)**	**Starch (% Dry wt.)**	**Sugar (%)**	**ß-carotene (mg / 100g dry wt.)**	**Flesh colour**
TSp16-3	22.17	17.68	2.78	2.17	Orange
TSp16-5	25.33	21.06	2.32	4.88	Orange
TSp16-6	25.58	21.21	2.43	2.45	Orange
TSp16-7	28.99	23.74	2.68	4.17	Orange
TSp16-9	27.28	21.72	2.82	3.39	Orange
TSp 16-10	30.01	22.73	4.64	3.81	Orange
Bhu Sona (ST-14)	32.40	21.61	2.42	12.99	Deep Orange
Local (Sree Bhadra)	30.86	24.24	3.13	2.12	Cream
SE(m)	0.45	0.20	0.045	0.033	
CD@ 5%	1.91	0.86	0.19	0.14	
CV(%)	2.82	1.62	2.69	1.28	

tuber yield of 26.67 and 23.17 t ha^{-1}, respectively. While lower total and marketable tuber yield of 14.53 and 11.67 t ha^{-1}, respectively was recorded in TSp 16-5. Several researchers found that yield potentiality of sweet potato depends on the genetic make-up of plants (Miller *et al*., 2013; Surajit *et al*., 2012).

Significantly least weevil infestation of 6.59 per cent was recorded in TSp 16-5. TSp 16-3 (6.71 per cent) and TSp 16-9 (6.79) were on par with TSp 16-5. Similar results were reported by Allolli and Shetty (2012).

The results on the dry matter such as, starch content and ß –carotene determined during 2020-21 showed higher dry matter and starch content of 22.17 and 17.68 per cent respectively in TSp 16-3. The ß–carotene content in ST-14 was highest (12.99 mg 100g dry^{-1} weight). The entries TSp 16-5 (4.88 mg 100g dry^{-1} weight) and TSp 16-7 (4.17 mg 100g dry^{-1} weight) were on par with each other. Similar results were reported by Desai *et al.* (2013). In another study, the orange flesh sweet potato varieties exhibited high amounts of ß –carotenoids ranging between 7.91 and 12.85 mg 100 g^{-1} (Donado *et al*., 2012).

Significantly higher sugar content of 4.64per cent was recorded in TSp 16-10.

CONCLUSION

The study concludes that the sweet potato genotypes having large genetic diversity in terms of yield, weevil incidence, dry matter, starch, sugar ß –carotenoids and flesh colour can act as a barrier to many types of cancer and other chronic diseases. The study also revealed that orange flesh sweet potato genotypes that are rich in carotenoids and are also assumed to be rich in ß –carotene, a precursor of vitamin A, plays an important role in the alleviation of VAD in the children of developing countries. The study recommends consumption of orange flesh sweet potato to address nutrient deficiencies.

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Heterosis studies on yield components in hybrid rice (*Oryza sativa*. L)

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ABSTRACT

Thirty-two hybrids developed from crossing four CMS lines and eight testers were evaluated for relative heterosis, heterobeltiosis and standard checks for yield traits in rice (*Oryza sativa* L.) during *kharif* 2019. The results indicated that heterobeltiosis for grain yield was significantly positive and superior over eleven hybrids ranging from -52.94 (CMS 64A X JGL 34985) to 149.24% (JMS 19A X JGL 34985) and ten hybrids over check (PA 6444) ranging from -47.76 (CMS 64A X JGL 34985) to 67.35% (JMS 19A X JGL 34985). Most of the crosses exhibiting superiority over better parent or standard check for grain yield showed significant heterosis for spikelet fertility and 1000 grain weight. The top hybrids JMS 19A X JGL 34985, CMS 14A X NSR 61 and CMS 64A X JGL 34984 exhibiting significant positive heterosis for yield over standard hybrid (PA 6444) were can be further exploited commercially.

Keywords: heterosis, heterobeltiosis, hybrids, rice, standard heterosis, yield

Rice (*Oryza sativa* L.) is the most important cereal food crop of India. At present the global area under rice is 1.58 billion hectares with a production of 470.2 million tonnes per annum (www.indiastat.com, 2018-19). In the current scenario it is very important to ensure food and nutritional security for the increasing population. Therefore, enhancing productivity of rice through novel genetic approaches like hybrid rice was felt necessary. In plant breeding, heterosis is considered to be one of the outstanding achievements.

The degree of heterosis is calculated as the difference in the phenotypic performance of a trait between a hybrid and the average of its two distinct parents. Heterosis for a trait could be both positive and negative while the desired value of heterosis is dependent on the nature of the particular trait. The positive heterosis in general is used for yield whereas heterosis in negative direction is desired for earliness. The exploitation of heterosis can enhance yield from 30 to 40 per cent and can also enrich the domesticated crops with most important traits of qualitative and quantitative nature. Increased yield and good quality of rice hybrids can increase the profitability of the farmers. Khush *et al*. (1988) studied this subject intensively and concluded that hybridity *per se* did not harm grain quality in terms of physical and chemical characteristics as long as both parents possess acceptable grain quality, hybrid rice breeding programs must give emphasis to the critical evaluation of parental lines and hybrids for grain quality before these are released for commercialization.

MATERIAL AND METHODS

The experiment was conducted at Regional Agricultural Research Station (RARS), Polasa, Jagtial of Telangana state during *kharif*, 2019. Four CMS lines i.e., CMS 64A, JMS 19A, JMS 13A, CMS 14A and eight restorers *i.e.*, JGL 34984, JGL 34986, JGL 34551, JGL 34452, JGL 34985, JGL 32467, NSR 42 and NSR 61 were used to generate 32 hybrids through Line x Tester mating design. These 32 hybrids along with lines, testers and checks (PA 6444 and US 312) were planted at 20 x 15 cm spacing in two replications and grown in Randomized Block Design. Observations were recorded on eight characters *viz*., days to 50 per cent flowering, plant height (cm), panicle length (cm), number of productive tillers per plant, 1000 grain weight (g), number of grains per panicle, spikelet fertility (%) and grain yield per plant (g). The estimation of heterosis was calculated as procedure given by Fonseca and Patterson (1968).

RESULTS AND DISCUSSION

Heterosis was computed as percent increase or decrease in F_1 value over mid parent, better parent (heterobeltiosis) and best commercial hybrid (standard heterosis) presented in tables 1, 2 and 3 respectively. The

relative magnitude of heterosis over mid parent, better parent and standard checks were studied for eight yield characters *viz*., days to 50 per cent flowering, plant height, panicle length, number of productive tillers per plant, 1000 grain weight, number of grains per panicle, spikelet fertility and grain yield per plant in 32 hybrids. The five best cross combination and *per se* performance were given in table 4. The salient results obtained on different aspects and conclusions drawn from the experiment are summarized below.

Days to 50 per cent flowering: Negative heterosis is considered as desirable for days to 50 per cent flowering. Mid parent heterosis ranged from -10.84 (CMS 14A X JGL 34985) to 5.19 percent (CMS 14A X JGL 34984) and 13 crosses recorded significant negative heterosis. Better parent heterosis ranged from -13.40 (CMS 14A X JGL 34985) to -2.86 percent (CMS 14A X JGL 34986). 16 crosses showed negative significant heterosis over better parent. Standard heterosis over two checks was estimated and the lowest heterosis exploited was -12.56 per cent (over PA 6444) and -10.40 per cent (over US 312) in the cross CMS 14A X JGL 34985. The highest heterosis exploited was 3.86 per cent (over PA 6444) and 6.44 per cent (over US 312) in the cross JMS 19A X NSR 61. Significant negative heterosis was observed in 10 crosses over PA 6444 and 8 crosses over US 312.

Similar finding were reported by Gokulakrishnan (2018) and Shukla *et al*. (2020) indicating the possibility of exploiting heterosis for earliness.

Plant height

For plant height, heterosis is desirable in negative direction. Hetorisis over mid parent was ranged from -5.88 (CMS 64A X NSR 42) to 6.42 per cent (CMS 14A X JGL 34984). Among 32 hybrids, 7 recorded significant and negative values, whereas the better parent heterosis ranged from -15.31 (JMS 19A X NSR 61) to 5.75 percent (CMS 14A X JGL

Table 1. Estimates of heterosis over mid parent for yield traits in rice

Crosses	Days to 50 per cent flowering	Plant height	Panicle length	No. of productive tillers per	No. of grains panicle^{-1}	Spikelet fertility	1000-grain weight	Grain yield plant^{-1}
CMS 64A X JGL 34984	1.00	1.86 *	5.03	-8.57	-31.97 **	22.48 **	76.20 **	93.73 **
CMS 64A X JGL 34986	1.49	-0.68	4.33	10.53	-7.32	-54.25 **	11.26 *	-28.25 **
CMS 64A X JGL 34551	2.01	-0.14	-5.46	-13.51	4.40	-40.59 **	0.63	-30.73 **
CMS 64A X JGL 34452	2.24 *	4.96 **	2.39	-26.32 **	-17.02 *	-3.81	20.04 **	-26.50 **
CMS 64A X JGL 34985	1.74	5.56 **	0.10	11.76	-5.47	-12.03 **	3.52	-33.68 **
CMS 64A X JGL 32467	-6.47 **	1.72 *	7.34	-8.11	-30.74 **	-14.02 **	41.26 **	-18.33 **
CMS 64A X NSR 42	2.49 *	-5.88 **	-0.21	-16.28	25.85 *	-26.41 **	11.04 **	-9.61
CMS 64A X NSR 61	-2.40 *	-3.28 **	-2.77	-11.63	-29.85 **	-0.29	14.89 **	20.22 **
JMS 19A X JGL 34984	1.48	2.42 **	-14.00 **	13.33	-6.15	-1.12	14.19 **	51.00 **
JMS 19A X JGL 34986	1.47	6.29 **	5.94	21.21	-21.27 **	-10.41 **	47.35 **	42.98 **
JMS 19A X JGL 34551	-2.49 *	4.58 **	0.89	6.25	-22.65 *	-4.84	-0.62	113.38 **
JMS 19A X JGL 34452	1.23	4.83 **	4.44	15.15	-18.24 *	3.41	-6.16	13.10
JMS 19A X JGL 34985	-5.65 **	5.28 **	7.65 *	31.03 *	9.54	5.30	-4.97	194.43 **
JMS 19A X JGL 32467	-4.93 **	-0.15	9.54 *	68.75 **	3.45	-36.14 **	27.90 **	19.95 *
JMS 19A X NSR 42	1.97	-2.25 **	5.73	21.05 *	0.74	4.60	20.70 **	46.09 **
JMS 19A X NSR 61	2.38 *	-5.06 **	2.16	21.05 *	37.81 **	-7.26 *	-8.20 *	51.00 **
JMS 13A X JGL 34984	-1.93	3.39 **	0.73	33.33 **	-5.81	-17.95 **	2.52	35.21 **
JMS 13A X JGL 34986	-0.48	3.64 **	2.24	51.52 **	27.66 **	-80.06 **	15.99 **	-25.65 *
JMS 13A X JGL 34551	-3.90 **	1.83 *	-3.08	12.50	35.43 **	-10.99 **	-13.75 **	116.84 **
JMS 13A X JGL 34452	-1.21	-0.70	3.44	3.03	30.65 **	-8.43 **	2.21	-37.02 **
JMS 13A X JGL 34985	-0.24	4.74 **	7.01	24.14	-3.89	-3.27	4.21	-5.92
JMS 13A X JGL 32467	-6.76 **	0.26	13.15 **	56.25 **	29.38 **	1.79	9.88	40.71 **
JMS 13A X NSR 42	-3.38 **	-1.76 *	-0.32	21.05 *	-7.91	-1.14	-1.85	50.97 **
JMS 13A X NSR 61	-4.67 **	-2.82 **	2.15	10.53	17.31	-9.94 **	-8.70 *	22.30 *
CMS 14A X JGL 34984	5.19 **	6.42 **	8.81 *	48.72 **	89.28 **	-60.74 **	5.37	9.96
CMS 14A X JGL 34986	0.25	3.55 **	7.96 *	4.76	27.61 **	-70.44 **	30.96 **	-0.33
CMS 14A X JGL 34551	-5.24 **	1.94 *	14.17 **	-12.20	94.11 **	-11.36 **	-13.28 **	112.40 **
CMS 14A X JGL 34452	0.99	0.91	13.76 **	0.00	67.31 **	-17.28 **	13.28 *	20.08 *
CMS 14A X JGL 34985	-10.84 **	0.75	12.94 **	31.58 **	-5.40	-12.87 **	54.45 **	5.74
CMS 14A X JGL 32467	-0.25	2.85 **	27.81 **	-21.95 *	-12.05	-39.94 **	41.96 **	-30.32 **
CMS 14A X NSR 42	-3.70 **	-1.11	18.24 **	6.38	-2.37	-13.47 **	5.85	26.67 **
CMS 14A X NSR 61	-2.15 *	-2.79 **	15.43 **	19.15 *	66.01 **	-12.53 **	8.06 *	93.37 **

*Significant at 5% level, ** Significant at 1% level

Table 2. Estimates of heterosis over better parent for yield traits in rice

Crosses	Days to 50 per cent flowering	Plant height	Panicle length	No. of productive tillers	No. of grains panicle^{-1}	Spikelet fertility	1000 grain weight	Grain yield plant^{-1}
CMS 64A X JGL 34984	-2.40	0.68	3.57	-23.81 *	-38.19 **	1.94	46.20 **	36.40 **
CMS 64A X JGL 34986	-2.38	-1.79	3.27	0.00	-21.95 **	-54.84 **	-7.60	-41.18 **
CMS 64A X JGL 34551	-0.49	-3.72 **	-7.93 *	-23.81 *	-5.94	-45.41 **	-0.06	-51.10 **
CMS 64A X JGL 34452	-0.97	2.35 *	0.61	-33.33 **	-25.51 **	-11.83 **	-0.09	-34.11 **
CMS 64A X JGL 34985	-1.91	4.51 **	-0.41	-9.52	-6.74	-17.42 **	-12.14 *	-52.94 **
CMS 64A X JGL 32467	-9.62 **	-1.53	-3.06	-19.05	-41.45 **	-16.87 **	4.96	-19.35 *
CMS 64A X NSR 42	-0.96	-14.36 **	-1.02	-18.18	6.53	-31.87 **	-1.31	-12.41
CMS 64A X NSR 61	-8.56 **	-12.76 **	-5.75	-13.64	-42.77 **	-9.90 **	0.18	18.01 *
JMS 19A X JGL 34984	-0.96	0.00	-14.68 **	6.25	-18.45 *	-19.88 **	-9.33	26.44 *
JMS 19A X JGL 34986	-1.43	3.82 **	4.23	17.65	-36.31 **	-14.46 **	17.10 **	39.08 **
JMS 19A X JGL 34551	-3.92 **	-0.37	-1.16	6.25	-26.97 *	-9.73 **	-5.39	79.33 **
JMS 19A X JGL 34452	-0.97	0.99	2.02	11.76	-29.76 **	-2.15	-25.27 **	-16.33 *
JMS 19A X JGL 34985	-8.13 **	2.96 **	6.45	18.75	2.89	-4.22	-22.94 **	149.24 **
JMS 19A X JGL 32467	-7.21 **	-4.49 **	-1.61	68.75 **	-16.01 *	-36.14 **	-8.34	-4.66
JMS 19A X NSR 42	-0.48	-12.04 **	4.23	4.55	-10.96	0.00	12.97 **	14.48
JMS 19A X NSR 61	-3.15 **	-15.31 **	-0.38	4.55	17.11	-13.54 **	-15.81 **	22.90 **
JMS 13A X JGL 34984	-2.40	2.37 *	-3.97	25.00	-16.02 *	-34.12 **	-15.71 **	10.98
JMS 13A X JGL 34986	-1.43	2.66 **	-0.21	47.06 **	5.69	-81.18 **	-4.55	-25.86 *
JMS 13A X JGL 34551	-4.37 **	-1.65	-8.70 *	12.50	24.38 *	-14.59 **	-14.16 **	78.61 **
JMS 13A X JGL 34452	-1.45	-3.01 **	1.69	0.00	15.12	-12.37 **	-15.71 **	-52.62 **
JMS 13A X JGL 34985	-0.96	3.88 **	3.92	12.50	-7.13	-12.94 **	-12.39 *	-21.97
JMS 13A X JGL 32467	-7.21 **	-2.78 **	5.47	56.25 **	7.52	0.59	-18.97 **	13.98
JMS 13A X NSR 42	-3.85 **	-10.46 **	-2.90	4.55	-20.66 *	-4.40	-11.87 **	20.52 **
JMS 13A X NSR 61	-8.11 **	-12.21 **	-4.21	-4.55	-2.69	-15.10 **	-19.59 **	1.53
CMS 14A X JGL 34984	2.40	5.75 **	2.98	16.00	31.39 **	-68.26 **	-16.76 **	1.92
CMS 14A X JGL 34986	-2.86 *	2.95 **	4.58	-12.00	-15.45 *	-71.86 **	3.54	-12.93
CMS 14A X JGL 34551	-6.86 **	-1.19	6.77	-28.00 **	54.57 **	-15.68 **	-17.98 **	97.69 **
CMS 14A X JGL 34452	-1.45	-1.08	10.99 *	-16.00	15.28	-21.51 **	-10.26 *	-17.20 **
CMS 14A X JGL 34985	-13.40 **	0.29	8.87 *	0.00	-30.83 **	-20.96 **	24.58 **	-0.77
CMS 14A X JGL 32467	-2.88 *	0.09	20.00 **	-36.00 **	-41.59 **	-40.12 **	1.32	-48.92 **
CMS 14A X NSR 42	-6.25 **	-9.57 **	14.32 **	0.00	-17.71	-17.03 **	-0.28	-8.28
CMS 14A X NSR 61	-7.66 **	-11.89 **	7.47	12.00	45.45 **	-18.23 **	-0.27	44.66 **

Significant at 5 per cent level ** Significant at 1 percent level

34984). 12 crosses recorded significant heterosis in desired direction. The lowest standard heterosis exploited was -6.66 per cent (over PA6444) and -7.01 per cent (over US 312) in the cross CMS 64A X JGL 34986. The highest heterosis exploited was 2.48 per cent (over PA 6444) and 2.11 per cent (over US 312) in the cross CMS 14A X NSR 42. Significant negative heterosis was noticed in 16 crosses over PA 6444 and 18 in US 312.

Gokulakrishnan (2018), Sari *et al*. (2019) and Shukla *et al*. (2020) also emphasized the importance of negative significant heterosis for plant height to develop dwarf plant types.

Panicle length

With regard to panicle length, heterosis in positive direction is desirable. Out of 32 crosses, 11 exhibited significant positive heterosis over mid parent and values varied from -14.00 (JMS 19A X JGL 34984) to 27.81 per cent (CMS 14A X JGL 32467). Better parent heterosis ranged from -14.68 (JMS 19A X JGL 34984) to 20.00 per cent (CMS 14A X JGL 32467) and four crosses exhibited positive significant heterosis. The lowest standard heterosis exploited was -17.47 per cent (over PA 6444) and -11.52 per cent (over US 312) in the cross JMS19A X JGL 34984, while the highest standard heterosis exploited was 15.43 per cent (over US 312) in the cross CMS 14A X NSR 61. Significant positive standard heterosis was manifested in six crosses over US 312 and none of the crosses exhibited significant positive heterosis over PA 6444.

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Table 3. Estimates of heterosis over checks for yield traits in rice

Crosses	Days to 50 per cent flowering		Plant height		Panicle length		No. of productive tillers per plant		No. of grains panicle^{-1}		Spikelet fertility		1000 grain weight		Grain yield plant^{-1}	
	checks		checks		checks		checks		checks		checks		checks		checks	
	PA 6444	US 312	PA 6444	US 312	PA 6444	US 312	PA 6444	US 312	PA 6444	US 312	PA 6444	US 312	PA 6444	US 312	PA 6444	US 312
CMS 64A X JGL 34984	-1.93	0.50	-4.23 **	-4.58 **	0.19	7.41	-33.33 **	-20.00	-9.91	-23.75 *	-13.19 **	2.60	9.30 *	12.88 **	51.43 **	-9.51
CMS 64A X JGL 34986	-0.97	1.49	-6.66 **	-7.01 **	-2.88	4.12	-12.50	5.00	35.85 **	14.97	-61.54 **	-54.55 **	-30.92 **	-28.66 **	-34.69 **	-60.98 **
CMS 64A X JGL 34551	-1.93	0.50	-3.63 **	-3.98 **	-8.64 *	-2.06	-33.33 **	-20.00	12.03	-5.19	-44.51 **	-34.42 **	-24.25 **	-21.77 **	-45.71 **	-67.56 **
CMS 64A X JGL 34452	-0.97	1.49	0.09	-0.27	-5.37	1.44	-41.67 **	-30.00 **	11.56	-5.59	-9.89 **	6.49	-25.30 **	-22.86 **	-7.76	-44.88 **
CMS 64A X JGL 34985	-0.97	1.49	-0.92	-1.28	-6.33	0.41	-20.83 *	-5.00	14.15	-3.39	-29.67 **	-16.88 **	-34.31 **	-32.17 **	-47.76 **	-68.78 **
CMS 64A X JGL 32467	-9.18 **	-6.93 **	-2.25 *	-2.61 **	-8.83 *	-2.26	-29.17 **	-15.00	0.94	-14.57	-24.18 **	-10.39 *	-21.53 **	-18.96 **	-8.16	-45.12 **
CMS 64A X NSR 42	-0.48	1.98	-2.94 **	-3.30 **	-6.91	-0.21	-25.00 **	-10.00	26.89 *	7.39	-31.87 **	-19.48 **	-5.10	-2.00	3.67	-38.05 **
CMS 64A X NSR 61	-1.93	0.50	0.83	0.46	-5.57	1.23	-20.83 *	-5.00	-31.84 **	-42.32 **	-4.95	12.34 **	0.67	3.97	31.02 **	-21.71 **
JMS 19A X JGL 34984	-0.48	1.98	-4.87 **	-5.22 **	-17.47 **	-11.52 **	-29.17 **	-15.00	18.87	0.60	-26.92 **	-13.64 **	-23.98 **	-21.49 **	-15.10	-49.27 **
JMS 19A X JGL 34986	0.00	2.48	-1.33	-1.69	-0.77	6.38	-16.67	0.00	10.85	-6.19	-21.98 **	-7.79	-1.82	1.39	-1.22	-40.98 **
JMS 19A X JGL 34551	-5.31 **	-2.97 *	-0.28	-0.64	-1.92	5.14	-29.17 **	-15.00	-21.46	-33.53 **	-8.24 *	8.44 *	-20.67 **	-18.08 **	20.41 *	-28.05 **
JMS 19A X JGL 34452	-0.97	1.49	-1.24	-1.60	-2.88	4.12	-20.83 *	-5.00	5.19	-10.98	0.00	18.18 **	-37.35 **	-35.30 **	17.14	-30.00 **
JMS 19A X JGL 34985	-7.25 **	-4.95 **	-2.39 *	-2.75 **	1.34	8.64 *	-20.83 *	-5.00	25.94 *	6.59	-12.64 **	3.25	-35.39 **	-33.28 **	67.35 **	0.00
JMS 19A X JGL 32467	-6.76 **	-4.46 **	-5.19 **	-5.54 **	-6.33	0.41	12.50	35.00 **	44.81 **	22.55 *	-41.76 **	-31.17 **	-23.15 **	-20.63 **	8.57	-35.12 **
JMS 19A X NSR 42	0.00	2.48	-0.32	-0.69	-0.77	6.38	-4.17	15.00	-4.25	-18.96	0.00	18.18 **	8.63 *	12.18 **	35.51 **	-19.02 **
JMS 19A X NSR 61	3.86 **	6.44 **	-2.11 *	-2.47 **	-0.19	7.00	-4.17	15.00	25.94 *	6.59	-8.79 *	7.79	-15.39 **	-12.62 **	31.43 **	-21.46 **
JMS 13A X JGL 34984	-1.93	0.50	-2.62 **	-2.98 **	-7.10	-0.41	-16.67	0.00	22.41	3.59	-38.46 **	-27.27 **	-35.51 **	-33.40 **	-21.63 *	-53.17 **
JMS 13A X JGL 34986	0.00	2.48	-2.44 *	-2.79 **	-8.06 *	-1.44	4.17	25.00 *	83.96 **	55.69 **	-82.42 **	-79.22 **	-26.97 **	-24.58 **	-47.35 **	-68.54 **
JMS 13A X JGL 34551	-4.83 **	-2.48	-1.56	-1.92 *	-9.40 *	-2.88	-25.00 **	-10.00	41.98 **	20.16 *	-13.19 **	2.60	-34.31 **	-32.17 **	26.12 **	-24.63 **
JMS 13A X JGL 34452	-1.45	0.99	-5.15 **	-5.49 **	-7.68	-1.03	-29.17 **	-15.00	72.41 **	45.91 **	-10.44 **	5.84	-35.51 **	-33.40 **	-33.67 **	-60.37 **
JMS 13A X JGL 34985	0.00	2.48	-1.52	-1.88 *	-3.26	3.70	-25.00 **	-10.00	13.68	-3.79	-18.68 **	-3.90	-32.97 **	-30.77 **	-44.90 **	-67.07 **
JMS 13A X JGL 32467	-6.76 **	-4.46 **	-3.49 **	-3.85 **	-7.49	-0.82	4.17	25.00 *	85.38 **	56.89 **	-6.04	11.04 *	-38.00 **	-35.97 **	29.80 **	-22.44 **
JMS 13A X NSR 42	-3.38 **	-0.99	1.47	1.10	-10.17 *	-3.70	-4.17	15.00	-9.43	-23.35 *	-4.40	12.99 **	-15.26 **	-12.49 **	42.65 **	-14.76 **
JMS 13A X NSR 61	-1.45	0.99	1.47	1.10	-4.03	2.88	-12.50	5.00	11.08	-5.99	-10.44 **	5.84	-19.19 **	-16.55 **	8.57	-35.12 **
CMS 14A X JGL 34984	2.90 *	5.45 **	0.60	0.23	-0.38	6.79	20.83 *	45.00 **	91.51 **	62.08 **	-70.88 **	-65.58 **	-29.24 **	-26.92 **	-45.92 **	-67.68 **
CMS 14A X JGL 34986	-1.45	0.99	-2.16 *	-2.52 **	-3.65	3.29	-8.33	10.00	47.17 **	24.55 *	-74.18 **	-69.48 **	-11.98 **	-9.10 *	-38.16 **	-63.05 **
CMS 14A X JGL 34551	-8.21 **	-5.94 **	-1.10	-1.47	5.95	13.58 **	-25.00 **	-10.00	47.64 **	24.95 *	-14.29 **	1.30	-30.27 **	-27.99 **	4.90	-37.32 **
CMS 14A X JGL 34452	-1.45	0.99	-3.26 **	-3.62 **	0.77	8.02	-12.50	5.00	72.64 **	46.11 **	-19.78 **	-5.19	-23.71 **	-21.21 **	15.92	-30.73 **
CMS 14A X JGL 34985	-12.56 **	-10.40 **	-4.92 **	-5.27 **	1.34	8.64 *	4.17	25.00 *	-15.33	-28.34 **	-27.47 **	-14.29 **	5.91	9.38 *	-47.35 **	-68.54 **
CMS 14A X JGL 32467	-2.42	0.00	-0.64	-1.01	3.65	11.11 *	-33.33 **	-20.00	0.71	-14.77	-45.05 **	-35.06 **	-13.87 **	-11.05 *	-41.84 **	-65.24 **
CMS 14A X NSR 42	-5.80 **	-3.47 **	2.48 **	2.11 *	5.76	13.37 **	4.17	25.00 *	-32.08 **	-42.51 **	-17.03 **	-1.95	-4.11	-0.97	8.57	-35.12 **
CMS 14A X NSR 61	-0.97	1.49	1.84 *	1.47	7.68	15.43 **	16.67	40.00 **	9.43	-7.39	-13.74 **	1.95	0.22	3.50	54.69 **	-7.56

*Significant at 5 per cent level ** Significant at 1 percent level

Panicle length is one of the main attributes to higher yields and the results are in conformity with the findings of Patel *et al*. (2018) and Shukla *et al*. (2020).

Number of productive tillers per plant

For number of productive tillers per plant, relative heterosis ranged from -26.32 (CMS 64A X JGL 34452) to 68.75 per cent (JMS 19A X JGL 32467) and 11 crosses manifested significant positive heterosis. Whereas the better parent heterosis ranged from -36.00 (CMS 14A X JGL 32467) to 68.75 per cent (JMS 19A X JGL 32467) and only three crosses exhibited positive significant heterobeltiosis. The lowest standard heterosis exploited was -41.67 per cent (over PA 6444) and -30.00 per cent (over US 312) in the cross CMS 64A X JGL 34452. The highest standard heterosis exploited was 20.83 per cent (over PA 6444) and 45.00 (US 312) in the cross CMS 14A X JGL 34984. Among 32 hybrids studied, significant positive heterosis was recorded in 7 hybrids over US 312 and one hybrid over PA 6444.

In present investigation, some of the hybrids exhibited positive significant heterosis which are in accordance with the earlier findings of Prem Kumar *et al*. (2017) and Alice *et al*. (2018).

Number of grains per panicle

Relative heterosis ranged from -31.97 (CMS 64A X JGL 34984) to 94.11 per cent (CMS 14A X JGL 34551) and 11 crosses exhibited significant positive heterosis, while the heterobelpiosis ranged from -42.77 (CMS 64A X NSR 61) to 54.57 percent (CMS 14A X JGL 34551). Four crosses showed positive and significant heterobeltiosis. For number of grains per panicle the lowest standard heterosis exploited was -32.08 per cent (over PA 6444) and -42.51 per cent (over US

Table 4. Standard heterosis, heterobeltiosis and relative heterosis for top crosses for each trait in rice

Character/cross	Standard heterosis		Hetero-beltiosis	Relative heterosis
	PA 6444	US 312		
Days to 50% flowering				
CMS 14A X JGL 34985	-12.56**	-10.40**	-13.40**	-10.84**
CMS 14A X JGL 34551	-8.21**	-5.94**	-6.86**	-5.24**
JMS 13A X JGL 32467	-6.76**	-4.46**	-7.21**	-6.76**
JMS 19A X JGL 34985	-7.25**	-4.95**	-8.13**	-5.65**
CMS 64AX JGL 32467	-9.18**	-6.93**	-9.62**	-6.47**
Plant height				
CMS 64A X JGL 34986	-6.66**	-7.01**	-1.79	-0.68
CMS 64A X NSR 42	-2.94**	-3.30**	-14.36**	-5.88**
JMS 19A X NSR 61	-2.11**	-2.47**	-15.31**	-5.06**
JMS 13A X JGL 34452	-5.15**	-5.49**	-3.01**	-0.70
JMS 13A X NSR 61	1.47	1.10	-12.21**	-2.82**
Panicle length				
CMS 14A X JGL 34551	5.95	13.58**	6.77	14.17**
CMS 14A X JGL 34985	1.34	8.64*	8.87**	12.94**
CMS 14A X JGL 32467	3.65	11.11*	20.00**	27.81**
CMS 14A X NSR 42	5.76	13.37**	14.32**	18.24**
CMS 14A X NSR 61	7.68	15.43**	7.47	15.43**
No. of productive tillers per plant				
JMS 19A X JGL 32467	12.50	35.00**	68.75**	68.75**
JMS 13A X JGL 34986	4.17	25.00*	47.06**	51.52**
JMS 13A X JGL 32467	4.17	25.00*	56.25**	56.25**
CMS 14A X JGL 34984	20.83*	45.00**	-4.55	48.72**
CMS 14A X NSR 61	16.67	40.00**	12.00	19.15**
No. of grains per panicle				
JMS 19A X NSR 42	41.98**	20.16*	24.38**	0.74
JMS 13A X JGL 34551	85.38**	56.89**	7.52	35.43**
JMS 13A X JGL 32467	91.51**	62.08**	-2.69	29.38**
CMS 14A X JGL 34551	47.64**	24.95*	54.57**	94.11**
CMS 14A X JGL 34452	72.64**	46.11**	15.28	67.31**
Spikelet fertility				
CMS 64A X NSR 61	-4.95	12.34**	-9.90**	-0.29
JMS 19A X JGL 34551	0.00	18.18**	-2.15	-4.84
JMS 19A X NSR 42	0.00	18.18**	0.00	4.60
JMS 13A X JGL 32467	-6.04	11.04*	0.59	1.79
JMS 13A X NSR 42	-4.40	12.99**	-4.40	-1.14
1000 Grain weight				
CMS 64A X JGL 34984	9.30*	12.88**	46.20**	76.20**
JMS 19A X JGL 34986	-1.82	1.39	17.10**	47.35**
JMS 19A X NSR 42	8.63*	12.18**	12.97**	20.70**
CMS 14A X JGL 34985	5.91	9.38*	24.58**	54.45**
CMS 14A X JGL 32467	-13.87**	-11.05*	1.32	41.96**
Grain yield per plant				
CMS 64A X JGL 34984	51.43**	-9.51	36.40**	93.73**
JMS 19A X JGL 34551	20.41*	-28.05**	79.33**	113.38**
JMS 19A X JGL 34985	67.35**	0.00	149.24**	194.43**
JMS 13A X JGL 34551	26.12**	-24.63**	78.61**	116.84**
CMS 14A X NSR 61	54.69**	-7.56	44.66**	93.37**

* Significant at 5 per cent level ** Significant at 1 percent level

312) in the cross CMS 14A X NSR 42. While the highest standard heterosis exploited was 91.51 per cent (over PA 6444) and 62.08 per cent (over US 312) in the cross CMS 14A X JGL 34984. Significant positive standard heterosis was recorded in 13 crosses over PA 6444 and 9 crosses over US 312.

Similar kind of heterotic pattern was observed by Thakor *et al.* (2018) and Sari *et al.* (2019) who reported high heterotic effects for number of grains per panicle.

Spikelet fertility

The range of relative heterosis obsereved from -80.06 (JMS 13A X JGL 34986) to 22.48 per cent (CMS 64A X JGL 34984) and only one cross exhibited significant positive relative heterosis. The range of heterobeltiosis was -81.18 (JMS 13A X JGL 34986) to -9.73 per cent (JMS 19A X JGL 34551) and no crosses exhibited significant positive heterobeltiosis for this trait. The lowest standard heterosis manifested was -74.18 per cent (over PA 6444) and -69.48 per cent (over US 312) in the cross CMS 14A X JGL 34986, while the highest heterosis manifested was -8.24 per cent (over PA 6444) and18.18 per cent (over US 312) in the crosses JMS 19A X JGL 34551 and JMS 19A X NSR 42 respectively. Six crosses over US 312 and no crosses over PA 6444 exhibited significant and positive standard heterosis.

Similar results have been reported by Bedi and Sharma (2016) and Alice *et al*. (2018).

1000-grain weight

Total 16 crosses recorded significant positive heterosis over mid parent for 1000- grain weight with a range from -13.75 (JMS 13A X JGL 34551) to 76.20 per cent (CMS 64A X JGL 34984), whereas the significant positive heterobeltiosis was exhibited in 4 crosses which varied from -25.27 (JMS 19A X JGL 34452) to 46.20 per cent (CMS 64A X JGL 34984). The lowest standard heterosis recorded was -38.00 per cent (over PA 6444) and -35.97 per cent (over US 312) in the cross JMS 13A X JGL 32467, while the highest standard heterosis recorded was 9.30 per cent (over PA 6444) and 12.88 per cent (over US 312) in the cross CMS 64A X JGL 34984. Further, significant positive standard heterosis was recorded in 2 crosses over PA 6444 and 3 crosses over US 312.

The results are in akin with the findings of Priyanka and Jaiswal (2017), Sari *et al*. (2019) and Shukla *et al*. (2020).

Grain yield per plant

With respect to grain yield per plant 18 crosses exhibited significant positive relative heterosis in desired direction. The values ranged from -37.02 (JMS 13A X JGL 34452) to 194.43 per cent (JMS 19A X JGL 34985). Significant positive heterobeltiosis was recorded in 11 crosses and heterotic values over better parent ranged from -52.94 (CMS 64A X JGL 34985) to 149.24 (JMS 19A X JGL 34985). The highest standard heterosis recorded was 67.35 per cent in JMS 19A X JGL 34985 followed by 54.69 (CMS 14A X NSR 61), 51.43 (CMS 64A X JGL 34984) and 42.65 (JMS 13A X NSR 42) over

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PA 6444 and a total of 10 crosses exhibited significant and positive heterosis, whereas none of the hybrids exhibited significant heterosis over US 312 in desirable direction.

The findings are in consonance with earlier reports of Bedi and Sharma (2016), Krishna *et al.* (2016), Devi *et al.* (2017), Priyanka and Jaiswal (2017) and Shukla *et al*. (2020).

CONCLUSION

Out of 32 hybrids studied, the highest significant heterotic hybrids over mid parent identified were JMS 19A X JGL 34985, JMS 13A X JGL 34551 and JMS 19A X JGL 34551 and over better parent JMS 19A X JGL 34985, CMS 14A X JGL 34551 and JMS 19A X JGL 34551 in desirable direction. The highest significant standard heterosis was observed in hybrids CMS 64A X JGL 34984, JMS 19A X JGL 34551, JMS 19A X JGL 34985, JMS 13A X JGL 32467 and CMS 14A X NSR 61 over the check PA 6444. These hybrids exhibiting good heterotic expression may further be studied to isolate superior transgressive segregants in later generations.

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Application of robotics in changing the future of agriculture

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ABSTRACT

Agricultural production system has witnessed drastic changes in the last few decades with advancements in robotics and artificial intelligence based technologies. Scarcity of labour during peak cropping season has also highlighted the need for an alternative option for safe and sustainable agricultural system using IoT, machine learning and robotics for carrying out agricultural operations. Augmented use of electronics and computer application has made the working of robotic system possible for various field operations viz. transplanting, harvesting, and interculture, etc. for agricultural as well as horticultural crops. These technologies can be integrated with vision based system and GPS for more precise application. Robotic transplanter for transplanting plug-type seedlings may be a good option for future agriculture. It can use robotic arm, manipulator and an end-effector to perform the operation by using computer vision and motion planning algorithm or an artificial intelligence system. The application of robotics will help in various field operations for movement, localization, capturing, targeting and moving to the next target using drones for addressing spatial as well as temporal management of crops. Same operation can be used in spraying, weeding as well as harvesting of fruits. However, the robotic technology seems to be at nascent stage and there is a need to adopt these technologies due to non-availability of labour and their higher wages and to ensure timeless in field operations. Although many research attempts have been made for development of robots for agriculture application, more research should be focused towards the development of next generation robots for difficult and labourious farm operations.

Keywords: Agricultural Robotics; IoT; Artificial Intelligence; Machine Learning, Precision Agriculture

Agriculture is the backbone of our country and we need food for our survival. Conventional agricultural equipment and practices require large number of labour. The agriculture sector should focus on all possible directions including current agricultural practices and technology as well as modern equipment like agricultural robots so that proper care will be taken for sustainable agricultural production without compromising safety of farmers and agricultural labour. The application of robotics in agricultural operations is being attempted for transplanting, weeding, spraying, harvesting, etc. Many agri-robots have been developed in developed countries whereas research has initiated in some developing countries like India. A few researchers have carried out research in crop mapping, identification of fruits and disease using vision-based systems. Drones (UAV) have been extensively used for crop health monitoring and for spraying operation. Table 1 gives a brief summary of research attempts carried out on application of robotics in the agriculture sector. Though, most of the developed technologies are in proto-type stage, which needs prime concern for its research and development to bring these technologies at farmers' field in order to achieve sustainable agricultural production with minimal manpower.

Table 1. Studies carried out on the application of robotics in agriculture (Bechar and Vigneault, 2016)

Agricultural operation	Crops	Research attempts
Harvesting	Apples, citrus, strawberry, melons, etc.	302
Transplanting and seeding	Tomato, chilli, eggplant, etc.	255
Plant protection and weed control	Field crops	326
Grasping of fruits and vegetables	Apples, strawberry, tomato, capsicum, etc.	125
Navigation and mapping	-	700
Multi-robot interaction	-	60

Since limited work has been done on robotics application in agriculture and only a few successful technologies have been developed, this article emphasizes on the available robotic systems for various farm operations either in the field crop or horticulture. A brief details of these technologies for undertaking various agricultural operations have been discussed in the present paper.

Transplanting

Manual vegetable transplanting of plug seedlings is the most time intensive and laborious work (Khadatkar *et al*., 2018). Robotic transplanter may be a solution for the operation it not only saves time but also requires less labour. The robotic transplanters use computer graphics or machine vision system for simulating transplanting operation (Kim *et al*., 1995; Ryu *et al*., 2001; Choi *et al*., 2002; Khadatkar *et al*., 2018). They consist of a robotic arm for seedling pick-up, a path manipulator and an end-effector (Kang *et al*., 2012; Ma

et al., 2013; Mao *et al*., 2014; Han *et al*., 2015; Xin *et al*., 2018; Han *et al*., 2019). An intelligent transplanting system (Fig. 1) consists of 5 bar picking mechanism with fixed gear train, the seedling tray conveying mechanism, the planting mechanism, the seedling detection system using PLC (Xin *et al*., 2018). The PLC is used to control transmission and detect the void cell of seedling tray. By using robotic transplanter for transplanting seedlings of vegetables and crops, it will assure precision with safe and comfortable operation.

Interculture

Intercultural operation such as weeding is done to kill the weeds by mechanical weeders or chemical spraying. Manual hand weeding in field crop is considered as the most drudgerious farm operation and demands huge manual labour. Weeding by herbicides not only involves high input costs but also degrade the environment and hence the overall productivity. Robotic weeding may offer a potential alternative for conventional weeding practices using hand tools. Also, due to the strict protocols and restriction on the use of herbicides, robotic weeding offers the best alternative to manual weeding. Robotic weeder uses vision-based systems for weed detection, guiding weeder and uprooting weeds mechanically (Melander *et al*., 2015; Meng *et al*., 2015; Midti by *et al*., 2016). Co-robots (Fig. 2) developed by US National Science Foundation can work as human partner as a co-worker to perform a task jointly with ease (Perez-Ruiz *et al*., 2014). Gonzalez-de-Soto *et al*. (2016) developed a robotic patch spraying system for precision application of herbicide (Fig. 3).

Spraying

During spraying of agro-chemicals, contamination is the major problem which may cause threat to human health, if proper protection is not taken. Robotic sprayers are being developed and extensively used in orchards like apples, grapevine, cherries, etc. and to some extent in greenhouses. These sprayers are developed for target oriented application and to enhance input use efficiency. Oberti *et al*. (2016) stated that "In current farming practice, pesticides are typically applied uniformly across fields, despite many pests and diseases exhibiting uneven spatial distributions and evolving around discrete foci". The amount of agrochemicals used in precision horticulture can be reduced by effective site-specific application of pesticide (Maghsoudi *et al*., 2015). The recently developed automatic variable-rate sprayers requires an accuracy in measurement of location, canopy size and application of adequate amount of agro-chemicals to reduce environmental losses and save inputs (Jeon *et al*., 2011; Escolà *et al*., 2013; Gil *et al*., 2013). An autonomous system (Fig. 4) can also reduce labour requirement which can be employed for other activities, hence increases the crop yield, agricultural profitability and economic survival (Zaidner and Shapiro, 2016).

Harvesting

Fruit selection as well as detachment is one of the essential tasks for efficient harvesting. Most of the robotic harvester have been developed for fruits like apple, citrus (Fig. 5 & Fig. 6), cherries, strawberries, etc. (De-An *et al*., 2011; Silwal *et al*., 2017; Wang *et al*., 2019; Amatya *et al*., 2016; Zion *et al*., 2014). However, some harvesters for crops grown in greenhouse such as tomatoes, capsicum, etc. were also developed. The harvesting of fruits is accomplished by grasping the fruit with grippers and then detaching it on the basis of shape, size, colour and texture.

Aerial robot or drones

Aerial robot universally known as drone or unmanned aerial vehicle (UAV) is the new way to perform agricultural operations viz. crop mapping, scouting, spraying, etc during shortage of labour and precise management of agricultural inputs viz. chemicals, fertilizer, etc. UAVs, drones and radio-controlled model aircraft can be flown at lower altitudes to increase spatial resolution possibly at lower cost (Hunt et al., 2005). The advantage of UAV platform for agricultural management over conventional satellite imagery are very high pixels resolution, independent of cloud cover factors during critical periods of growth and instant information communication (Sinha, 2020; Fig. 7). Images acquired from UAV are used for determining utilization of shrub in vineyard management, mapping grass species, measuring shrub biomass, mapping crop vigour (Primicerio et al. 2012). Apart from these, vegetation mapping of rangeland (Laliberte and Rango 2009), patches of weed detection (Hardin et al. 2007), crop water stress monitoring, crop biomass recording (Hunt et al. 2005, Swain et al. 2010), and evaluating nitrogen treatments on crops (Hunt et al. 2005, Swain et al. 2010) are the other uses. UAV can also be used as geo-fencing to fend off animals' attacks by alerting the owner. Thakur (2016) reported reduction in cost of production by 25-30% by early detection of pest and efficient spraying by using drones in Chhattisgarh state of India. The aerial robot based spraying system enables farmers to apply protectants in time and safely.

Limitations in use of robots

The viability of agrobots for a wide range of agricultural applications has been evaluated extensively but the commercial applications of robots in field conditions are yet to be available. Field applications of robotics are still in the nascent stage due to some of the limitations as mentioned below.

1) The algorithms based on AI for sensing, mapping as well as controlling need to be work under tough, unstructured and dynamic field conditions.

2) The time bound activity of agriculture makes it challenging to attain the high level of utilization as it is found in manufacturing or industrial sectors.

3) Restricted automation as well as man-robot interfaces resulted in low production leading to delay in operations, low detection rates and inability to perform field operations under uncontrolled environments.

4) The use of aerial robots or drones (UAV) for agricultural operations is limited due to low payload capacity and battery life. Also, high initial as well as maintenance cost, fragile structure, skilled operator and a little knowledge are the major constraints to ensure use of aerial robotics at farm level.

The use of robots for agricultural operation seems to be impracticable at first instance, but its use has become crucial to provide food security for the burgeoning population of the world and to address the issue of labour shortage in agriculture. Due to migration of labour from agriculture to other sectors, there is shortage of labour and increase in demand for food day by day. The robotic machinery have potential for adoption for repetitive farm operations such as transplanting, weeding and harvesting which demand huge labour. It also offers additional advantages to overcome limited or non-availability of labours for transplanting, weeding, spraying and harvesting of fruits and vegetables. Research also shown that drones can be used to assess the spatial and temporal management of field crop which can be utilized for better crop planning and monitoring at large scale.

While data oriented digital solutions are available in the form of mobile app, the adoption of robotic technology is at a nascent phase. Some technologies viz. driverless tractors, laser land levelers, and harvesters for strawberries, tomatoes, capsicum in greenhouse are being developed, but the major constraints are in the adoption as well as commercialization of such high-end technologies. It may be due to climatic as well as geographical conditions, high cost, less efficient, need of skilled operator, etc. However, some robotics applications are now commercially available for fruit harvesters, sprayers and autonomous combines or tractors. These technologies need to be tested, improved, popularized and adopted on mass scale to address labour shortage in future. The process for implementing these autonomous robots highlights the need to have collaborative human-robot systems. Hence, multi-stakeholder initiative might speed up the adoption of robots in the agricultural sector.

CONCLUSION

Application of robotics technology in agriculture needs to be adopted and to ensure timely field operations due to higher labour wages.

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Integrated nutrient management in green gram (*Vigna radiata* L.)

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ABSTRACT

The study carried out at CCS Haryana Agricultural University, Hisar during *kharif* 2019 revealed that the treatment combination of 75 per cent RDF+vermicompost @ 1t ha^{-1} + PSB + *Rhizobium*. recorded maximum plant height (68.50cm), dry matter accumulation per plant (48.61g ha^{-1}), pod length (8.21 cm), number of pods (13.61), number of seeds per pod (7.49), seed index (4.78), grain yield (13.26 q ha^{-1}), straw yield (26.52 q ha^{-1}), biological yield (39.78 q ha^{-1}), harvest index (28.13), cost of cultivation (21200 Rs ha^{-1}), gross and net monetary returns (98124 Rs ha^{-1} and 76929 Rs ha^{-1}, respectively) and BC ratio (3.62) against minimum plant height (52.46 cm), plant dry matter accumulation (35.68g ha^{-1}), pod length (5.80 cm), number of pods (9.72), number of seeds per pod (6.93), grain yield (7.46 q ha^{-1}), straw yield (14.92 q ha^{-1}), biological yield (22.38 q ha^{-1}), harvest index (26.90), cost of cultivation (17790 Rs ha^{-1}), gross and net monetary returns (55204 Rs ha^{-1} and 37414 Rs ha^{-1}, respectively) and BC ratio (2.10) recorded in control. Cultivation of green gram cv. MH 421 was found more productive and profitable with the application of 75 per cent RDF + vermicompost @ 1t ha^{-1} + PSB + *Rhizobium*.

Key words: Integrated nutrient management, vermicompost, Rhizobium, green gram

Green gram [*Vigna radiata* (L.) Wilczek], also known as mung bean, is one of the excellent sources of high quality protein (25%) and ranks third after chickpea and pigeon pea. In India it occupies an area of 4.26 million hectares with a production of 2.01million tonnes in 2017-18 (Anonymous, 2018). The productivity of this crop is very low because of its cultivation generally on marginal and sub marginal lands having lesser soil fertility and little attention paid on proper fertilization (Saravanan *et al*., 2013). Chemical fertilizers play an important role in meeting the nutritional requirement of mungbean crop, but imbalanced and continuous use of chemical fertilizers adversely affect the physical, chemical and biological properties of the soil affecting sustainable crop production, besides causing a huge environment and soil pollution (Virmani, 1994). The regular use of chemical fertilizers adversely affects the physical and chemical properties of soil making it acidic or saline. Therefore, there is an immediate need to reduce of replace the use of chemical fertilizers and in the usage of organic sources of nutrients. The scope for improving the production potential of this crop can be increased with the use of inorganic manures, organic manures and biofertilizers in conjugation or in different combinations. Therefore, a way of integration, to grow green gram by combining organic and inorganic fertilizers along with biofertilizers is a better alternative. The integration of various sources of nutrients *i.e.* organic as well as inorganic may become an important strategy for sustainable crop production.

The INM techniques have provided way to grow mungbean crop by using both inorganic and organic nutrient sources like vermicompost along with biofertilizers like *Rhizobium* and PSB. Its main focus is to achieve four major goals viz., to maintain soil productivity, ensure sustainable production, prevent degradation of the environment and reduce expenditure on the chemical fertilizers. Keeping these facts in view, the effect of integrated nutrient management on growth, yield and economics of mungbean was studied.

MATERIAL AND METHODS

The experiment was conducted on green gram *Cv.* MH 421 at Agronomy Farm of CCS Haryana Agricultural University, Hisar located at 29.09^{o}N and 75.43^{o}E during *Kharif* 2019. The climate of this region is typically semi-arid, characterized by extremes of temperatures during summers and winters (as high as 45^{0}C in summer and as low as–1.0 ^{0}C in winter). The average annual rainfall of this tract ranges between 250-300 mm, but only 247.2 mm rainfall occurred during experiment period (Figure1). The soil of experimental field was loamy sand in texture, alkaline in reaction, low in nitrogen and phosphorus and medium in potassium.

The experiment consisted of thirteen treatment combinations involving different levels of fertilizers, vermicompost, biofertilizer inoculations (PSB and *Rhizobium*) and an absolute control laid out in randomized block design in three replications. The thirteen treatment combinations were T_1-control, T_2-vermicompost@2.5t ha^{-1}, T_3-*Rhizobium* + PSB, T_4-50 per cent RDF, T_5-75 per cent RDF, T_6-RDF, T_7-50 per cent RDF + *Rhizobium* + PSB, T_8-50 per cent RDF + vermicompost @ 1t ha^{-1}, T_9-50 per cent RDF +1t ha^{-1} vermicompost+ PSB + *Rhizobium,* T_{10}-75 per cent RDF +

Rhizobium + PSB, T_{11}-75 per cent RDF + vermicompost @ 1t ha^{-1}, T_{12}-75 per cent RDF + vermicompost@ 1t ha^{-1} + PSB + *Rhizobium* and T_{13}-RDF + *Rhizobium* + PSB. Observations on plant stand, plant height, dry matter accumulation per plant, number of pods per plant, number of seeds per pod, seed index, biological yield, seed yield, straw yield and harvest index were recorded.

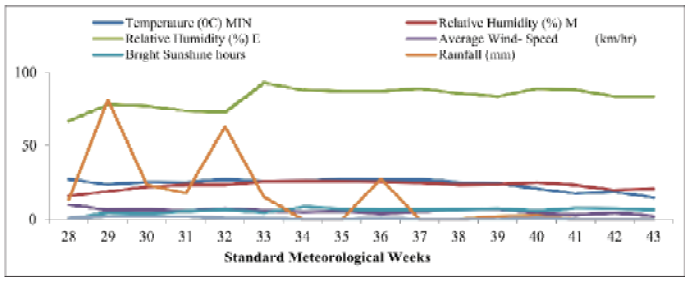

Fig. 1: Mean weekly meteorological data during experimentation

RESULTS AND DISCUSSION

The results revealed that plant height was not affected by different treatments at 20 DAS, It may be because all kinds of nutrients or nutrient combinations do not differ in their capacity to increase the plant height and all the efficiency is utilized for increasing the yield. Maximum plant height (55.16 & 68.50cm) was recorded in T_{12} (75% RDF + vermicompost @ 1t ha^{-1} + PSB +*Rhizobium*) 40 DAS and maturity, respectively. Similar results were observed by Patel *et al.,* (2016).

Maximum dry matter accumulation per plant was also recorded in T_{12} (75% RDF+vermicompost @ 1t ha^{-1} + PSB + *Rhizobium)*. This might be due to better growth and development obtained in this treatment. Singh *et al.* (2019) also reported that with the application of intergrated nutrients as 75 per cent RDF + PSB + VC + R*h* gave maximum dry matter per plant as compared to other treatments. Similar findings were reported by Rathore *et al.,* (2007), Sheoran *et al.,* (2008), Dhakal *et al.,* (2016) and Singh *et al.,* (2019). Plants with combined application of 75 per cent RDF + vermicompost @ 1t ha^{-1} + PSB + *Rhizobium* (T_{12}) had longer pods (8.21 cm) which was markedly at par with RDF + PSB + *Rhizobium* (T_{13}) Minimum pod length (5.8 cm) was under T_1 (control) which was statistically at par with application of *Rhizobium* + PSB (T_3).

Conjugation of different sources of nutrients like chemical fertilizer with organic source of nutrient and biofertilizer provide better environment for the growth and development, gave proper nourishment for plant growth resulting in maximum length of pods. Similar result was reported by Meena *et al.* (2015) and Singh *et al.,* (2019). Maximum number of pods per plant (13.61) were recorded in T_{12} (75% RDF + vermicompost @ 1t ha^{-1} + PSB + *Rhizobium)* and minimum (9.72) were found in control which was found statistically at par with vermicompost @ 2.5t ha^{-1} (T_2). The increase in the yield attribute might be due to increased supply of major nutrients, directly as well as by inoculating with biofertilizers. Vermicompost increases the carbon status, soil water holding capacity, availability of macronutrients as well as micronutrients which lead to sustainable crop production (Rajkhowa *et al.*, 2000). The increased yield attribute might be because of availability of almost all essential nutrients required by plants. All this might lead to more translocation which would ultimately result in increased number of pods. Synergistic effect of *Rhizobium* and PSB might also increase the number of pods. Similar findings were obtained by Patel *et al.* (2003), Yadav *et al.* (2004), Kumar and Kushwaha (2006), Yadav *et al.* (2007). The significant increase in the number of pods, which is an important yield attribute, with the integrated use of organic (Vermicompost @ 1t ha^{-1}), inorganic (75% RDF) fertilizers and biofertilizer (PSB and *Rhizobium*) was also observed by Kumpawat *et al.,* 2010.

Use of vermicompost @ 2.5t ha^{-1} (T_2) as well as integration of *Rhizobium* with PSB (T_3) showed significant increase in yield over control (T_1). Maximum grain yield (13.26 q ha^{-1}) was found in case of combined application of 75 per cent RDF + Vermicompost @ 1t ha^{-1} + PSB + *Rhizobium* (T_{12}). This might be due to the conjugation of chemical fertilizer with organic source of nutrient and biofertilizer usage which might have provided better environment and nourishment for plant growth resulting in maximum grain yield. Similar findings were reported in the studies of Amruta *et al.,* (2015), Choudhary *et al.,* (2003), Dhakal *et al.*(2016), Meena *et al.,* (2015), Patel *et al.,* (2013), Rajkhowa *et al.,* (2003), Reddy *et al.,* (2011), Saini *et al.,* (2014), Singh *et al.* (2019), Tyagi *et al.,* (2014)and Yakadri *et al.,* (2004). Minimum grain yield in case of control treatment could be attributed to the minimum values of all the yield attributing characters *i.e.* pods per plant, seeds per pod and seed index, which was also due to the fact that minimum biomass accumulation during vegetative growth phase which lead to less bearing and thus minimum grain yield. Similarly, maximum straw yield (26.52 q ha^{f}) and harvest index also was found in T_{12}. Results indicated that organic source can substitute up to 25 per cent of chemical fertilizers. Addition of organic manures along with chemical fertilizers significantly increased the yield over chemical fertilizers alone might be due to it provides favourable physico-chemical soil environment for microbes as well as plant. Maximum cost of cultivation (Rs

Table 1: Effect of various nutrients combinations on plant height, dry matter accumulation, no.of pods per plant, no.of seed per pod and seed index in mung bean

Treatment	Plant height (cm)			Dry matter accumulation g per plant^{-1}			Number of pods plant^{-1}	Number of seeds pod^{-1}	Seed Index(g)
	At 20 DAS	At 40 DAS	At Harvest	At 20 DAS	At 40 DAS	At Harvest			
T_1	22.13	41.46	52.46	5.59	15.62	35.68	9.72	6.93	3.94
T_2	22.06	52.40	59.73	6.83	17.27	36.46	10.70	6.97	3.96
T_3	22.76	47.76	56.76	6.77	16.94	36.88	10.25	6.95	3.92
T_4	22.13	47.80	54.46	7.23	17.42	39.37	10.81	7.17	4.01
T_5	22.16	47.50	60.50	7.25	17.86	39.58	10.97	7.22	4.02
T_6	23.20	50.70	59.90	8.94	18.87	44.23	11.85	7.36	4.40
T_7	22.63	46.30	56.63	8.39	17.94	42.00	11.44	7.22	4.14
T_8	22.80	52.43	60.10	8.42	18.39	42.12	11.46	7.23	4.27
T_9	22.90	46.80	58.13	8.55	18.55	42.25	11.49	7.26	4.27
T_{10}	22.86	45.20	56.86	8.64	18.6	42.46	11.63	7.29	4.28
T_{11}	23.36	46.23	55.70	9.01	18.94	45.60	12.03	7.34	4.53
T_{12}	23.56	55.16	68.50	9.13	22.42	48.61	13.61	7.49	4.78
T_{13}	23.50	54.20	66.20	9.12	19.95	43.81	12.91	7.42	4.63
S.E.(m)	0.54	1.71	1.76	0.84	0.84	1.51	0.49	0.35	0.02
C.D. at 5%	NS	5.04	5.19	NS	2.46	4.45	1.46	1.16	1.01

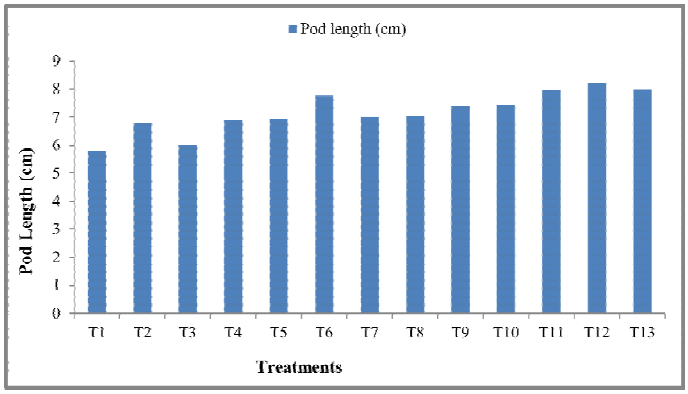

Fig. 2: Pod length as affected by different nutrient treatments

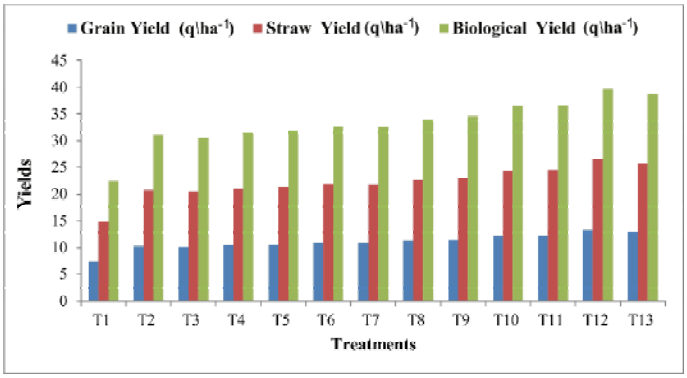

Fig. 3: Grain yield, straw yield and biological yield as effected by different nutrient treatments

Table 2: Cost of cultivation, gross monitory returns (Rs ha^{-1}), net monitory returns (Rs ha^{-1}) and benefit cost ratio (B-C ratio) under different treatments in mungbean

Treatment	Cost of cultivation (Rs ha^{-1})	Gross monitory returns (Rs ha^{-1})	Net monitory returns (Rs ha^{-1})	Benefit cost ratio (B-C)
T_1	17790	55204	37414	2.10
T_2	22790	76664	53874	2.36
T_3	17910	75480	57570	3.21
T_4	18790	77626	58836	3.13
T_5	19100	78662	59562	3.11
T_6	20540	80660	60120	2.92
T_7	18890	80216	61326	3.24
T_8	20790	83620	62830	3.02
T_9	20890	85248	64358	3.08
T_{10}	19200	89910	70710	3.68
T_{11}	21100	90428	69328	3.28
T_{12}	21200	98124	76929	3.62
T_{13}	20640	95219	74579	3.61

21200 ha^{-1}), net monetary returns (Rs 76929 ha^{-1}) and maximum BC ratio (3.62) were calculated in T_{12} (Table 2). This might be due to maximum grain and straw yield. Rajkhowa *et al.*, (2003) with the same application of organic and inorganic sources, observed increased economics of moongbean crop. Results of Yadav *et al.*, (2004) and Singh *et al.*, (2019) are also in conformity of the result of present study.

CONCLUSION

The study conclude that application of 75 per cent RDF + vermicompost @ 1t/ha^{-1} + PSB + *Rhizobium* in green gram cultivation is more productive and profitable.

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Effect of integrated nutrient management on growth, yield and economics of potato (*Solanum tuberosum* L.) in Jalandhar (Punjab)

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ABSTRACT

The field experiment laid out in randomized block design with three replications of ten different combinations of organic and chemical fertilizer i.e., (FYM, poultry manure, vermicompost) and the chemical fertilizers at different levels, using kufri Pukhraj variety of potato, showed that the growth parameters like percentage of plant emergence, percent tubers emerged, plant height at 30 and 75 DAP, number of haulms plant^{-1} (75 DAP), number of compound leaves plant^{-1} at 75 DAP as well as the yield attributing characters (ie., number of tubers plant^{-1}, tuber weight plant^{-1}, marketable yield plot^{-1}, and non-marketable yield plot^{-1} and total yield plot^{-1}) were significantly influenced by different treatment combination of FYM, NPK, poultry manure and vermicompost. However, the combination of poultry manure and the RDF applied @10t h^{-1} + 50 per cent R.D.F exhibited maximum weight of tuber plant^{-1}, marketable yield plot^{-1}, non marketable yield plot^{-1}, total yield plot^{-1}, gross income (Rs. 2,79.666), net income (Rs 163914) and B: C ratio (141), minimum weight of tubers plant-1, marketable yield plant^{-1}, the total yield gross income, net income and the Benefit: Cost and the lowest was recorded in T4 (vermi compost @ 20 t ha^{-1}).

Key words: Potato, INM, Vermicompost, Poultry Manure, FYM, Sustainable Agriculture

Potato (*Solanum tuberosum* L.) is one of the most popular and widely grown vegetable crops in the world belonging to solanaceae or nightshade family. This herbaceous crop is rich in dry matter, dietary fibre, quality protein, minerals, vitamins, carbohydrates and amino acids like leucine, tryptophan and isoleucine (Khurana and Naik, 2003). The annual world production of potato is 352,161,052 MT under 47,697,763.36 MH area (Anonymous, 2019). India ranks second in terms of potato production after China, where potato is cultivated in over 2.18 M ha^{-1} with annual production of 52.58 MT and productivity of 24.07 tonne ha^{-1} (Anonymous, 2019).

Major potato growing states in India are Madhya Pradesh, Uttar Pradesh, West Bengal, Haryana, Punjab, Bihar, Gujarat and Maharashtra. In Punjab, the area under potato cultivation is 86,000 hectares with a productivity of about 20 tons ha^{-1}. Of these 86,000 hectare land, 80,000 ha (93%) are located in Doaba region (Anonymous, 2019). It is Doaba ("land of two rivers") is the region located in between the Sutlej and Beas rivers, with the city of Jalandhar as a central point. Fertilizer is one of the most important inputs for increasing the productivity of crops and modern varieties of different crops (Ali et al., 2009). Chemical fertilizers are not only in limited supply but also very expensive in developing countries like India. Potato is one of the heavy feeders requiring relatively large quantities of fertilizers. However, continuous dependence on chemical fertilizer causes nutritional imbalance and adverse effects on physio-chemical and biological properties of the soil. Continuous application of heavy doses of chemical fertilizer without organic manure has led to a deterioration of soil health in terms of physical and chemical properties of soil, increased soil, water and air pollution. Integrated nutrient management (INM) is a better approach for supplying nutrition or food to the crop by including organic and inorganic sources of nutrients (Arora, 2008). Integration of organic manure and inorganic sources of nutrient is necessary for sustainable agriculture as it not only provides greater viability in production, but also helps in restoration and maintenance of soil fertility (Nambiar et al., 1998) as also as reduces the production costs. The application of organic manures like farm yard manure, vermi-compost and poultry manure etc, not only supply nutrients to the crop but also improve the physico-chemical properties of the soil and its water retention capacity. Farm yard manure (FYM) is one of the very popular forms of organic fertilizer.

The farm yard manure contains 0.4-0.5 per cent nitrogen, 0.2-0.3 per cent phosphorus and 0.4-0.5 per cent potassium and many other micro- nutrients (Yawalkar et al., 1996). The farm yard manure is the best organic manure but, it is required in bulk quantity to supply desired quantity of nitrogen. Poultry manure contains all 13 of the essential plant nutrients that are used by plants. These include nitrogen (N), phosphorus (P), potassium (K), calcium (Ca), magnesium (Mg), sulfur (S), manganese (Mn), copper (Cu), zinc (Zn), chlorine (Cl), boron (B), iron (Fe) and molybdenum

(Mo). Vermicompost used as fertilizer and soil conditioner (Munroe, 2007 and Rajesh et al., 2003) is responsible for the improvement of the physical properties of soil and supply vital plant nutrients (Smith et al., 2014). The growth and yield of potato largely depends on the soil and soil conditions which can be improved through the use of different organic fertilizers. Though single nutrient source may not supply the rest of required nutrients for the plant but integrated use of all sources is required for balanced plant nutrition (Arora, 2008). Keeping in view the importance of crop and need to standardize the doses of organic and in-organic manures for potato production in Doaba region of Punjab, the effect of integrated nutrient management on growth, yield and economics of potato cultivation under different treatment combinations was studied.

MATERIALS AND METHODS

The study was carried out at the experimental farm of Faculty of Agricultural Sciences, DAV University, Jalandhar in 2019 in Randomized Block Design. Ten treatment combinations of different organic and chemical fertilizer i.e., (FYM, poultry manure, vermicompost) with chemical fertilizer at different levels were used in the experiment by keeping three FYM levels (20 t ha^{-1}), (10 t ha^{-1} + 50% recommended dose of fertilizer), (5 t ha^{-1} + 50% recommended dose of fertilizer) , three vermicompost levels (20 t ha^{-1}), (10 t ha^{-1} + 50% recommended dose of fertilizer), (5 t ha^{-1} + 50% recommended dose of fertilizer), three poultry manure levels (20 t ha^{-1}), (10 t ha^{-1} + 50% recommended dose of fertilizer), (5 tha^{-1} + 50% recommended dose of fertilizer) and one control treatment (recommended dose of fertilizer) was also included in the present study. Kufri Pukhraj variety of potato was used in the study. The treatment effects on growth yield and economics were assessed and analyzed statistically.

Table1: Analysis of variance for different growth and yield contributing traits in potato

Characters	Mean sum of square treatments (MST)	Error
Plant emergence per cent	46.74*	18.54
Plant height (cm)	44.69*	0.96
Number of haulms plant-1	0.93*	0.05
Number of compound leaves plant-1	149.88*	2.55
Average number of tuber plant-1	5.74*	0.53
Average tuber weight plant-1	0.024*	0.002
Marketable yield plot-1	32.47*	0.50
Non-marketable yield plot-1	0.72*	0.22
Total yield plot-1	40.82*	1.82

RESULTS AND DISCUSSION

The analysis of variance for the different growth and yield contributing traits in potato

showed that all treatments were significant at 5 per cent significance level (table 1). The data on the growth parameters i.e. plant emergence per cent, plant height (cm), number of haulm $plant^{-1}$, number of compound leaves $plant^{-1}$, influenced by different levels of FYM, NPK, poultry manure and vermicompost furnished in (tables 2) revealed that per cent emergence of plants in all the treatments were statistically at par except in T_7 (poultry manure @10 h^{-1} + 50 per cent recommended dose of fertilizer) and T_{10} (poultry manure @5 h^{-1} + 50 per cent recommended dose of fertilizer). Minimum percent of tuber emergence (80.53) was observed in T_4 (vermicompst @20t h^{-1}) which was statistically at par with all the treatments except T_7 and T_{10}. Maximum percent of tubers emerged (92.74) was observed in T_7 (poultry manure @10 h^{-1} + 50 per cent recommended dose of fertilizer) which was statistically at par with T_9 (vermicompost @ 5t h^{-1} + 50 per cent of R.D.F) in which percent emergence of tubers was

Tables 2: Effect of integrated nutrient management on growth parameters as influenced by FYM, NPK, poultry manure and vermicompost

Treatments	Plant emergence (%)	Plant height	Number of halms $Plant^{-1}$	Number of compound leaves $plant^{-1}$
Recommended dose of fertilizer	84.96	40.84	5.46	46.73
FYM @ 20t h^{-1}	87.31	37.68	5.13	39.86
FYM @ 10t h^{-1} + 50 per cent R.D	83.96	38.34	5.06	43.40
Vermicompost @ 20th^{-1}	80.53	35.66	4.60	39.80
Vermicompost @ 10th^{-1} + 50 per cent R.D	87.21	36.60	5.06	43.46
Poultry @20t h^{-1}	87.73	43.49	5.80	52.13
Poultry @10t h^{-1} + 50 per cent R.D	92.74	48.16	6.46	59.46
FYM@5t h^{-1} +50 per cent R.D	84.07	40.33	5.20	50.33
Vermicompost @5th^{-1} + 50 per cent R.D	81.08	37.78	5.46	46.04
Poultry @5th^{-1} + 50 per cent R.D	91.06	43.40	6.13	58.73
CD	7.44	1.70	0.40	2.76
SE(m)	2.48	0.56	0.13	0.92

(91.07). The results are in line with the findings of Hossain *et al.* (2003), Chopra *et al*. (2006), Chhonkar *et al.* (2011), Verma *et al*. (2011) Patel (2013), and Banerjee *et al.* (2016) who also observed significant differences for plant emergence among different integrated nutrient management for plant emergence percent.

The plant height recorded at 75 DAP were significantly influenced by different treatment combinations. Data (table 2) revealed minimum plant height (35.60 cm) with treatment T_4 (vermicompost @20t h^{-1}) which was statistically at par with T_5 (vermicompost @10t h^{-1} + 50 per cent R.D.F) which produced plants with 36.60 cm height. Significantly higher plant height (48.16 cm) than in all other treatment combinations was reported in treatment T_7 (poultry manure @10 h^{-1} + 50% R.D.F) The increase in plant height can be due to the fact that higher nitrogen concentrate stimulates the assimilation of carbohydrates and protein which in turn enhanced cell division and formulation of more tissues enhancing vegetative growth of the plants. T_7 i.e poultry manure +10 ha^{-1}+50 per cent R.D.F increased the available nitrogen in the soil and resulted into increased plant height. These findings are in line with the findings of earlier researchers i.e. Santlal (2003), Hossain *et al*. (2003), Chettri *et al*. (2004), Chopra *et al*. (2006), Nag (2006), Nandekar *et al*. (2006), Pandey *et al*. (2007), Raghav *et al*. (2008), Solanke *et al*. (2009), Verma *et al*. (2011), Chhonkar *et al*. (2011), Sarkar *et al*. (2011), Verma *et al*. (2011), Dubey *et al*. (2012) and Patel (2013).

The data on number of shoots $plant^{-1}$ at 75 DAP were significantly influenced by levels of FYM, NPK, poultry manure and vermicompost. The result revealed significant difference among treatments for number of Haulms $plant^{-1}$. Minimum number of haulms $plant^{-1}$ (4.60) was observed in treatment T_4 (vermicompost @20t h^{-1}) which was statistically lower than other treatments. Maximum number of haulms $plant^{-1}$ (6.47) was observed in treatment T_7 (poultry manure @10t h^{-1} + 50% R.D.F) which was significantly at par with treatment T_{10} (poultry manure @5t h^{-1} + 50% R.D.F) which produced 6.13 haulms $plant^{-1}$. Maximum number of Haulms $plant^{-1}$ in the treatment combination producing maximum plant height reveals that this might be due to the fact the high dose of nitrogen would have stimulated the assimilation of carbohydrates and proteins. Barman *et al.* (2014) reported the beneficial response by the use of integrated levels of NPK, FYM, Vermicompost and neem Cake. Patel (2013) also recorded significantly higher number of shoots $plant^{-1}$ under treatment 150 per cent RDF in potato. Similarly, Kumar and Sharma (2002) and Raghav et al. (2008) reported the maximum number of shoots $plants^{-1}$ with application of FYM @ 10t ha^{-1} + 100 per cent RDF.

Significant differences for number of leaves $plant^{-1}$ was observed among different levels of FYM, NPK, poultry manure and vermicompost. Data (table 2) reveals minimum number of compound leaves (39.80) in treatment T_4 (vermicompost @20t h^{-1}) which was at par with T_2 (FYM @20t h^{-1}) producing (39.87) number of compound leaves $plant^{-1}$. Maximum number of compound leaves $plant^{-1}$ (59.47) was found in treatment T_7 (poultry manure @10t h^{-1} + 50% R.D.F) which was at par with treatment T_{10} (poultry manure @5 h^{-1} + 50% R.D.F) which produced (58.73) compound leaves $plant^{-1}$. These results corroborates with the finding of Nag (2006), Raghav *et al* (2008), Patel (2013) and Barman *et al*. (2014) who reported higher number of compound leaves per plant in differentiation integrated nutrient combinations.

The data on the yield parameters i.e. average number of tubers $plant^{-1}$, average tuber weight $plant^{-1}$, marketable yield $plot^{-1}$, non- marketable yield $plot^{-1}$ and total yield $plot^{-1}$,influenced by different levels of FYM, NPK, Poultry Manure and Vermicompost are furnished in Table 3. The number of tubers $plant^{-1}$ were significantly influenced by different levels of FYM, NPK, poultry manure and the vermicompost. Perusal of data clearly indicates that significantly higher number of tubers $plant^{-1}$ (10.60) in treatment T_7 (poultry manure @10t h^{-1} + 50% R.D.F) which was statistically at par with number of tuber $plant^{-1}$ observed in T_{10} (poultry manure @5t h^{-1} + 50% R.D.F) and T_6 (poultry manure @20t h^{-1}). Minimum number of tuber $plant^{-1}$ (6.27) was observed in treatment T_4 (vermicompost @20t h^{-1}) which was statistically at par with T_2 (FYM 20t/h^{-1}) producing (6.80) tubers per plant. Similar results were reported Imas and Bansal (2002), Das *et al.* (2004), Chetiri *et al.* (2005), Nag (2006), Bose *et al.* (2008), Raghav *et al*. (2008), Patel (2013), Amara and Mourad (2013), Chandrakar *et al*. (2013), Barman *et al.* (2014) and Das *et al*. (2004).

The tuber weight $plant^{-1}$ was significantly influenced by different levels of FYM, NPK, poultry manure and vermicompost. Data presented in (table 3) reveals that treatment T_7 (poultry manure @10t h^{-1} + 50% R.D.F) exhibited maximum weight of tuber $plant^{-1}$ (0.47 kg), which was significantly higher than all other treatments except T_{10} (poultry manure @5t h^{-1} + 50% R.D.F) producing 0.42 kg average weight of tuber $plant^{-1}$. However, minimum weight of tuber $plant^{-1}$ (0.18 kg) was observed in treatment T_4 (vermicompost @20t h^{-1}) which was significantly lower than all other treatments. Highest weight of tuber $plant^{-1}$ in treatment combination producing maximum number of leaves per plant may be linked to the increased total leaf area which in turn increased the amount of solar radiation intercepted and more assimilate might have been produced and assimilated in tubers. These findings corroborates with

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Tables 3: Effect of Integrated Nutrient Management on yield parameters as influenced by FYM, NPK, Poultry Manure and Vermicompost

Treatments	Average number of tubers plant^{-1}	Average tuber weight plant^{-1}	Marketable yield plot^{-1} (kg)	Non- marketable yield plot^{-1} (kg)	Total yield plot^{-1} (kg)
Recommended dose of fertilizer	8.53	0.30	7.16	0.64	7.77
FYM @ 20t h^{-1}	6.80	0.20	5.15	0.96	5.99
FYM @ 10t h^{-1}+50 per cent R.D	8.80	0.36	7.39	1.19	7.94
Vermicompost @20t h^{-1}	6.26	0.18	3.50	1.11	4.79
Vermicompost @ 10t h^{-1}+ 50 per cent R.D	8.06	0.34	7.27	1.61	8.85
Poultry @20t h^{-1}	9.53	0.39	11.22	1.70	13.20
Poultry @10t h^{-1}+ 50 per cent R.D	10.60	0.47	14.30	2.34	16.78
FYM@5t h^{-1} + 50 per cent R.D	7.86	0.33	6.17	1.01	7.57
Vermicompost @5t h^{-1} + 50per cent R.D	7.66	0.30	5.97	1.48	7.76
Poultry @5t h^{-1} + 50 per cent R.D	10.10	0.42	11.16	0.93	12.52
1.61CD	1.26	0.07	1.22	0.81	2.33
SE(m)	0.42	0.02	0.40	0.27	0.78

Table 4: Effect of integrated nutrient management on economics of potato

Treatments	Treatment cost	Cost of cultivation	Yield ha^{-1} (q ha^{-1})	Gross income in (Rs)	Net income (Rs)	B.C Ratio
Recommended dose of fertilizer	16,305	83,905	86.3	129,499.5	45594.5	0.54
FYM @ 20t h^{-1}	1,00,000	1,67,600	66.5	99,832.5	-67,7675	-0.40
FYM @ 10t h^{-1} +50 per cent R.D	58,152	1,25,752	88.2	132,333	6581	0.05
Vermicompost @20t h^{-1}	120,000	187,600	53.2	79,833	-107,767	-0.57
Vermicompost @ 10t h^{-1}+ 50 per cent R.D	68,152	135,752	98.3	147,499.5	11,747.5	0.09
Poultry @20t h^{-1}	80,000	147,600	146.6	219,999	72,399	0.49
Poultry @10t h^{-1} + 50 per cent R.D	48,152	115,752	186.4	279,666	163,914	1.41
FYM@5t h^{-1} + 50 per cent R.D	33,152	100,752	84.1	126,166.5	25,414.5	0.25
Vermicompost @5t h^{-1} + 50 per cent R.D	38,152	105,752	86.1	129,333	23,581	0.22
Poultry farm @5t h^{-1} + 50 per cent R.D	28,152	95,752	139.1	208,666.5	112,914.5	1.17

the finding of Taya *et al.* (1994), Pandey *et al.* (2007) and Zelalem *et al.* (2009). Other like Abou- Hussein *et al.* (2003), Santlal (2003), Kate *et al.* (2005), Singh and Rai (2007), Amara and Mourad (2013), Islam *et al.* (2013) also observed significant effect of different INM treatments on average tuber weight plant^{-1}.

Maximum marketable yield plot^{-1} (14.30kg) was observed in treatment T_7 (poultry manure @10t h^{-1} + 50% R.D.F) which was significantly higher than all other treatments. The T4 treatment recorded the lowest-marketable yield plant^{-1} (3.50 kg). The potato tuber yield plant^{-1} is directly dependent on the supply of N, P and K through excessive supply of nitrogen that may substantially delay leaf senescence leading to enhanced leaf area duration and probable increase in tuber yield (Mackerron and Helbronn, 1985). The other yield contributing traits like number of tubers per plant, weight of tuber per plant and the number of leaves plant^{-1} were highest in T_7 (poultry manure @10 t h^{-1} + 50% R.D.F). The increased number of leaves per plant increased the total leaf area available for photosynthesis and thus increased food assimilates. Saikia and Rajkhowa (1998), Chatterjee *et al.* (2014) also observed significant effect of INM on marketable yield plant^{-1} in potato.

The data on non-marketable yield plot^{-1} as influenced by different levels of FYM, NPK, poultry manure and vermicompost showed maximum non-marketable yield (2.34kg) with T_7 (poultry manure @10t h^{-1} + 50% R.D.F), which was statistically at par with T_6 (poultry manure @20t h^{-1}) and T_5 (Vermicompost @10t h^{-1} + 50% R.D.F) producing non-marketable tubers plot^{-1} to the tune of (1.70kg) and (1.61kg), respectively. Minimum non-marketable tubers (0.64kg) was found in T_1 (Chemical fertilizer R.D.F), which was statistically at par with T_2 (FYM @20t h^{-1}), T_3 (FYM @10t h^{-1} + 50% R.D.F), T_4 (vermicompost @20t h^{-1}), T_8 (FYM @5t/h + 50% R.D.F) and T_{10} (poultry manure 25t h^{-1} + 50% R.D.F) which produced (0.96kg), (1.19kg), (1.11kg), (1.01kg) and (0.93kg) non-marketable tubers plot^{-1}, respectively. The significant difference of integration of different rates of inorganic fertilizers with various organic manures indicates that the higher yield revealed by treatment T_7 (poultry manure @10t h^{-1} + 50% R.D.F) could be due to increased number of non-marketable tubers plot^{-1} which in turn increased the total yield.

The yield followed similar trend as observed with the marketable yield. The treatment with (poultry manure @10t h^{-1} + 50 per cent R.D.F (T_7) exhibited maximum yield $plot^{-1}$ (16.78kg) which was significantly higher than all other treatments. T_4 (vermicompost @20t h^{-1}) resulted in minimum tuber yield (4.79kg) which was significantly lower than all other treatments except T_2 (FYM @20t h^{-1}) with 5.99kg tuber yield. The result were in agreement of Kang *et al.* (2004), Tsegerro and Hammes (2005). These findings are in line with the findings of Zaman *et al.* (2011), Raghav et al (2008) and Bongkyoon*i* (2004)., Zabihi *et al.* (2010), Nag (2006) and Banjare (2012) and Patel (2013) who also reported higher marketable yield with the high nutrient supply.

The data on economics of different treatments (table 4) showed maximum gross income (Rs 279,666), net income (Rs. 163,914) and benefit: cost ratio (B:C ratio) (1.41) with T_7 (poultry manure @ 10 t ha^{-1} + 50% RDF) followed by T_{10} (poultry manure @ 5 t h^{-1} + 50% RDF) and T_6 (poultry manure @ 20 t ha^{-1}) providing a net income of (Rs 72,399). The lowest gross income, net income and Benefit: Cost ratio was recorded in control T_4 (vermicompost @ 20 t ha^{-1}) followed by T_2 (FYM @ 20t ha^{-1}). Higher benefit cost ratio for T_7 (1.41) and T_{10} (1.17) could be because of low inputs cost. The findings are in line with Jaipaul *et al*., (2011)

CONCLUSION

It was concluded that the use of organic manures (poultry manure @ 10 t ha^{-1} + 50 per cent of recommended dose of fertiliser proved highly effective in increasing the value of various parameters of potato growth, yield and the economics including the benefit cost ratio and therefore be employed in sustainable potato production.

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Influence of post flowering foliar nutrient application on yield and quality attributes of mango (*Mangifera indica* L.) cv. Alphonso under south Konkan conditions

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ABSTRACT

The experiment conducted at Mango Research Sub-Centre, Rameshwar, Tal. Deogad, Dist. Sindhudurg (Maharashtra) during the year 2015-16 to assess the influence of post flowering sprays of nutrients. viz; Urea (2%) NPK 19 : 19 : 19 – (1%), potassium nitrate (2%), potassium sulphate (0.5%) and monopotassium phosphate (0.5%) with an untreated control revealed highest fruit retention (8.05 per cent) with the potassium nitrate application against the lowest (93.85 per cent) registered in control. The fruit yield was also significantly maximum (127 fruits tree^{-1}, 29.13 kg tree^{-1} and 2.91 t ha^{-1}) with the potassium nitrate treatment registering an increase in yield by 38.57 per cent than the control. The maximum fruit weight (233.8 g) was recorded in control (T_6). The lowest intensity (10.0 per cent) was observed with potassium nitrate and potassium sulphate sprays.

Keywords: *Mango, Alphonso, foliar nutrition, fruit retention, yield*

Mango (*Mangifera indica* L.) is rightly known as 'National Fruit of India', owing to it's nutritional richness, unique taste and flavour as well as religious and medicinal importance. It is the third widely produced fruit crop of the tropics after banana and citrus. It belongs to family Anacardiaceae and originated from South East Asia, the Indo-Burma region, in the foothills of the Himalayas (Mukherjee, 1951). In the world, India is the leading mango producer with an area of 2.26 million ha and annual production of 19.69 million tonnes with productivity of 8.7 MT ha^{-1} (Anon., 2017). Konkan is the major and famous Alphonso mango producing region located on the west coast of Maharashtra. The hot and humid climate throughout the year with dry season from November to May in Konkan region is most ideal for Alphonso mango. Its virtual dominance in domestics and markets abroad because of its typical highly appreciable taste and flavor, sugar-acid blend and pleasant aroma, benefit growers in getting premium rates compared to other commercial mango varieties.

The Alphonso mango although is precious, it is poor yielder, has irregular bearing and is sensitive to climatic aberrations as well as physiological processes and fruiting hampering its production and productivity. At various stages of fruit growth and development (peanut, marble and egg stage), heavy fruit drop is a serious problem and is a limiting factor for increasing productivity in Alphonso mango. The fruit drop is as natural phenomenon which may occurs due to several factors like hormonal imbalance, nutrition, recurrent flowering, adverse weather, etc. The foliar sprays of nutrient reduce the fruit drop in most of the commercial fruit crops. In mango, the effectivity of foliar sprays of various nutrients to reduce the fruit drop was studied by several researchers and recommended for adoption by the mango growers. Keeping this in view the present investigation was undertaken.

MATERIAL AND METHODS

The experiment was conducted at Mango Research Sub-Centre, Rameshwar, Tal. Deogad, Dist. Sindhudurg (Maharashtra) during the year 2015-16 on 25 years old mango trees (cv. Alphonso). It was laid out in randomized block design with four replications and six treatments viz; Urea - 2% (T_1), NPK 19 : 19 : 19 – 1% (T_2), Potassium nitrate – 2% (T_3), Potassium sulphate – 0.5% (T_4), Monopotassium phosphate – 0.5% (T_5) and Control (T_6). Each treatment was given for two uniformly growing trees. First spray was given after fruit set (peanut stage of fruit development) and second at marble stage. The recommended cultural practices were followed in experimental trees block. The data on yield attributing characters like fruit set, fruit retention, fruit weight, number of fruits were recorded. The occurrence of spongy tissue, TSS, acidity in ripe fruit and shelf life of fruits were also recorded. The data were statically analyzed by the method described by Panse and Sukhatme (1985).

RESULTS AND DISCUSSION

The data on the fruit retention percentage at harvest presented in Table 1 revealed that the post flowering

nutrition with NPK grades significantly improved the retention percentage over control. The highest fruit retention (8.05 per cent) was observed in potassium nitrate treatment (T_3) whereas the lowest fruit retention 93.85 per cent) was registered in control (T_6). The rest of the treatments were at par among themselves. It also implied that the fruit drop was control in potassium nitrate and other nutrient sprays given to the mango trees rather than control. The role of potassium is known in increasing turgidity while nitrogen assists in transporting photoassimilates toward the sink (fruit) and as a result as a result of this combination, higher fruit retention was observed. These results are in the line earlier findings of Sudha *et al* (2012) and Sarker and Rahim (2013) in mango.

The data on the days required for fruit set indicated that the period required for fruit development was not altered by the post flowering nutritional sprays (Table 1). On an average, 90.2 days were taken for the harvesting of fruits from fruit set and it was in the range of 86.8 to 92.0 days. The days taken for fruit maturity is predominantly governed by the prevailing environmental conditions of the particular location and the degree days required to complete the heat units. The non-significant effect of different foliar sprays on number of days required for harvesting of Alphonso mango fruits was also reported by Bansode (2012) under Dapoli condition.

The data regarding yield as influenced by the different post flowering foliar nutrition are presented in Table 1. The yield of Alphonso mango, quantified in terms of number of fruits per tree and in kilograms, significantly increased due to the foliar nutrition during fruit development. The highest number of fruits (127 tree^{-1}) was harvested in treatment T_3 (Potassium nitrate) and it was at par with the rest of the nutrient treatments while least number of fruits (89.8 tree^{-1}) were obtained in control. Similarly, the fruit yield was significantly maximum (29.13 kg tree^{-1} and 2.91 t ha^{-1}) in potassium nitrate (T_3) treatment. It was at par with T_4 and T_2. Whereas, the lowest fruit yield (21.03 kg tree^{-1} and 2.10 t ha^{-1}) was in control.

The result revealed that foliar nutrition after flowering and during fruit development stage significantly reduced the fruit drop which eventually ensured better fruit yield. The yield data that the yield in T_3 treatment (Potassium nitrate) increased by 38.57 per cent over the control, followed by T4 (30.0 per cent), T_2 (28.57 per cent), while with the urea treatment, the yield improved by 18.57 per cent and with 0 : 52 : 34 treatment by 18.10 per cent (Table 1). The beneficial effect of supplementary nutrition in increasing the fruit yield might be due to the increased fruit retention. Besides this, potassium increases resistance of plants to various biotic and abiotic stresses which ultimately lend a hand to control

Table 1. Effect of post flowering foliar nutrition on yield and quality attributes of mango cv. Alphonso

Treatments		Fruit retention at harvest (%)	Days required to harvest from fruit set	No. of fruits tree^{-1}	Fruit Yield (kg tree^{-1})	Fruit Yield (t ha^{-1})	Yield improvement over control (%)
T_1	Urea (46:0:0) – 2 per cent	6.56^{b}	89.5	112.5^{a}	24.91^{abc}	2.49	18.57
T_2	NPK 19 : 19 : 19 – 1 per cent	6.91^{b}	91.8	124.0^{a}	26.98^{ab}	2.70	28.57
T_3	Potassium nitrate – 2 per cent % KNO_3 (13:0:45)	8.05^{a}	89.5	127.0^{a}	29.13^{a}	2.91	38.57
T_4	Potassium sulphate – 0.5 per cent KSO_4 (0 : 0 50)	6.89^{b}	86.8	119.0^{a}	27.25^{ab}	2.73	30.00
T_5	Monopotassium phosphate - 0.5 per cent (0-52-34)	6.64^{b}	91.8	111.5^{a}	24.80^{bc}	2.48	18.10
T_6	Control	3.85^{c}	92.0	89.8^{b}	21.03^{c}	2.10	-
Mean		6.48	90.2	113.96	25.68	2.57	-
S. Em ±		0.28	1.80	6.42	1.43	-	-
C. D. at 5%		0.81	NS	18.78	4.19	-	-
C. V. (%)		8.5	3.99	11.27	11.15	-	-

Average fruit weight (g)	Occurrence of spongy tissue (%)	T. S. S. (^{0}B)	Titratable acidity (%)	Shelf life (days)
222.0^{bc}	20.0 (26.57)*	19.80	0.34	13.5
217.4^{c}	13.8 (21.77)	19.90	0.35	14.0
229.1^{ab}	10.0 (18.43)	20.25	0.33	14.8
228.6^{ab}	10.0 (18.43)	19.90	0.33	14.5
228.7^{ab}	16.3 (23.77)	19.90	0.35	14.0
233.8^{a}	25.0 (30.00)	19.70	0.33	14.3
226.61	15.83 (22.98)	19.91	0.34	14.17
2.71	1.83	0.14	0.10	0.42
7.93	5.36	NS	NS	NS
2.39	15.93	1.36	5.15	5.91

(* Figures in parenthesis are the arcsine value)

the fruit drop. Nitrogen also has stimulating effect in translocation of photosynthetes towards the developing fruits. This finding confirms earlier work in mango done by Shinde *et al*. (2006), Sarker and Rahim (2013), Galande (2015) and Chaudhari (2016).

The data presented in Table 2 revealed that the fruit weight was significantly influenced by the foliar nutrition. The maximum fruit weight (233.8 g) was recorded in control (T_6) and it was at par with T_3, T_5 and T_4. The lowest fruit weight (217.4 g) was in T_2 (19 : 19 : 19). The highest fruit weight was found in control treatment which may be because of less number of fruits retained per trees which gained more food material. Bansode (2012) also observed the similar trend in mango.

The spongy tissue in Alphonso mango, a physiological disorder, may be caused due to several factors. The data presented in Table 2 revealed significant reduction in the spongy tissue intensity in the mango fruits due to post flowering foliar nutrition. The lowest intensity (10.0 per cent) was observed in T_3 and T_4 treatments and it was closely followed by T_2. The spongy tissue occurrence was highest in control (25.0 per cent) and it war at par with T_1. The potassium has role in major plant metabolic processes such as protein synthesis, enzyme activation, water uptake, transpiration, etc. which in some way helps in reduction of spongy tissue. The similar findings were also reported in mango by Shinde *et al*. (2009) and Malshe *et al*. (2018).

The total soluble solids (T.S.S.) and titratable acidity were not influenced significantly due to post flowering foliar nutrition. The mean values for T. S. S. and acidity were 19.91^{0}B and 0.34 per cent, respectively (Table 2). The post flowering foliar nutrition did not significantly influence the shelf life of mango fruit indicating range of 13.5 to 14.8 days with a mean value of 14.17 days. The similar findings were also noted by Bansode (2012) in Alphonso mango.

CONCLUSION

The present investigation revealed that the supplementary nutrition during the fruit development stage reduced the fruit drop and increased the yield of mango cv. Alphonso. The intensity of the spongy tissue could be reduced by nutrition with potassium containing water soluble fertilizers. The fruit yield was improved by taking only two sprays of nutrients. As the outcomes are only from one year experiment, the confirmation is essential by taking trials for two or three years at multy locations.

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Effect of drought stress on phenological and yield attributes in Wheat (*Triticum aestivum L.*)

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ABSTRACT

Investigations on drought stress treatment in terms of phenological and yield attributes carried out with wheat varieties (WH 1105 and WH 1025) under field conditions revealed that the number of days to heading, under drought stress conditions decreased by 5 days. The number of days to anthesis were also found less in drought conditions. It was 100 and 103 days in WH 1105 and WH 1025, respectively. The grain filling period was shortened in WH 1025. The drought-stressed varieties reached to maturity 5 days earlier compared to irrigated ones. The yield attributes such as grain number per spike, grain weight per spike (g), test weight (g), and grain yield per square meter (g/m ^{2}) under drought also decreased to a great extent. The Drought Susceptibility Index (DSI) calculated for two wheat varieties was 1.25 and 0.71 in WH 1105 and WH 1025, respectively. The results showed that the wheat variety WH 1025 registered better drought tolerance than WH 1105.

Agriculture is facing the greatest challenge to meet the food requirements of the ever-growing population in the world. There is intense pressure on the scientific community to further enhance the productivity of major cereal crops. Wheat is one of the major food crops that feed nearly half of the world's population (Dhakal 2021). According to the predicted trends, wheat production will be affected due to climate change (Aaron *et al.,* 2020). Scenarios of changing climate suggest that water-deficient conditions at the grain filling stage could reduce crop yields in arid and semi-arid regions in most parts of the world (Hafiz *et al.,* 2020). Wheat growing areas in India are frequently hit by moderate to severe droughts particularly when the crop is at the stage of anthesis. Less availability of water during drought periods forces plants to use limited moisture as efficiently as possible. High-temperature during extreme in of weather conditions associated with drought further aggravate the risk of drought (IPCC 2007). Significant changes are expected to occur with regard to wheat phenological parameters (Jessica *et al.,* 2020). The negative changes that occur in phenological traits considerably reduce the time of maturity of the crop and ultimately affecting the yield attributes (Hester and Alison 2021). Wheat plants, stressed for water at different stages of growth, mature earlier than the plants under normal conditions (Nisar *et al.* 2007). Drought adversely affects the crop developmental stages and shorten the grain filling period by decreasing potential yields (Brooks *et al.* 1982).

Shortening of days to heading, days to maturity, and grain filling period under high-temperature stress in wheat have been reported (Mahboob *et al.* 2005). The variability of climate and the frequent onset of droughts are responsible to cause a change in yield attributes such as number of spikes per square meter, test weight, the weight of grains per spike and biological yield were the most effective variables influencing grain yield and suggesting that high yield of wheat plants under drought conditions could be obtained by selecting suitable breeding materials (Leilah & Al-Khateeb, 2005). Previous investigators have also studied the impacts of drought stress on phenological parameters in highly controlled environments or pot culture experiments with no adequate data of crop performance under field conditions. Empirical studies on wheat in open airfields are scarce. Hence, the present study about the impacts of drought stress on various phenological parameters and yield attributes under field conditions in (varieties WH 1105 and WH 1025) was carried out. The aim of this experiment was to determine how drought stress treatment at the grain filling stage, influence phenological parameters and yield attributes.

Keywords : Wheat *(Triticum aestivum L.*), drought stress, phenological parameters and yield attributes.

MATERIALS AND METHODS

Seeds of two varieties of wheat *viz.* WH 1105 (drought sensitive) and WH 1025 (drought tolerant) were obtained from the wheat and barley section, Department of Genetics and Plant Breeding, College of Agriculture, CCSHAU, Hisar. Seeds were sown in micro plots on the university farm. Drought stress was created by giving pre-sown irrigation only for the micro plots designated for this purpose. Normal recommended irrigations were given for other micro plots. Various phenological parameters and yield attributes were studied.

Observations on different phonological traits, namely days to heading, days to anthesis, days to maturity, green filling duration and yield attributes such as grain number spike^{-1} grain weight spike^{-1}, (g) grain yield square meter^{-1} (g), and test weight (g) of (thousand grains) were recorded. The

drought susceptibility index (DSI) was calculated by the method of Fischer & Maurer (1978) on the basis of mean grain yield:

Where,

Yd= Mean yield under drought,

Yi= Mean yield under irrigated condition and

D= Environmental stress intensity

D = 1- (Mean yield of two varieties under drought/ mean yield of two varieties under irrigated conditions).

The soil samples of 75-100 g taken from both drought and irrigated fields at a depth of 15 cm by using a soil auger were stored in soil moisture boxes. Dry weight after drying in the oven at 105°C for 24 hours) of the samples was taken and the extent of soil moisture content was calculated according to Ge *et al.* (2012). The data was analyzed by analysis of variance for the randomized block design (RBD) using OPSTAT software available on www. http// hau. ernet. in home page (Sheoran, O.P.) where each observation was replicated thrice and CD at 5 per cent was calculated.

RESULTS AND DISCUSSION

Effect of drought stress on different phenological parameters

The data (table 1) showed that days to heading anthesis, grain filling duration and grain maturity under drought condition in both the wheat varieties was lower than under irrigated conditions. It was 96, 100, 39 and 137 days in WH 1105 and 96, 103, 40 and 141 days in WH 1025 under drought condition, respectively.

Effect of drought stress on yield attributes

Almost similar effect of drought stress on yield attributes were also observed. Data presented in table 2 shows that drought stress led to a decline in grain number over irrigated conditions. Maximum percent reduction was observed in WH 1105 (25.20) and minimum in WH 1025 (14.10). Contrary to this, the percentage decline in grain weight per spike was maximum (38.48%) in WH 1025 and 33.33 percent in WH 1105.

Considerable reduction in the test weight and grain yield on both wheat varieties under drought stress conditions was also noticed. The test weight noted for WH 1105 was 40.10 g and 36.10 g, while for WH 1025 it was 38.80 g and 36.60 g under irrigated and drought stress conditions respectively. The grain yield noted for WH 1105 was 735 and 628 g and for WH 1025 680 and 515 g under irrigated and drought stress conditions, respectively. The percent decrease in test weight was 9.97 and 5.67 in WH 1105 and WH 1025, respectively.

Drought susceptibility index

It is a very important parameter indicating the tolerance of wheat variety. The drought susceptible index is given in Table 2 DSI of WH 1025 was less than WH 1105. WH 1025 exhibited a DSI value of 0.71 while it was 1.25 in WH 1105.

Soil moisture content

The results presented in table 3 show deficit soil moisture content (%) under drought and irrigated fields. The soil moisture content ranged from 9.00 to 13.99 per cent and 11.60 to 18.07 percent under drought and irrigated fields

Table 1: Effect of drought stress on different phenological traits of wheat

Treatment	Days to heading		Days to anthesis		Grain filling duration		Days to maturity	
	WH 1105	WH 1025	WH 1105	WH 1025	WH 1105	WH 1025	WH 1105	WH 1025
Irrigated	102	101	104	108	39	43	142	146
Drought	96	96	100	103	39	40	137	141
CD at 5%	G = 2.4, E = 2.4, GXE=NS		G = 1.6, E = 1.7, GXE=NS		G = 0.9, E = 0.9, GXE= 1.3		G = 1.9, E = 1.9, GXE=NS	

Table 2: Effect of drought stress on yield attributes in wheat

Treatment	Grain number spike^{-1}		Grain weight spike^{-1}		Test weight (g)		Grain yield (g/m 2)		Drought susceptible index	
	WH 1105	WH 1025	WH 1105	WH 1025	WH 1105	WH 1025	WH 1105	WH 1025	WH 1105	WH 1025
Irrigated	56.20	62.40	2.13	3.17	40.10	38.80	735	680	1.25	0.71
Drought	42.00	53.60	1.42	1.95	36.10	36.60	628	515		
CD (5%)	G = 8.8, E = 8.8, GXE=NS		G = 0.5, E = 0.1, GXE=NS		G=0.1, E =0.3, GXE=0.04		G = 39.2, E = 39.2, GXE=55.4			

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Table 3: Soil moisture content in drought and irrigated fields

Days after sowing	Drought (%)	Irrigated (%)
0	11.60	11.60
40	12.53	17.60
70	13.99	18.07
90	10.80	14.22
120	9.00	14.06

from a day of sowing to 120 days after sowing. Soil moisture levels were relatively lower under drought fields as compared to irrigated fields.

The results on effect of drought stress on days to heading are well supported by the previous results of Hasan & Tacettin (2010) Mahboob *et al.* (2005) and Saadollah *et al.* (2005). They reported that the number of days to heading decreased under salt stress in durum wheat in wheat. Anthesis is the pivotal phenological stage for the effect of environmental conditions on yield components (Abdelali *et al.*, 2019). Drought stress decreased the number of days to reach to anthesis in both the wheat varieties. However, WH 1105 reached to anthesis early (100 days) as compared to WH 1025 (103 days) and the interaction between genotypes and environment was non-significant. Similar results were also given by Jacques *et al.* (2000) in winter wheat. Date of maturity was recorded when the peduncle was completely yellow for about half of the plants in the plot. Drought stress reduced the number of days to maturity in both the wheat varieties and WH 1105 reached maturity earlier (137 days) than WH 1025 (141 days). The observed results are in accordance with the previous results of Hasan & Tacettin (2010) who reported that drought stress decreased the number of days to maturity in wheat. Saadollah *et al.* (2005) also showed a reduction in the number of days to maturity in wheat under saline stress. Mahbood *et al.* (2005) also showed the reduction in days to maturity in wheat under temperature stress. Similarly, the number of days to heading was also reported by Nisar *et al.* (2007) and Pasquale *et al.* (2007). The results appreciate the survival pressure imposed by water scarcity on plant phenology where early maturity allows better management of drought period. Wheat varieties matured early indicating forced maturity due to drought stress. Generally, the early maturing plants would escape the deleterious effects of environmental and soil drought as compared to ones that mature late. Wheat plants stressed for water at different stages of growth matured earlier than plants under normal conditions. It is evident that earlier maturity of wheat may be caused by soil moisture deficit during seed development (Day & Intlap, 1970).

For most small crops, such as wheat, environmental potential yield is determined by duration in days of grain filling. Plant responses were determined from the day of drought implementation to plants reaching maturity and the grain filling duration was calculated (table 1). Drought stress reduced the number of days of grain filling in WH 1025 (40 days) while no change was observed in WH 1105 (39 days). The shortening of the grain filling duration with drought stress has previously been reported (Brooks *et al.* 1982). The results are consistent with the results of Gooding *et al.* (2003) who reported shortening of grain filling duration under drought stress in winter wheat. The results are also at par with the previous results of Shah & Paulsen (2003) who observed a reduction in grain filling duration under stress in wheat. Mahbood *et al.* (2005) also showed a reduction in grain filling period in wheat under temperature stress. Yashawanthakumar *et al.,* (2021) reported that yield is positively associated with grain filling duration under drought stress. Victoria *et al.,* (2020) had shown that terminal drought conditions shortened grain filling duration in wheat.

Drought is polymorphic stress and is considered as one of the most important factors limiting crop yields around the world (Hasan & Tacettin, 2010). As climate change leads to increasingly hotter and drier summers, the importance of drought constraints on yield and yield components has increased. The response of plants to water stress depends on several factors such as developmental stage, severity and duration of stress, and cultivar genetics (Beltrano & Marta, 2008; Qasin *et al.,* 2019). The yield of grain crops integrates two main components *viz*. grain number and grain weight. It is commonly accepted that the determination of these components scarcely overlaps the crop cycle in wheat (Slafer & Rawson, 1994). The data on the number of grains per spike is given in table 2. Drought stress markedly decreased the number of grains per spike in both the wheat varieties with a more percent reduction in WH 1105 as compared to WH 1025. The results are in accordance with the previous results of Hafiz *et al.* (2012) who reported that drought stress adversely affected the number of grains per spike and showed a significant decrease in wheat. Fatah *et al.* (2014) also showed similar results in wheat. The results are also at par with the reports of Ali *et al.* (2001); Naseri *et al.* (2010). Mirazaei *et al.* (2011) who showed a decrease in the number of grains per spike under drought stress in wheat. Parvaneh *et al.* (2014) also showed low grains per spike under drought stress as compared to irrigated conditions in wheat. Nasir *et al.* (2007) reported that the number of grains per spike was relatively low under drought stress in wheat. Mirza *et al.,* (2019) reported that drought stress had a higher reducing effect on grain yield and caused 45 percent reduction in mean value. Dwivedi *et al.,* 2018 showed an increase in the number of grains per spike when drought-stressed plants are treated with plant growth regulators.

Drought stress reduced the grain weight in both the wheat varieties (table 2) but more reduction was observed in WH 1025 (38.48 %) as compared to WH 1105 (33.33 %). The results are in accordance with the previous observations of Maqbool & Afzal, (1999) who reported that complete drought stress showed a significant reduction in the grain weight. Similarly, Gooding *et al.* (2003) showed a reduction in grain weight under water deficit conditions in wheat. Reduction in grain weight due to drought and variable response of the tested wheat genotypes to water stress has already been reported (Khan *et al.* 1993). A decrease in grain size may occur in part due to a decrease in endosperm cell number in response to water limitation (Nicolas *et al.* 1985). The maximum potential grain size is often related to the maximum amount of water per grain (Schnyder & Baum, 1992). In fact, grain weight is a function of the speed and duration of the grain formation period. The existence of environmental stresses such as drought stress especially at the grain formation period leads to a reduction in the amount of photosynthesis, the speed, and duration of the grain formation period, and finally, it leads to a reduction in grain weight (Parvaneh *et al.* 2014).

Test weight is greatly determined by climatic parameters, particularly drought stress during the final phase of grain filling. The data on test weight of both wheat varieties indicate that drought stress significantly reduced test weight, however, more reduction was observed in WH 1105 (9.9 %) than WH 1025 (5.6 %) (table 2). The results are corroborated with the results of Hasan & Tacettin (2010) who reported a significant reduction in the test weight of wheat under drought conditions as compared to well-watered conditions. Dwivedi *et al.*, 2018 showed that the test weight was increased when drought-stressed plants were treated with plant growth regulators. Drought stress impaired grain filling due to the loss in the partitioning of assimilates and supply of photosynthesis resulted in decreased test weight and ultimately grain yield per plant. The results are in harmony with those of Saadollah *et al.* (2005); Farooq *et al.* (2009); Fatah *et al.* (2014). Similarly, Ivanova *et al.* (2013) reported that the test weight of wheat varieties decreased under stress. Drought stress adversely affects the test weight in wheat (Nesmith & Ritchi, 1992; Hafiz *et al.* 2012). Gooding *et al.* (2003) in a study about intensity and duration of drought stress in wheat reported that drought stress reduced test weight by shortening the grain formation period. Wasaya *et al.*, 2021 reported that the highest test weight was observed in the Galaxy-2013 wheat genotype.

The grain yield of a genotype is the most integrative trait because it is influenced by all known and unknown factors (Araus *et al.* 2001). Drought stress at the grain formation stage leads to a reduction in grain yield. The data about grain yield per square meter is given in Table 4. Drought stress significantly reduced grain yield in both the wheat varieties however, more reduction was observed in WH 1025 (24.2 %) as compared to WH 1105 (14.5 %). The results are similar to the earlier results of Shokouh *et al.* (2015) who also reported that drought stress reduces grain yield in wheat. Foulkes *et al.* (2002) reported that moisture stress is ultimately reflected in depressed yield and the magnitude of yield depression is perhaps the most practiced measure of drought resistance of wheat plants (Reynolds *et al.* 2001; Foulkes *et al.* 2004). Nisar *et al.* (2007) also stated that wheat plants growing under drought stress conditions resulted in lower grain yields. Gooding *et al.* (2003) in a study conducted on the intensity and duration of applied drought stress reported that drought stress caused a reduction in grain yield of wheat. Wasaya *et al.*, 2021 reported that the Galaxy-2013 wheat genotype recorded the highest grain yield under severe drought conditions.

One important index to determine drought tolerance is the drought susceptibility index (DSI). It is a yield stability parameter that is based on how much reduction is realized under drought stress (Bilge & Mehmet, 2010). The stability of grain yield for each genotype is estimated by the drought susceptibility index (DSI), derived from the yield difference between stress and non-stress environments (Blum *et al.* 1989). Fischer & Maurer (1978); Langer *et al.* (1979) involved the use of DSI which characterizes yield stability between two environments. There are many reports in the literature about the use of DSI for identifying genotypes with yield stability in moisture-limited environments (Clarke *et al.* 1984; Bruckner & Frohberg, 1987; Bansal & Sinha, 1991; Ahmad *et al.* 2003). The data on the drought susceptibility index is given in Table 4. The drought susceptible index was more to WH 1105 (1.25) than WH 1025 (0.71). Genotypes with low DSI values (less than 1) can be considered as drought-resistant (Bruckner & Frohberg, 1987) because they exhibited smaller yield reductions under water stress compared with the well-watered condition. The results are at par with previous results of Golabadi *et al.* (2006) who showed lower DSI for wheat resistant genotypes. Some researchers found that cultivars that had the lowest DSI values were drought-resistant than cultivars with the highest DSI values (Zarea-Fizabady & Ghodsi, 2004; Golabadi *et al.* 2006; Maciej *et al.*, 2019).

The data presented in table 3 indicate the soil moisture content of experimental fields. Soil moisture content in drought fields considerably decreased to as low as 9.0 percent at 28th DAA where no irrigation was given after sowing.

Journal of Eco-friendly Agriculture **17**(1) 2022
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However, the irrigated field maintained adequate soil moisture throughout the experimental period. The adverse effects of soil moisture deficit in drought fields on wheat varieties were reflected among various parameters studied. Wheat variety WH1025 performed better under drought fields as compared to WH 1105. The presented results are in accordance with previous results of Ge *et al.* (2012) who showed a significant reduction of soil moisture content under drought in wheat. Similar results were also reported by various workers (Hafiz *et al.* 2012; Farzad *et al.* 2013; Ivanova *et al.* 2013).

CONCLUSIONS

Considerable decrease in grain yield of both wheat varieties was observed under stress conditions. The wheat variety WH 1025 recorded comparatively higher grain yield than the WH 1105. The drought susceptibility index was higher in WH 1105 than WH 1025. Grain number per spike and test weight also decreased under drought. Grain number per spike and test weight reduced under drought stress in both the wheat varieties. However, more per cent reduction was observed in WH 1105. Grain weight decreased under drought stress, but higher percent reduction was observed in WH 1025.

Phenological parameters and yield attributes under drought conditions at different wheat grown areas across different climatic zones are to be further studied to assess the extent of effect caused and use selected varieties as useful genetic resources for further development of better drought-tolerant genotypes.

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Enhancement of cashew yield through foliar feeding of nutrients

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ABSTRACT

The experiment, conducted to access the effect of water soluble nutrients on growth and yield of cashew under *rainfed* conditions at farmer's field during the years 2017-18 and 2019-20 with nine treatments *viz*., control (no spray), urea (2%), 0:52:34 (1% and 2%), 13:0:45 (1% and 2%), 19:19:19 (1% and 2%), using 6 years old trees of cashew cv. Vengurla-4 planted at 7m x 7m spacing, showed that three foliar applications of nutrients at 19:19:19 @ 2 per cent recorded the highest sex ratio (0.24), nut weight (8.50g) and yield (8.58 kg tree^{-1} and 1.75t ha^{-1}) and appeared best for cashew yield enhancement.

Keywords: Cashew, flushing, flowering, panicle, rainfed, water soluble nutrients

Cashew is one of the most important dollar earning crops of Konkan region of Maharashtra which is being widely cultivated on an area of 1.91 lakh ha with production of 2.69 lakh MT and productivity is 1367 kg ha^{-1}. Maharashtra state ranks first in area, production and productivity of cashew in the country (Anon., 2017). However, there is still a good scope for improvement in its productivity.

Cashew is grown in heavy rainfall area of Konkan especially on hill slopes. Most of the applied nutrients get leached leading to low availability of nutrients during the critical stages and adversely affecting the growth, flowering, fruit set and yield of cashew. Therefore, foliar feeding of nutrient at different phenological stages (critical demand) may play a major role in increasing the fruit set, retention and overall production and quality of cashew by optimum uptake of nutrient with minimum losses.

Foliar sprays are an excellent supplement to soil application as they provide rapid absorption of nutrients for immediate utilization in metabolic activities beside ensuring economization of fertilizer use. Keeping in view a meager availability of information on the use of water soluble fertilizers in cashew in India and more particularly Konkan agro-climatic conditions, the present trial at farmer's field was taken up.

MATERIALS AND METHODS

The experiment was conducted by Regional Fruit Research Station, Vengurla, Dist. Sindhudurg, (Dr. B. S. Konan Krishi Vidyapeeth, Dapoli, Dist. Ratnagiri) Maharashtra at the field of Shri. Santosh Satardekar, Tendoli, Tal. Kudal, Dist. Sindhudurg during the years 2017-18 and 2019-20 under *rainfed* conditions. Nine treatments *viz*., control (no spray), urea (2%), 0:52:34 (1% and 2%), 13:0:45 (1% and 2%), 19:19:19 (1% and 2%) and water spray under randomized block design with three replications were evaluated. Uniform six years old plantations of cashew variety Vengurla-4, planted at 7m x 7m distance, were selected for experimentation. The nutrients were sprayed at flushing, flowering and fruit setting stages. Uniform recommended package of practices including recommended fertilizers doses i.e. 4 pots of FYM, 1000 g N, 250g P_2O_5 and 250 g K_2O per tree were applied during August to all the treatments including control and plant protection schedule were followed. Observations on flowering, fruiting and yield attributes of cashew were recorded at appropriate stages. The data collected for all the attributes were subjected to the statistical analysis for Randomized Block Design (RBD) as suggested by Panse and Sukhatme (1985).

RESULTS AND DISCUSSION

Effect of water soluble nutrients on flowering attributes of cashew

The three years as well as pooled data on effect of application of water soluble nutrients on flowering attributes i.e. flowering duration (days), number of laterals/m^2, number of panicles/m^2 and sex ratio of cashew presented in table 1 and table 2. The data in table 1 revealed that the foliar feeding of nutrients did not significantly influence the flowering duration (days), number of laterals m^2 and number of panicles m^2 during the year 2017-18 and 2019-20. However, different treatments significantly influenced the flowering

duration (days) during the year 2018-19, number of laterals/ m^2 during the year 2019-20 and sex ratio during the year 2017-18, 2018-19.

Minimum flowering duration (89.67 days) was observed with the application of urea @ 2 per cent and it was at par with in T_6 (90.33 days), T_8 (91.00 days), T_7, T_9, and T_5, (92.67 days). Maximum number of laterals/m^2 observed in T_6 (32.00/m^2) was at par with in T_5 (30.00/m^2), T_7 (29.33/ m^2), T_2 (28.67/m^2), T_3& T_8 (27.67/m^2) during the year 2019-20. The highest sex ratio of 0.24 was recorded in treatment T_8 (19:19:19 @ 2 %). However, it was at par with the treatments T_5 & T_9 (0.23) and T_1, T_3 and T_6 (0.22) during the year 2017-18. The T_8 treatment also recorded highest sex ratio of 0.20 during the year 2019-20 and was at par with T_7(0.19), T_2 & T_5 (0.18). The pooled data revealed significantly highest sex ratio of 0.24 with foliar application of 19:19:19 @ 2 per cent.

The results showed that the treatment 19:19:19 (2%) appeared the best with respect to flowering attributes, particularly the sex ratio. This may be due to higher C/N ratio as well as higher auxin level within the flowering shoots of treated trees. The results are in close conformity with Gawankar (2010) who also reported that 19:19:19 @ 2 per cent spray gave significantly higher sex ratio than the control.

Effect of water soluble nutrients on fruiting and yield attributes of cashew

The data on fruiting parameters i.e. fruit set/m^2 and number of nuts per panicle presented in table 2 showed that the foliar application of water soluble nutrients significantly altered the fruit set/m^2 during the year 2018-19 and number of nuts per panicle during the year 2017-18 & 2018-19. However, non-significant results for rest of the years and also in pooled study were obtained. The highest number of fruit set/m^2 (29.33) was recorded in t_2 treatment (urea @ 2%). While, significantly the highest number of nuts panicle^{-1} (5.83 & 9.87) was recorded during the year 2017-18 and 2018-19, respectively.

The data on yield attributes such as nut weight (g), apple weight (g) and shelling are presented in table 3. Different treatments significantly influenced the nut weight (g) and apple weight (g) during the year 2017-18. However, the results obtained during the year 2018-19 and, 2019-20 were non-significant. The highest nut weight (9.40g) recorded in T_8 (19:19:19 @ 2%) was at par with T_6 (8.97g), T_9 (8.57g), T_5 (8.53g) and T_1 (8.50g) during the year 2017-18. During the same year, the highest apple weight (100.0g) noted in T_5 (13:0:45 @ 1%) was at par with treatment T_9 (95.83g), T_8 (92.50g), T_4 (89.33), T_1 & T_6 (85.00g).

The data on effect of foliar application of nutrients on yield of cashew cv. Vengurla-4 (kg tree^{-1} & yield t ha^{-1}) are presented in table 4. The data revealed highest yield kg tree^{-1} (10.71, 9.57 and 5.47) and t ha^{-1} (2.18, 1.95 & 1.11) with application of 19:19:19 @ 2 per cent (T_8) during the year 2017-18, 2018-19 and 2019-20, respectively. The low yield during the year 2019-20 was due to cold and high dews prevailed during fruit set stage. The three years pooled data also revealed significantly highest mean pooled yield (8.58 kg tree^{-1} & 1.75 t ha^{-1}) with 19:19:19 @ 2% (T_8) and was at par with treatments T_7 - 19:19:19 @ 1 per cent (7.59 kg tree^{-1} & 1.55 t ha^{-1}). The lowest mean pooled yield of 4.75 kg tree^{-1} and 0.97 t ha^{-1} was recorded under T_1 control (no spray) during both the years.

Foliar feeding of 19:19:19 @ 2 per cent registered 44.57 per cent increment of mean pooled yield (t ha^{-1}) over control during both the years, respectively and was found best in enhancing the cashew yield. This may be because the treatment was applied at flushing, flowering and fruit set

Table 1: Effect of foliar application of nutrients on flowering in cashew during the year 2017-18 to 2019-20

	Treatment	Flowering duration (days)				No. of laterals/m^2				No. of panicles/m^2			
		2017-18	2018-19	2019-20	Pooled Mean	2017-18	2018-19	2019-20	Pooled Mean	2017-18	2018-19	2019-20	Pooled Mean
T_1	Control (No spray)	93.33	**94.67**	94.33	94.11	29.67	25.33	25.67	26.89	16.00	16.67	16.33	16.33
T_2	Urea 2%	92.33	89.67	**89.67**	90.56	28.33	25.33	**28.67**	27.44	17.00	15.67	**22.33**	18.33
T_3	0:52:34 (1%)	87.00	**95.00**	94.67	92.22	30.00	26.33	**27.67**	28.000	19.33	15.33	19.00	17.89
T_4	0:52:34 (2%)	89.33	**94.00**	**95.67**	93.00	23.33	26.67	23.67	24.56	14.67	15.33	**16.00**	15.33
T_5	13:0:45 (1%)	95.33	**92.67**	95.00	94.33	25.67	26.00	**30.00**	27.22	16.00	15.00	19.67	16.89
T_6	13:0:45 (2%)	90.67	90.33	91.67	90.89	26.00	26.67	**32.00**	28.22	16.00	17.00	**22.33**	18.44
T_7	19:19:19 (1%)	93.67	**92.67**	93.33	93.22	32.67	26.33	**29.33**	29.44	21.33	14.67	20.33	18.78
T_8	19:19:19 (2%)	86.00	91.00	91.67	89.56	27.00	26.00	**27.67**	26.89	17.00	17.00	17.67	17.22
T_9	Water spray	91.33	**92.67**	90.67	91.56	30.00	24.33	26.67	27.00	19.00	16.00	20.00	18.33
	SEm±	2.82	1.16	2.24	1.26	2.28	1.79	1.49	1.26	1.61	1.15	1.55	1.04
	CD @ 5%	N.S	3.47	N.S.	N.S.	N.S	N.S.	4.47	N.S.	N.S	N.S.	N.S.	N.S.

Table 2: Effect of foliar application of nutrients on flowering and fruiting in cashew during the year 2017-18 to 2019-20

Treatment	Sex ratio				Fruit set/m²				No. of nuts panicle⁻¹			
	2017-18	2018-19	2019-20	Pooled Mean	2017-18	2018-19	2019-20	Pooled Mean	2017-18	2018-19	2019-20	Pooled Mean
T_1 Control (No spray)	**0.22**	0.20	0.17	0.20	21.00	19.67	13.00	17.89	3.37	3.43	**3.13**	3.31
T_2 Urea 2%	0.21	0.20	**0.18**	0.20	16.33	**29.33**	13.00	19.55	2.93	5.77	3.30	4.00
T_3 0:52:34 (1%)	**0.22**	0.20	0.17	0.20	21.33	19.00	14.00	18.11	2.67	5.03	3.43	3.71
T_4 0:52:34 (2%)	0.20	0.22	0.17	0.20	17.67	19.67	14.67	17.34	2.83	6.27	3.50	4.20
T_5 13:0:45 (1%)	**0.23**	0.22	**0.18**	0.21	16.00	23.66	**12.67**	17.44	2.10	7.10	3.27	4.16
T_6 13:0:45 (2%)	**0.22**	0.22	0.17	0.20	13.00	16.67	13.33	14.33	1.97	3.87	3.27	3.04
T_7 19:19:19 (1%)	0.23	0.21	**0.19**	0.21	20.00	17.00	13.67	16.89	3.27	4.60	3.43	3.77
T_8 19:19:19 (2%)	**0.24**	0.23	**0.24**	**0.24**	27.67	23.00	**15.00**	21.89	**5.83**	**9.87**	**3.67**	6.46
T_9 Water spray	**0.23**	0.22	0.17	0.21	15.67	14.33	14.33	14.78	3.10	3.20	3.43	3.24
SEm±	0.005	0.008	0.006	0.007	3.03	1.07	1.60	2.00	0.41	0.83	0.38	0.67
CD @ 5%	0.02	N.S.	0.02	0.02	N.S	3.21	N.S.	N.S.	1.24	2.49	N.S.	N.S.

Table 3: Effect of foliar application of nutrients on yield parameters in cashew during the year 2017-18 to 2019-20

Treatment	Nut wt. (g)				Apple wt. (g)				Shelling (%)			
	2017-18	2018-19	2019-20	Pooled Mean	2017-18	2018-19	2019-20	ooled mea	2017-18	2018-19	2019-20	Pooled Mean
T_1 Control (No spray)	**8.50**	7.23	7.37	7.70	**85.00**	110.00	82.73	92.58	29.00	30.33	**27.50**	28.94
T_2 Urea 2%	8.23	6.90	**8.30**	7.81	74.17	100.00	87.33	87.17	29.93	29.67	27.97	29.19
T_3 0:52:34 (1%)	7.60	7.30	**7.13**	7.34	78.33	93.33	81.00	84.22	30.17	30.67	28.93	29.92
T_4 0:52:34 (2%)	8.23	8.00	7.50	7.91	**89.33**	76.67	81.77	82.59	29.27	30.00	28.03	29.10
T_5 13:0:45 (1%)	**8.53**	7.20	7.77	7.83	**100.00**	103.33	87.77	97.03	29.93	29.67	29.73	29.78
T_6 13:0:45 (2%)	**8.97**	6.20	7.27	7.48	**85.00**	100.00	81.10	88.70	28.60	30.67	28.03	29.10
T_7 19:19:19 (1%)	7.80	6.50	7.17	7.16	70.00	80.00	84.33	78.11	29.67	30.00	29.70	29.79
T_8 19:19:19 (2%)	**9.40**	8.17	7.93	8.50	**92.50**	73.33	**93.83**	86.55	30.00	31.17	**30.20**	30.46
T_9 Water spray	**8.57**	7.70	7.47	7.91	**95.83**	100.00	**77.33**	91.05	30.40	29.83	28.10	29.44
SEm±	0.32	0.60	0.32	0.27	5.38	13.57	3.59	5.76	0.55	0.52	0.82	0.37
CD @ 5%	0.96	N.S.	N.S.	N.S.	16.11	N.S.	N.S.	N.S.	N.S	N.S.	N.S.	N.S.

Table 4: Effect of foliar application of nutrients on yield of cashew during the year 2017-18 to 2019-20

Treatment	Yield (kg tree⁻¹)				Yield (t ha⁻¹)			
	2017-18	2018-19	2019-20	Pooled Mean	2017-18	2018-19	2019-20	Pooled Mean
T_1 Control (No spray)	5.99	5.49	2.77	4.75	1.22	1.12	0.56	0.97
T_2 Urea 2%	7.31	6.17	3.40	5.63	1.49	1.26	0.69	1.15
T_3 0:52:34 (1%)	5.99	7.12	3.08	5.40	1.22	1.45	0.63	1.10
T_4 0:52:34 (2%)	7.61	6.90	4.19	6.23	1.55	1.41	0.86	1.27
T_5 13:0:45 (1%)	7.05	6.94	**4.77**	6.25	1.44	1.42	**0.97**	1.28
T_6 13:0:45 (2%)	7.05	**8.12**	4.35	6.51	1.44	**1.65**	**0.89**	1.33
T_7 19:19:19 (1%)	**8.99**	**8.94**	**4.83**	**7.59**	**1.83**	**1.82**	**0.99**	**1.55**
T_8 19:19:19 (2%)	**10.71**	**9.57**	**5.47**	**8.58**	**2.18**	**1.95**	**1.11**	**1.75**
T_9 Water spray	**8.76**	**8.38**	3.96	7.03	**1.79**	**1.71**	0.81	1.44
SEm±	0.76	0.61	0.37	0.36	1.16	0.12	0.08	0.07
CD @ 5%	2.28	1.83	1.10	1.09	0.47	0.37	0.23	0.22

stages that might have produced pronounced effect of all physiological processes.

In large number of horticultural crops, foliar spray of various nutrients in the form of either nutrients from the chemical substances or from water soluble fertilizers have played a vital role in increasing growth, flowering and yield attributes. Gawankar *et al.* (2010) under Konkan conditions recorded significantly the higher fruit set, retention, yield and shelling per cent in cashew with 19:19:19 (2%) sprays. Likewise, Ghosh (1990) observed up to 2-3 folds increase in nut production and shelling percentage with highest level of N. Harishu Kumar and Sreedharan (1988) observed improvement in shelling (%) with NPK recorded Chaurasia *et al.* (2005) and Premsekhar and Rajashree (2009) significantly highest fruit set and yield with 19:19:19 by in tomato; which are in conformity with the present study.

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The increase in yield due to spraying of water soluble fertilizer may be due to absorption of nutrients and water resulting in more photosynthesis and increased nutrients accumulation in the fruits. Moreover, foliar feeding of water soluble nutrients directly to the metabolite sites considerably enhanced fruit yield by increasing number of nut, size and weight. In present findings, the beneficial effect of 19:19:19 (2% and 1%) in increasing nut yield may be due to an increase of sex ratio, number of nuts per panicle, retention of nuts up to harvest and nut weight. The optimum supply of nutrients to the bearing mango trees help in retaining more number of fruits (Singh, 1972; Sharma *et al*., 1990 in mango). The macronutrient, nitrogen, is the most widely needed fertilizer element in cashew. Nitrogen is used by plants to synthesize amino acids and nucleic acids that are necessary for all functions of the plant. Nitrogen application may increase the supply of some hormones to the fruit that tend to reduce abscission, probably auxin (Addicot, 1970). Size of fruit and yield of plant is the cumulative effect of various attributes as affected by macronutrients through higher rate of cell division and enlargement, photosynthesis and increase in enzymatic activities as well as biosynthesis of hormones especially auxin which plays a vital role in physiology of growth and development. Further, NPK application i.e. 19:19:19 fertilizer might have enhanced the translocation of metabolites from source (leaf) to sink (fruit) and increased accumulation of dry matter within the fruits resulting into higher yield. Accumulation and partitioning of biochemical constituent's *viz*., carbohydrates, nitrogen, starch, phenol and chlorophyll in cashew mature shoots and reproductive flushes was observed to be taking place in a favourable direction towards increasing nut set and yield in plants that received this particular treatment (Pushpalatha *et al*., 2000).

The increase in yield in water spray treatment (T_9) might be due to availability of water to cashew tree at the critical stage and helping in readily availability of major nutrients to cashew plant. Present findings are in conformity with results reported by Gawankar *et al*. (2010).

CONCLUSION

It was concluded that the foliar spray of 19:19:19 @ 2% (T_8) recording highest sex ratio, number of nuts per panicles, nut weight and raw nut yield of cv. Vengurla-4 (kg tree^{-1} and t ha^{-1}) appeared as the best treatment for cashew yield enhancement. The treatment registered 44.57 per cent increase in mean pooled yield over control.

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Evaluation of pigeon-pea based intercropping systems under irrigated conditions in coastal Odisha

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ABSTRACT

The field experiment conducted for over two years (2016-17 and 2017-18) at the Centre for Pulses Research (OUAT), Berhampur-761001, under irrigated *rabi* conditions in coastal Odisha testing six intercropping combinations of pigeon pea (cv. manak) with three vegetables (yam bean (safed hyb. sakalu), french bean (falguni latika) and cowpea (kashi kanchan) in two proportions (1:1 ; 2:2) and 4 sole crops revealed that intercropping combinations recorded higher pigeon pea seed equivalent yield than the respective sole crops. Performance of paired row sowing was found superior with 38.24 g ha^{-1} of pigeon pea seed equivalent yield than with single row intercropping (32.71 q ha^{-1}). Among the three intercrops, the french bean system was found most remunerative with 41.13 q ha^{-1} pigeon pea seed equivalent yield followed by yam bean (37.84 q ha^{-1}), but superior to cowpea systems (27.46 q ha^{-1}). Maximum pigeon pea seed equivalent yield (44.09 q ha^{-1}), net return (Rs. 2,31,473/-) and B:C ratio (3.99) were recorded with pigeon pea: french bean =2:2 proportion. This system also registered maximum land equivalent ratio (1.62), monetary advantage index (Rs. 74,176 ha^{-1}) and system relative crowding coefficient (9.86). Aggressivity and competition ratio indicated that french bean and cowpea were dominant on pigeon pea .

Key words: Aggressivity, B:C ratio, land equivalent ratio, monetary advantage index, net return, pigeon pea seed equivalent yield, relative crowding coefficient.

The rapidly rising food needs due to mounting population pressure and progressively shrinking per capita agricultural land availability warrants the intensification of the cropping system in the country. Pulses can play an important role in sustaining intensive agriculture by improving physical, chemical and biological properties of soil. Being nutrient efficient and sustainable, these are considered excellent crops for diversification in cropping systems. Pigeon pea (*Cajanus cajan* L. Millsp) is one of the most ancient and versatile grain legume crop in India with 90 per cent of global production and grown across the country under diverse agro-ecosystem. Production and productivity of this nutrient trove pulse is constrained due to biotic and abiotic stresses during *kharif*. Pigeon pea always face challenges from natural calamities, aberrant weather condition, severe attack of pod borer complex and competitive remunerative rainfed crops such as hybrid maize, sweet corn, cowpea etc. during *kharif* season. Need for horizontal expansion of pigeon pea encouraged for searching new niche. Pigeon pea can be grown as *rabi* crop in areas with mild and short winter (Roy Sharma et. al. ,1980). Puste and Jana (1990) recommended september sowing of pigeon pea in West Bengal. Venugopal and Rao (1999) advised for september sowing of rabi pigeon pea in Andhra Pradesh. Panda *et.al.* (2003) studied on the effect of NK fertilization on performance of yam bean-pigeon pea intercropping system during pre-*rabi* and its residual effect on succeeding mungbean in West Bengal. Pigeon pea can successfully be grown during *rabi* in coastal Odisha with 45X 10cm planting geometry (Panda *et al*. 2019). Pigeon pea genotypes such as laxmi, manak and CORG-9701 were found suitable for *rabi* sowing during november 1st fortnight in east and south eastern coastal plain zone of Odisha (Panda *et al*. 2019). Babu and Kalra (1989) studied nutrient management of *rabi* pigeon pea in Maharastra. Kanwar (1981) indicated the importance of early maturing pigeon pea for post-monsoon sowing. Mahalakshmi *et. al*. (2011) reported the positive response of *rabi* pigeon pea to drip irrigation. Behera *et al*. (1996) studied on prospects of vegetable crops in pigeon pea based intercropping system in rainfed lateritic soil of Odisha . Dubey *et al.* (1991) studied on intercropping in short duration pigeon pea. The development of cropping system adapted to climate change and meeting the nutritional and economic requirements of the local populations. In this context promotion of local cropping system firmly rooted in food and social habit of populations is recommended (Altieri, 1999). Intercropping systems for sustainable tropical agriculture is expected to withstand a wide range of ecological and climatic factors (Doubi *et al*., 2016). However, it is well established that different species growing together in the same place compete for nutrients, water and light (Ghose, 2004). Hence, effort has been made for evaluation of pigeon pea based intercropping systems under irrigated *rabi* condition in coastal Odisha. The trial was targeted to the vast alluvial up-medium land basin of the river Rushikulya and similar agro-ecological situations in east and south eastern coastal plain zone of Odisha after harvest of early duration paddy and finger millet grown during *kharif*.

MATERIALS AND METHODS

The field experiment was conducted over two consecutive years (2016-17 & 2017-18) at the Centre for Pulses Research, OUAT, Berhampur-761001, which comes under east and south eastern coastal plain zone of Odisha. Altogether 10 treatments including 6 intercropping combinations viz. T1: pigeon pea: yam bean =1:1 ; T2: pigeon pea: yam bean =2:2; T3: pigeon pea : french bean =1:1 ; T4: pigeon pea : french bean =2:2 ;T5: pigeon pea: cowpea =1:1 ; T6: pigeon pea: cowpea =2:2; and 4 sole crops ie.T7: sole pigeon pea (cv. manak); T8: sole yam bean (cv. safed hyb. sakalu) ; T9:Sole french bean (cv. Falguni latika) ; T10: Sole cowpea (cv. kashi kanchan) were laid out in randomized block design with three replications. Row spacing of sole crops were maintained as 50cm and intercrops were taken as additional series with row spacing of 25cm. Crop was sown during first fortnight of november in both the years (2016-17 & 2017-18). Finger millet was taken as preceding crop during *kharif* with fertilizer dose of 60:30:30 N,P_2O_5, K_2O kg ha^{-1}. For pigeon pea recommended package of practice for *rabi* was followed . Additional fertilizer was applied for intercrops as per respective recommended doses adjusting as per the plant population proportions. The soil was sandy loam with pH 6.2, low organic carbon (0.43 %), medium available phosphorus (21.67 kg ha^{-1}) and high available potassium (292 kg ha^{-1}). The crop was sown with pre-sowing irrigation and later four irrigations were given at critical stages of growth . Observations on yield of pigeon pea and component crops were taken at harvest and analysed as per statistical procedure described by Panse and Sukhatme (1985). Economics of the treatment combinations were calculated and compared on basis of pigeon pea seed equivalent yield for economic feasibility. Indices of competition such as land equivalent ratio (LER), monetary advantage index(MAI), competition ratio (CR), aggressivity(A), relative crowding coefficient (RCC) were calculated as follows .

The land equivalent ratio (LER), which is the relative land area under sole crop required to produce the same yield achieved in intercropping, was calculated using Willey and Osiru, 1972 formula.

LER = LERa + LERb = (Yab/Yaa) +(Yba/Ybb)

Where, LER a and LERb are individual LER of both crops ; Yab and Yba are yields of crop 'a' and 'b' respectively in mixed stand., Yaa and Ybb are pure crop yield of crop 'a' and 'b', respectively.

The monetary advantage index (MAI) as suggested by Willey (1979), gives the absolute value of the genuine yield advantage in intercropping. Higher the index , better is the cropping system.

MAI= Value of combined intercropping yield x (LER-1)/LER

Competition ratio (CR) is the ratio of individual LER of two component crops. It was calculated as under:

CRa = (LERa/ LERb) X(Zba/Zab) and CRb =(LERb/ LERa) X(Zab/Zba)

Where, CRa and CRb are the competition ratios of crop 'a' in mixture with 'b' and crop 'b' in mixture with 'a' respectively; LERa and LERb are individual LER of both crops ; Zba and Zab are sowing proportions of crop 'b' with 'a' and 'a' with 'b' respectively.

Aggressivity (A) determines the competitive abilities of the experimental crops. It is if zero, then both component crops are equally competitive. Positive value stands for dominant and negative value for dominated.

Aggressivity for 'a' (Aab) = Yab / (Yaa X Zab) – Yba / (Ybb X Zba) ;

Aggressivity for 'b' Aba= Yba / (Ybb X Zba)- Yab / (Yaa X Zab)

Where, Aab and Aba are the Aggressivities of crop 'a' in mixture with 'b' and crop 'b' in mixture with 'a' ; Yab and Yba are yields of crop 'a' and 'b' respectively in mixed stand., Yaa and Ybb are pure crop yield of crop 'a' and 'b' respectively. Zba and Zab are sowing proportions of crop 'b' with 'a' and 'a' with 'b' respectively.

Relative crowding co-efficient (RCC) indicates whether a species grown in mixture population has produced more or less yield than expected in pure stand (de Wit,1960 ; Hall, 1974).

RCCa=Kab = (Yab X Zba) / (Yaa –Yab)Zab ; RCCb= Kba = (Yba X Zab) / (Ybb –Yba) Zba Where, Kab is the Relative crowding co-efficient of crop 'a' in mixture with 'b' ; Yab and Yaa are yields of crop 'a' in mixed stand and pure crop respectively. Zba and Zab are sowing proportions of crop 'b' with 'a' and 'a' with 'b' respectively. When K>1, there is yield advantage, K=1, there is no difference and K < 1, there is yield disadvantage .

RESULS AND DISCUSSION

Yield and pigeon pea seed equivalent yield

The yield of intercrops were converted to pigeon pea seed equivalent yield considering the sale rate proportions for comparison (table-1). Pooled data of two years (2016-17

and 2017-18) revealed that yield of individual component crops in intercropping combinations were always less than their respective yield in sole crops. Similar results were reported by Panda *et al*. (2003) in case of pigeon pea and yam bean intercropping. The result also corroborated with the findings of Ahlawat (1998) and Das et al. (2002). The percentage of reduction in grain yield of pigeon pea in intercropping system with yam bean in 2:2 proportion was found minimum (22.8%) as compared to sole crop yield .Reddy *et al*. (1993) also reported that intercropping did not reduce the pigeon pea yield significantly. However combined yield expressed as pigeon pea seed equivalent yield were always found higher than both sole crops of intercropping system . Performance of paired row sowing (mean PSEY 38.24 q ha^{-1}) was found superior to that of single row intercropping (mean PSEY 32.71 q ha^{-1}). Among three combinations, intercropping with french bean was found most remunerative (mean PSEY 41.13q/ha) closely followed by that with yam bean (mean PSEY 37.84 q ha^{-1}),but conspicuously superior to intercropping with cowpea (mean PSEY 27.46 q ha^{-1}). Among intercrops, the french bean gave maximum PSEY (31.26 q ha^{-1}) when sown with pigeon pea in 2:2 proportion. This treatment also recorded maximum pigeon pea seed equivalent yield of the system (44.09 q ha^{-1}) .The next remunerative system was pigeon pea sown with yam bean in 2:2 proportion (system PSEY 41.15 q ha^{-1}, pooled).

Economics

Economics of individual treatments based on pooled PSEY and average minimum support price of pigeon pea for two years for evaluation of profitability are presented in table-1. Maximum gross monetary return (Rs. 2,31,473/ha), net monetary return (Rs. 231473 ha^{-1}) and B:C ratio (3.99) were recorded with T4 (Pigeon pea: French-bean =2:2 proportion). This system followed by pigeon pea and yam bean intercropping in 2:2 proportion with gross monetary return of Rs 2,16,038 ha^{-1} and net return of Rs. 1,54,038 ha^{-1}. The lowest values of gross monetary return (Rs. 90,090 ha^{-1}), net return (Rs. 52090/-) and B:C ratio (2.37) were associated with sole pigeon pea. Gross monetary return from intercropping system was always more than respective sole crops. Panda *et al*.(2003) also found similar results in pigeon pea-yam bean intercropping system in West Bengal .Therefore for enhancing farmers income from unit area pigeon pea can be introduced as intercrop with vegetables in irrigated areas during *rabi*.

Competition functions

Different competition functions such as land equivalent ratio (LER), monetary advantage index(MAI), competition ratio (CR), aggressivity(A), relative crowding coefficient (RCC) were calculated and interpreted for evaluation of intercropping compatibility of the systems(table-2) .Intercropping of compatible crops always have yield

Table 1: Yield and economics of *rabi* pigeonpea based intercropping systems (*Rabi*, 2016-18)

Sl. No.	Treatment	Yield of intercrop		Yield of pigeonpea	Yield reduction to sole crop	PSEY (q ha^{-1}) of system (pooled)	Gross return	Cost of prod.	Net return	B:C ratio
		(q ha^{-1})	PSEY (q ha^{-1})	(q ha^{-1})	(%)		(Rs)	(Rs)	(Rs)	(Rs)
T1	PP:YB =1:1	77.24	23.55	10.98	36.0	34.53	181282	62000	119282	2.92
T 2	PP:YB =2:2	91.56	27.91	13.24	22.8	41.15	216038	62000	154038	3.48
T 3	PP:FB =1:1	62.32	28.46	9.72	43.4	38.18	200445	58000	142445	3.46
T 4	**PP:FB =2:2**	**68.45**	**31.26**	**12.83**	**25.2**	**44.09**	**231473**	**58000**	**173473**	**3.99**
T 5	PP:CP =1:1	42.12	16.02	9.41	45.2	25.43	133508	45000	88508	2.97
T 6	PP:CP =2:2	50.78	19.31	10.18	40.7	29.49	154823	45000	109823	3.44
T 7	Sole PP	-	-	17.16	-	17.16	90090	38000	52090	2.37
T 8	Sole YB	112.64	34.34	-		34.34	180285	55000	125285	3.27
T 9	Sole FB	78.36	35.78	-		35.78	187845	48000	139845	3.91
T 10	Sole CP	68.82	26.17	-		26.17	137393	40000	97393	3.43
	SEm(±)					3.02				
	CD(5%)					9.14				

Avg. PSEY of paired rows:38.24q/ha
Avg. PSEY of single row: 32.71q/ha
Avg. PSEY frenchbean systems:41.13 q/ha
Avg. PSEY yam bean systems:37.84 q/ha
Avg. PSEY cowpea systems:27.46 q/ha

PP=Pigeonpea, YB=Yambean, FB= French bean, CP= Cowpea,
PSEY= Pigeonpea seed equivalent yield.
Average Sale rate (two years): PP= 5250/q, YB=1600/q, FB =2400/q & CP= 2000/q.

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Table 2: Evaluation of *rabi* pigeonpea based intercropping systems through various competition functions (*Rabi*, 2016-18)

Sl. No.	Treatment	Land equivalent ratio (LER)			MAI (Rs ha^{-1})	Competition ratio (CR)		Aggressivity (A)		Relative crowding coefficient (RCC)		
		LERa	LERb	LERa+b		CRa	CRb	Pigeon pea (Aa)	Intercrop (Ab)	Pigeonpea (Kab)	Intercrop (Kba)	System (Kab+Kba)
T1	PP:YB =1:1	0.64	0.64	1.28	34334	1.00	1.00	0.00	0.00	1.78	1.79	3.57
T 2	PP:YB =2:2	0.83	0.75	1.58	69686	1.11	0.90	0.16	-0.16	4.88	3.01	7.89
T 3	PP:FB =1:1	0.57	0.80	1.37	32437	0.71	1.40	-0.46	0.46	1.31	3.88	5.19
T 4	PP:FB =2:2	0.75	0.87	1.62	74176	0.86	1.16	-0.24	0.24	2.96	6.90	9.86
T 5	PP:CP =1:1	0.55	0.61	1.16	17377	0.90	1.11	-0.12	0.12	1.21	1.58	2.79
T 6	PP:CP =2:2	0.59	0.74	1.33	36407	0.80	1.25	-0.30	0.30	1.46	2.82	4.28
T 7	Sole PP	1.00	--	1.00	--							
T 8	Sole YB	--	1.00	1.00	--							
T 9	Sole FB	--	1.00	1.00	--							
T 10	Sole CP	--	1.00	1.00	--							

NB: Component 'a' is pigeon pea and 'b' is intercrop

advantages over respective sole crops (Panda *et al*. 2003). Land equivalent ratio of component 'a' (pigeon pea) in intercropping combinations was found maximum (0.83) with pigeon pea : yam bean in 2:2 proportion. Among intercrops (component 'b'), french bean sown in 2:2 proportion registered maximum LER (0.87). This indicates that both component crops have exhibited their maximum productivity under 2:2 proportion planting geometry, which proved their compatibility. When LER of the system was computed, T4 (Pigeon pea : french bean =2:2) also registered highest LER (1.62) showing its compatibility and high productivity. This intercropping combination also registered maximum MAI (Rs.74176/-) and proved its profitability. Higher the index better is the cropping system. Individual competition ratio of both component crops were computed and presented in table-2. Competition ratio of pigeonpea when sown with french bean and cowpea were less than one ,which indicates that intercrops were dominant on pigeon pea. However, pigeon pea was equally competitive with yam bean when sown in 1:1 proportion (CRa =1.0) and dominant when sown in 2:2 proportion (CRa =1.11). Aggressivities of crop 'a' (pigeon pea) and crop 'b' (intercrop) was calculated and presented in table-2. The data indicates that aggressivity of crop 'a' (pigeon pea) was found to have negative value when pigeon pea was sown with french bean and cowpea. This indicates that pigeon pea was dominated and french bean / cowpea was dominant component in the system. Conversely, aggressivity of crop 'b' (intercrop) was positive for french bean and cowpea proved their dominance over pigeon pea. Nevertheless, pigeon pea was found equally competitive with yam bean when sown in 1:1 proportion, having aggressivity value zero and dominant over yam bean when sown in 2:2 proportion with positive aggressivity value (0.16). The RCC of a given species in mixture indicates whether it has produced more or less yield than expected in pure stand. RCC of pigeon pea (RCCa), intercrops (RCCb) and RCC of the system (RCCa$_+$b) were computed and exhibited in table-2. The data revealed that pigeon pea has maximum yield advantage (4.88) when sown with yam bean in 2:2 proportion. Among intercrops french bean registered maximum co-efficient (6.9) when sown in 2:2 proportion with pigeon pea. RCC of pigeon pea and respective intercrops were added for computing total RCC of the system. The data revealed that the treatment T4 (Pigeon pea : French bean =2:2) registered highest system relative crowding coefficient (RCCa$_+$b) (9.86) showing its yield advantage over other cropping systems.

CONCLUSION

It was concluded that the pigeon pea can be introduced as intercrop with vegetables in the vast alluvial up-medium land basin of the river Rushikulya and similar agro-ecological situations with irrigation facility in east and south eastern coastal plain zone of Odisha during *rabi* season after harvest of finger millet grown during *kharif*. Therefore, for enhancing farmers income per unit area, intercropping of pigeon pea with french bean in 2:2 proportion or with yam bean in 2:2 proportion during *rabi* may be recommended.

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Management of white grubs through a novel technology in Uttarakhand hills of North-West Himalayas

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ABSTRACT

White grubs, a group of destructive insect pests of polyphagous nature, cause severe damage to crop plants in hill ecosystem. The grubs with subterranean habitat feed extensively on the roots and the adults defoliate the plants. A two-pronged strategy involving an efficient, light weight, eco-friendly, low cost, light-based insect trap for capturing the adults and a novel entomopathogen, *Bacillus cereus* strain WGPSB-2 for the management of grubs was developed. Large scale deployment of the above technology was done on community basis in 18 villages of low, mid and high-altitude areas including two experimental farms of Uttarakhand hills. Three years experimentation revealed drastic reduction in beetle population to the tune of 75.8, 78.5 and 80.5 per cent in low, mid and high-altitude villages respectively. A significant reduction of the grub population was recorded in the range of 87.8 to 95.7 per cent in three years across the different villages. As a result of reduction in grub population, per cent increase in yield of different crops was recorded from 23.8 to 187.9 per cent in different villages and experimental farms of low, mid and high altitudes. The technology is thus, capable of managing white grubs at different altitudes of hills in general and North Western Himalayas in particular.

Keywords: *Bacillus cereus*, entomopathogen, light trap, Uttarakhand Hills, white grubs.

White grubs (Coleoptera: Scarabaeidae) are polyphagous insect pests with cosmopolitan distribution. Crop damage by white grubs has been identified as one of the limiting factors in Indian agriculture (Sharma, 2002). Among the different agro–ecological regions of India, the North Western (NW) Himalayan region comprising of the states of Uttarakhand (UK), Himachal Pradesh (HP) and Jammu-Kashmir (J&K) has been identified as a hot spot for white grub diversity. The damage caused by the grubs under rainfed condition, which account for about 80 per cent of cultivated land in Uttarakhand and HP and 59 per cent in J&K, may vary on an average from 10-30 per cent but some times complete crop failure is also observed. In NW Himalayas, cereals, millets, pulses, oilseeds and vegetables are cultivated in 1.32 million hectare area with an estimated production of 3.44 million tonnes under rainfed condition during *kharif* season. The damage caused by the white grubs under such situation at the rate 10 per cent amounts to a loss of 0.34 million tonnes, which consequently causes an estimated economic loss of Rs 1.8 billion annually (Sushil *et al*. 2008 a).

Due to intensification of agricultural activities and destruction of forests, white grubs have attained the status of pest of national importance during the last three decades. The grubs, feed on the roots of many agricultural crops, weed hosts, forest trees, ornamental and horticultural crops where as the adults feed on foliage, flower, pollen and fruits of the crops. There is a complex of white grub species prevalent in the agricultural areas of NW Himalaya. Existence of as many as 78 species belonging to sub families Rutelinae, Melolonthinae, Dynastinae, Cetoniinae, Aphodiinae, Euchirinae and Scarabaeinae has been documented from the Uttarakhand alone (Garg, 1992). In Uttarakhand, *Anomala dimidiata, Holotrichia longipennis* and *H. seticollis* have been reported as the dominant white grub species (Garg, 1992; Bhatt and Sushil, 2004; Sushil *et al*., 2007) and cause severe damage to upland rice, maize, finger millet, barnyard millet, potato, chilli, cole crops, pulses and other high value crops.

In the NW Himalayas, majority of the cropped area is under well drained sandy soil along the hill slopes, which is favourable for growth and development of white grubs. Several insecticides are recommended for the control of white grubs, but as these do not provide satisfactory control unless used in a very high dose, become hazardous and uneconomical besides causing pollution. Therefore, the challenge before researchers is to preserve the eco-system of Himalayas on one hand and restrict the damage caused by phytophagous white grubs on the other. A satisfactory solution to these conflicting demands can be met through developing safer control methods. Of the four stages *viz*.,

egg, grub, pupa and adults in the life cycle of the white grubs, only the adult and grub stages are the most vulnerable for targeting the pest for their management (Sushil *et al.* 2008a). Considering the potential of natural bacterial epizootics on white grubs in this region, the concept of microbial control was considered as an alternative strategy for the management of the pest at the grub stage. Hence, an in-depth research on isolation and characterization of potential entomopathogenic bacteria was undertaken so as to develop a viable, environmentally safe microbial agent against white grubs. Similarly, mechanical means of management was considered as the most appropriate for managing the adult stage of the pest. Trapping of adult white grub beetles with a light source was also considered economical over pheromones in the context of multi species distribution. The earlier models of insect trap were found less effective due to one or other drawbacks. Hence, development of an efficient, low-cost user-friendly light trap was felt necessary considering the socio-economic condition of the farmers of the region.

We report here development of a technology comprising of two components *viz*., an insect trap for controlling the adults and a bio-agent, comprising of the talc-based formulation of the bacterium, *Bacillus cereus* strain WGPSB-2, for managing the grubs (Sushil *et al.*, 2008a; Sushil *et al.*, 2008b; Selvakumar *et al.*, 2007). The results demonstrate the effectiveness of both the components of the technology in a larger area covering 18 villages across the different altitudes of Uttarakhand hills of the NW Himalayas.

MATERIALS AND METHODS

Selection of villages

Severely affected eighteen villages (including two experimental farms) covering low, mid and high altitudes of Uttarakhand hills were selected for testing and demonstration of the newly developed technologies for the management of white grubs on community basis (Figure 1 & Table 1). The villages were selected based on random samplings for white grub occurrence. Only villages having more than 10 grubs/ m^2 at 10 locations were selected.

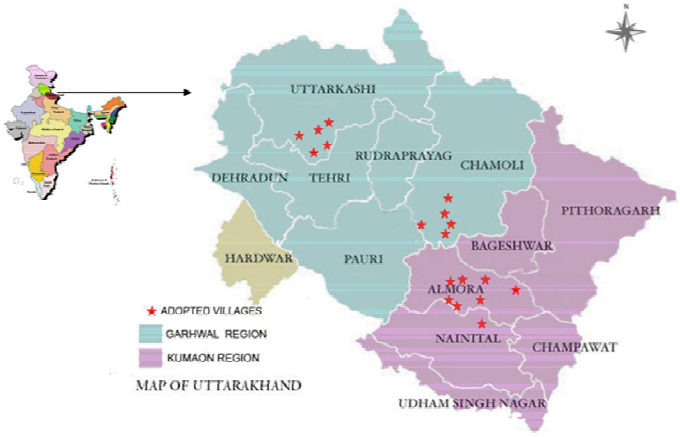

Figure 1. Map of Uttarakhand state showing locations of adopted villages.

Table 1. List of villages adopted for testing and demonstration of white grub management technologies

Sl. No.	Village	Altitude (m amsl)	Block	District
1. Low Hills (<1000 m amsl)				
1.	Mahargaon	900	Chinyalisaur	Uttarkashi
2.	Barethi	855	Chinyalisaur	Uttarkashi
3.	Tuliyara	850	Chinyalisaur	Uttarkashi
4.	Galari	850	Chinyalisaur	Uttarkashi
5.	K.V.K. Farm, Chinyalisaur	855	Chinyalisaur	Uttarkashi
2. Mid Hills (1000 – 1500 m amsl)				
1.	Chausali	1133	Hawalbagh	Almora
2.	Tipola	1090	Tarikhet	Almora
3.	Tunakot	1065	Tarikhet	Almora
4.	Daulaghat	1285	Hawalbagh	Almora
5.	Govindpur	1310	Hawalbagh	Almora
6.	Manan	1350	Takula	Almora
3. High Hills (>1500 m amsl)				
1.	Gwaldam	1960	Tharali	Chamoli
2.	Sunnowmalla	1650	Tharali	Chamoli
3.	Taal	1900	Tharali	Chamoli
4.	Patla	1850	Tharali	Chamoli
5.	Sainji	1650	Gairsain	Chamoli
7.	Bhagartola	1900	Dhauladevi	Almora
8.	Darim	1850	Mukteshwar	Nainital

Installation of insect traps

Considering the topography and aspect of the mountain, the newly developed light mediated insect trap, VL-White grub beetle Trap-1 (Figure 2) were installed at strategic locations of the adopted villages. Normally, one trap was installed in 1-2 ha area for mass trapping of the beetles. A diagrammatic representa-tion of the light trap installation at strategic locations of the village Manan is shown in Figure 3. A similar strategy was followed in other adopted villages.

Figure 2. VL White grub beetle Trap-1.

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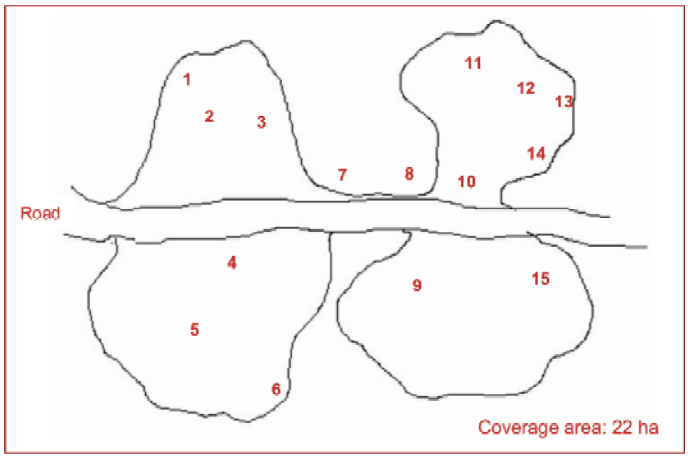

Figure 3. Diagrammatic representation of VL White grub beetle Trap-1 installation at the adopted village Manan (1350 m amsl).

Application of bio-pesticide

Talc-based formulation of *B. cereus* strain WGPSB-2 (Figure 4) with a spore load of $1x10^{10}$ / g was prepared as per the method of Sushil *et al*., 2008a. and the same was applied in all the compost pits made in the adopted villages and subsequently in the fields in order to get desired level of disease occurrence and mortality in the grub population (Figure 5). The talc-based formulation was applied in the compost pits at the rate of 1 kg/ton basis, a month before application in the field. It is noteworthy, that *B. cereus* strain WGPSB-2 has strong ability to colonize on different compost substrates (Sushil *et al.*, 2008a). In the worst affected fields, a dose of 10 kg/ha talc-based formulation of the bio-pesticide was applied directly.

Figure 4. Talc formulation of *Bacillus cereus* strain WGPSB-2.

Data recording and statistical analysis

Recording of the beetles trapped in the VL-White Grub Beetle Trap-1 was done regularly from June to October during 2006 to 2008 in all the adopted villages. The number of beetles trapped and per cent reduction during the study period of three years has been calculated. Samplings were done every month for recording the grub occurrence under field condition. Pits of 30 cm^2 were dug with a depth of 20 cm at 20 random locations for assessment of the white grub population in the village for one crop. A similar method was adopted for different crops grown in the adopted villages. Data obtained was subjected to standard deviations and mean was calculated for comparison. Mean yield of different crops grown in the village was recorded and per cent increase in yield was calculated in comparison of the non-adopted villages for consecutive three years.

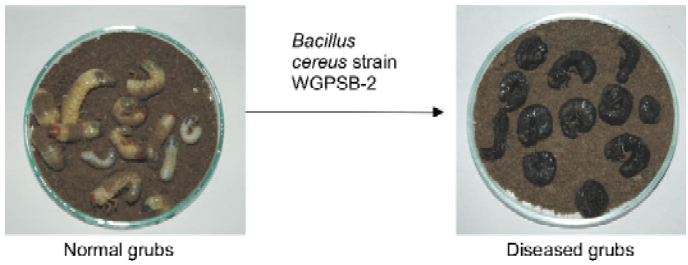

Figure 5. White grubs infected by *Bacillus cereus* strain WGPSB-2.

RESULTS AND DISCUSSION

Management of beetle stage

Altogether 161 newly developed insect traps (VL White Grub Beetle Trap-1) were installed at strategic locations of 16 adopted villages and 2 experimental farms in low, mid and high altitudes. In low hills, 47 traps were installed in 5 villages. Using these traps 1.25, 0.72 and 0.30 lakhs beetles were trapped during 2006, 2007 and 2008 respectively. Reduction in beetle's population to the tune of 42.12 and 75.79 per cent was recorded during 2007 and 2008 respectively over the beetle catches of 2006 (Table 2). In mid hills, 49 traps were installed in 6 villages resulting in beetle catches of 3.90, 2.18 and 0.83 lakhs during 2006, 2007 and 2008 respectively. A decline in beetle population to the tune of 43.90 and 78.45 percent was recorded during 2007 and 2008 respectively over the beetle catches of 2006 (Table 3). Similarly, in high hills, 65 traps were installed in 7 villages and 3.95, 2.12 and 0.53 lakhs beetles were trapped during 2006, 2007 and 2008 respectively. Mean per cent reduction in beetle population was 45.63 and 80.46 during 2007 and 2008 respectively over the beetle catches of 2006 (Table 4). The peak period of beetle emergence was July at all the altitudinal sites. Population density and species composition of scarabaeids differed in all the adopted villages located at different altitudes. Altogether, 36 species in low hills, 51 species in mid hills and 26 species in high hills were recorded in the insect traps, besides many of them were unidentified.

In low hills, *Anomala dimidiata*, *A. rugosa*, *A. tristis*, *Holotrichia longipennis*, *H. seticollis* and *Xylotrupes gideon* were dominant. In mid hills, *A. dimidiata*, *H. longipennis*, *Hemiserica nasuta* and *X. gideon* whereas in high hills, *A. dimidiata*, *H. longipennis*, *H. seticollis*, *H. rosettae* and *X. gideon* were found to be the dominant species.

Management of grub's population

***Low hills*:** Reduction of grub population over the years was recorded in five different villages through pit sampling. Among the adopted villages of low hills, population of white grubs recorded in the village Mahargaon is presented in Table 5. Crop wise pit sampling data revealed a substantial reduction of white grub population over the years. The grub population was very high in June 2006, which went up to 2.6 grubs/ 30 cm^2 in tomato, 2.3 in potato and 1.6 in French bean. However, due to the continuous mass trapping of beetles and application of the bio-agent, *B. cereus* strain WGPSB-2 through FYM, a remarkable reduction in the grub population to a tune of 0.6 grubs/ 30 cm^2 in Aug 2007 and 0.4 grubs/ 30 cm^2 in Aug 2008 were recorded in tomato fields. By the end of the cropping season in 2008, the grub population was reduced to 0.2 and 0.4/ 30 cm^2 in potato and French bean fields, respectively. The mean per cent reduction in the grub population was 75.4 per cent by the end of second year and it reduced further up to 83. 6 per cent by the end of the third year. Similar trend was recorded in the other adopted villages of low hills.

Table 2. Management of white grubs through insect traps on community basis in low hills (<1000 m amsl)

Adopted villages	Number of light traps installed	Beetles trapped (in lakhs)			Beetle reduction over 2006 (%)	
		2006	2007	2008	2007	2008
Mohargaon	12	0.25	0.15	0.064	42.36	74.79
Barethi	10	0.22	0.15	0.069	34.39	69.29
Tuliyara	10	0.24	0.14	0.039	39.99	84.67
Gailari	9	0.26	0.14	0.038	44.99	83.98
Chinyalisaur	6	0.28	0.14	0.095	48.90	66.24
Total	47	1.25	0.72	0.305	42.12 (Mean)	75.79 (Mean)

Table 3. Management of white grubs through insect traps on community basis in mid hills (1000 – 1500 m amsl).

Adopted villages	Number of light traps installed	Beetles trapped (in lakhs)			Beetle reduction over 2006 (%)	
		2006	2007	2008	2007	2008
Chausali	12	1.01	0.57	0.15	43.56	85.24
Tunakot-Tipola	07	0.57	0.37	0.16	35.09	72.42
Daulaghat	07	0.72	0.37	0.13	48.61	82.50
Govindpur	08	0.62	0.32	0.14	48.39	77.80
Manan	15	0.98	0.55	0.25	43.88	74.28
Total	49	3.90	2.18	0.83	43.90 (Mean)	78.45 (Mean)

Table 4. Management of white grubs through insect traps on community basis in high hills (>1500 m amsl)

Adopted villages	Number of light traps installed	Beetles trapped (in lakhs)			Beetle reduction over 2006 (%)	
		2006	2007	2008	2007	2008
Taal	6	0.43	0.21	0.081	51.16	81.06
Sunnow Malla	5	0.37	0.18	0.072	51.35	80.54
Patla	4	0.27	0.15	0.045	44.44	83.32
Sainji	5	0.33	0.16	0.065	51.52	80.18
Gwaldam	10	0.71	0.37	0.116	47.89	83.63
Darim	18	1.25	0.57	0.220	54.40	82.23
Bhagartola	17	0.59	0.48	0.160	18.65	72.28
Total	65	3.95	2.12	0.531	45.63 (Mean)	80.46 (Mean)

Mid hills: Pit sampling for grub population was done in tomato, capsicum, French bean and cabbage fields in six adopted villages of mid hills. The data presented in Table 6 reveals the reduction of grub population in the village Manan. In June 2006, the grub population ranged from 1.2 to 1.8 / 30 cm^2 in different crops. It was reduced to 0.1 to 0.3 grubs / 30 cm^2 in June 2007 and further to 0.05 to 0.1 grubs / 30 cm^2 at the end of the third year. The reduction in the grub population was in the range of 25.0 to 76.5 per cent in 2006 and 85.7 to 94.4 per cent in 2007. More than 90% reduction in grub population was recorded in all the crops and even up to 96.4 per cent in cabbage in the year 2008. The mean reduction of grub population was found to be 53.7, 92.0 and 93.8 per cent in 2006, 2007 and 2008, respectively. Similar trend was recorded in the other adopted villages of mid hills.

High hills: Pit sampling data on the population of white grubs of the village Bhagartola situated in high hills is presented in Table 7. The grub population was found to be as high as 2.6 to 3.4/ 30 cm^2 in June 2006 in different crops. The population of the grubs was reduced to 0.1 to 0.3 / 30 cm^2 in 2007 and 0.1 to 0.2 / 30 cm^2 in 2008. The reduction was found to be 50.0 to 89.2 per cent in the first year, 89.2 to 96.8 per cent in the second year and 92.9 to 97.0% in the third year. The mean reduction was 95.7 per cent at the end of three years of experimentation. Observations on the grub population of all the adopted villages of high hills revealed a similar trend of reduction.

Yield enhancement

Mean yield of different crops was recorded in all the villages and per cent increase in yield of the adopted villages was calculated and compared with the non-adopted villages of different altitudes. Data presented in the Table 8, reveal

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the per cent increase in yield of different crops in the adopted villages over non-adopted villages. In low hills, mean per cent increase in yield by 51.7 in tomato, 46.4 in potato, 39.0 in chilli and 59.2 in French bean was recorded during the three years of experimentation. In mid hills, the mean per cent increase in yield of tomato, capsicum, French bean and cabbage was 40.6, 68.6, 187.9, 68.8 respectively. In high hills, 23.8, 33.0, 39.5, 38.9 and 53.1 per cent increase in yield of tomato, potato, capsicum, French bean and cabbage was recorded in the adopted villages over non-adopted villages in three years.

A complex of white grub species has been recorded in Uttarakhand hills, where some of the species cause extensive damage to crop plants (Sushil *et al.*, 2008a). The present investigation for the management of white grubs targets two vulnerable stages of the pest *viz.*, adult and grub stages. The two-pronged strategy resulted in a significant reduction in the white grub population as evidenced by about 80 per cent reduction in beetle population within 2 to 3 years on entire village basis. Light based insect traps have been reported to be effective tool for mass trapping of the scarab beetles by other researchers too (Carne and Chinnick,1956; Hosking, 1979; Kato *et al.*, 2000). Light based insect traps, unlike pheromone traps, are known to attract insects of both sexes of the scarabaeid (Harai *et al.*, 2000) and this is in conformity of the findings of the present study, wherein both the sexes of more than 50 species of scarabaeids have been recorded through the light traps. The newly developed light trap has several advantages over the already reported models. The beetle hitting surface area (fins), their angle and arrangements around the light source have been designed in such a way, so as to trap more beetles into the trough. The trap is specific to the scarabaeids and was found to trap negligible number of beneficial insects. Further the new model is light weight, theft proof and user friendly too.

The early-stage grubs are known to feed on soil organic matter and hence it is the most suitable stage to control them through soil inhabiting entomopathogens. Therefore, microbial control was considered as viable and sustainable strategy for its management. The soil environment, rich in entomopathogens (Hochberg, 1989) predisposes the soil dwelling white grubs to a large array of diseases (Jackson, 1999). Pathogenicity and environmental competence are considered as the two key features of any potential microbial control agent used for soil pests (Klein, 1992). Bacteria such as *Bacillus thuringiensis*, *Paenibacillus popilliae*, *P. lentimorbus*, *Micrococcus* sp., *Serratia entomophila* and *S. proteamaculans* have been isolated from white grubs (Suzuki *et al.*, 1994; Bourner *et al.*, 1996). *P. popilliae* and *P. lentimorbus* are the causal agents of milky disease in scarab larvae. Although *in vivo*-produced spores of these bacteria were used for the control of a few scarab species, including *H. consanguinea*, their continued production and widespread use as biological control agents have been constrained by the inability of the researchers to mass-produce the spores *in vitro* (Vyas *et al.*, 1991; Stahly and Klein, 1992). In the present investigation, we isolated, characterized and evaluated the potential of bacterial pathogens as biological control agents against white grubs of north western Himalayan hill region. *Bacillus cereus* strain WGPSB-2 was found to cause about 90% mortality in predominant species of white grubs of the region. Thus, considering the potential of the bio-agent, *B. cereus* strain WGPSB-2, protocol for mass production and talc-based formulation was developed and found effective (Sushil *et al.*, 2008). This is in conformity with the earlier report by Sezen *et.al.*, 2005. in which the insecticidal effect of a *B. cereus* isolate on larvae of the scarab, *Amphimallon solstitiale* was found to an extent of 90% mortality. In addition, several strains of *B. cereus* have also been isolated from various insects (Lipa and Wiland, 1972; Sezen and Demirbag, 1999).

The two-pronged strategy using light trap and the entomopathogen (*B. cereus* strain WGPSB-2) for the control of adult and grub stages of the pest on entire village basis is the first attempt of this kind. Mass trapping of the beetles led

Table 5. Per cent reduction of grub population in different crops of the adopted village, Mahargaon (900 m amsl) in low hills

Crop	Mean number of grubs/square feet (30 cm²) ± standard deviation															Per cent reduction in grub population over June, 2006		
	2006					2007					2008							
	June	July	Aug	Sep	Oct	June	July	Aug	Sep	Oct	June	July	August	Sep	Oct	2006	2007	2008
Tomato	2.6±0.7 (1-5)	2.4±0.3 (0-5)	2.0±0.4 (0-3)	-	-	0.8±0.8 (1-2)	0.6±0.8 (1-2)	0.6±0.5 (0-1)	-	-	0.7±0.5 (0-1)	0.6±0.8 (0-2)	0.4±0.5 (0-1)	-		23.0	76.9	84.6
Potato	-	2.3±0.5 (1-3)	1.8±0.4 (0-3)	1.2±0.6 (0-3)	0.8±0.1 (0-2)	-	1.1±0.9 (1-2)	0.6±0.8 (1-2)	1.1±0.9 (1-2)	0.3±0.5 (0-1)	-	0.8±0.8 (0-2)	0.6±0.8 (0-2)	0.5±0.5 (0-1)	0.2±0.5 (0-1)	65.2	86.9	91.3
French bean	1.6±0.1 (0-2)	1.0±0.5 (0-1)	-	-	-	1.1±0.9 (1-2)	0.6±0.5 (0-1)	-	-	-	0.8±0.8 (0-2)	0.2±0.8 (0-2)	-	-		37.5	62.5	87.5
Mean per cent reduction in grub population																41.9	75.4	87.8

* Figures in parentheses are range.

Table 6. Per cent reduction of grub population in different crops of the adopted village, Manan (1350 m amsl) in mid hills

Crop	Mean number of grubs/square feet (30 cm²) ± standard deviation														Per cent reduction in grub population over June, 2006		
	2006				2007					2008					2006	2007	2008
	June	July	Aug	Sep	June	July	Aug	Sep	Oct	June	July	August	Sep	Oct			
Chilli	1.8±1.0 (1-4)	1.2±0.6 (0-2)	0.7±0.8 (0-2)	-	0.3±0.7 (0-2)	0.1±0.3 (0-1)	0.1±0.3 (0-1)	-	-	0.2±0.4 (0-1)	0.2±0.4 (0-1)	0.1±0.3 (0-1)	0.1±0.3 (0-1)	0.1±0.3 (0-1)	61.1	94.4	94.4
Radish	1.2±0.4 (0-1)	0.9±0.4 (0-2)	-	-	0.2±0.4 (0-1)	0.2±0.4 (0-1)	0.1±0.3 (0-1)	0.1±0.3 (0-1)	-	-	0.2±0.4 (0-1)	0.1±0.3 (0-1)	0.1±0.3 (0-1)	0.1±0.3 (0-1)	25.0	91.7	91.7
Cabbage	1.4±0.5 (1-2)	0.8±0.5 (0-1)	-	-	0.1±0.3 (0-1)	0.2±0.4 (0-1)	-	-	-	0.1±0.3 (0-1)	0.1±0.3 (0-1)	0.1±0.3 (0-1)	0.2±0.4 (0-1)	0.05±0.1 (0-1)	42.9	85.7	96.4
Capsicum	1.7±1.1 (0-2)	1.1±0.6 (0-2)	0.4±0.5 (0-1)	-	0.1±0.3 (0-1)	0.1±0.3 (0-1)	0.2±0.4 (0-1)	0.1±0.3 (0-1)	-	0.2±0.4 (0-1)	0.2±0.4 (0-1)	0.1±0.3 (0-1)	0.1±0.3 (0-1)	-	76.5	94.1	94.1
Tomato	1.4±0.7 (0-2)	0.9±0.7 (0-2)	0.9±0.7 (0-2)	0.4±0.3 (0-1)	0.1±0.3 (0-1)	0.1±0.3 (0-1)	0.2±0.4 (0-1)	0.2±0.4 (0-1)	0.1±0.3 (0-1)	0.2±0.4 (0-1)	0.1±0.3 (0-1)	0.1±0.3 (0-1)	0.1±0.3 (0-1)	-	71.4	92.9	92.9
Brinjal	1.5±0.5 (1-2)	1.0±0.7 (0-2)	0.8±0.2 (0-2)	-	0.1±0.3 (1-1)	0.2±0.4 (0-1)	0.2±0.4 (0-1)	0.2±0.4 (0-1)	0.1±0.3 (0-1)	-	0.1±0.3 (0-1)	0.1±0.3 (0-1)	0.1±0.3 (0-1)	-	46.7	93.3	93.3
Mean per cent reduction in grub population															53.9	92.0	93.8

Figures in parentheses are range.

Table 7. Per cent reduction of grub population in different crops of the adopted village Bhagartola (1900 m amsl) in high hills

Crop	Mean number of grubs/square feet (30 cm²) ± standard deviation												Per cent reduction in grub population over June, 2006		
	2006			2007					2008				2006	2007	2008
	July	Aug	Sep	June	July	Aug	Sep	Oct	June	July	August	Sep			
Cabbage	2.6±0.7 (2-4)	1.1±0.5 (0-3)	0.7±0.3 (0-2)	-	0.3±0.5 (0-1)	0.3±0.5 (0-1)	0.2±0.4 (0-1)	0.1±0.3 (0-1)	-	0.2±0.4 (0-1)	0.2±0.4 (0-1)	0.1±0.3 (0-1)	73.07	96.1	96.1
Cauliflower	3.2±0.7 (2-4)	1.9±0.7 (1-3)	0.6±0.1 (0-2)	-	0.4±0.7 (0-2)	0.2±0.4 (0-1)	0.1±0.3 (0-1)	0.1±0.3 (0-1)	-	0.2±0.4 (0-1)	0.1±0.4 (0-1)	0.1±0.3 (0-1)	81.25	96.8	96.8
Capsicum	2.8±0.9 (2-4)	1.4±0.5 (0-3)	0.3±0.1 (0-1)	0.2±0.4 (0-1)	0.2±0.4 (0-1)	0.2±0.4 (0-1)	0.3±0.4 (0-1)	-	0.2±0.4 (0-1)	0.1±0.4 (0-1)	0.2±0.4 (0-1)	0.2±0.4 (0-1)	89.28	89.2	92.9
Potato	3.4±0.8 (2-4)	1.7±0.6 (0-3)	-	0.1±0.3 (0-1)	0.4±0.7 (0-2)	0.3±0.6 (0-2)	-	-	0.3±0.6 (0-2)	0.2±0.4 (0-1)	0.1±0.4 (0-1)	-	50.0	91.1	97.0
Mean per cent reduction in grub population													73.4	93.3	95.7

*Figures in parentheses are range.

Table 8. Per cent increase in yield of different crops in the adopted villages over non adopted villages

Location	Crop*	Mean Yield (q ha^{-1})									Mean percent increase in yield
		2006-07			2007-08			2008-09			
		Adopted village	Non adopted village	Increase (%)	Adopted village	Non adopted village	Increase (%)	Adopted village	Non adopted village	Increase (%)	
Low hills	Tomato	171.0	105.0	62.8	170.6	104.0	64.0	237.6	185.5	28.4	51.7
	Potato	213.6	150.0	42.4	215.8	145.0	48.8	211.8	143.0	48.1	46.4
	Chilli	48.7	36.0	35.2	51.7	35.0	47.7	58.8	40.0	46.9	39.0
	French bean	88.1	50.0	76.2	89.2	50.0	78.4	86.2	80.0	23.1	59.2
Mid hills	Tomato	238.5	156.3	52.6	215.7	154.0	40.1	218.0	155.0	29.0	40.6
	Capsicum	190.0	102.5	85.4	179.6	-	-	182.0	120.0	51.7	68.6
	French bean	208.0	61.3	239.3	177.4	75.0	136.5	177.4	-	-	187.9
	Cabbage	185.0	85.0	117.6	120.5	85.0	41.8	124.8	85.0	47.0	68.8
High hills	Tomato	195.0	157.5	23.8	208.0	190.0	08.6	204.1	147.0	38.9	23.8
	Potato	153.0	115.0	33.0	157.2	118.0	33.2	159.4	120.0	32.8	33.0
	Capsicum	106.7	88.8	20.2	104.6	80.0	30.8	100.4	60.0	67.4	39.5
	French bean	70.0	60.0	16.7	74.7	47.0	58.9	67.7	48.0	41.1	38.9
	Cabbage	383.7	212.5	80.6	209.5	152.0	37.8	222.5	158	40.8	53.1

*Varieties: Tomato-Manisha/Himsona; Potato-Kufri jyoti; Chilli-Pant chilli; French bean-Contender/Arka komal; Capsicum-California wonder; Cabbage-Golden acre/Varun

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to the significant reduction in egg laying in the villages and application of the talc-based formulation of the *B. cereus* strain WGPSB-2 through compost led to reduction of the grub population. Based on periodic pit sampling, the overall reduction of grub population ranged from 70.5 to 84.3% in low hills, 85.3 to 90.9% in mid hills and 70.7 to 93.4% in high hills of Uttarakhand state with a significant increase in crop yield. Approximately 381 hectare area was covered across the Uttarakhand hills benefiting 1400 farm families during the course of experimentation. The present investigation on white grub management through combination of light mediated insect trap and bacterial formulation has led to the development of eco-friendly, cost effective and sustainable technology. Adoption of this approach is expected to help in the management of white grubs in other areas too.

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Integrated management of gundhi bug (*Leptocorisa* spp.) infesting aromatic rice

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ABSTRACT

The field trial conducted at Rice Research Farm of Birsa Agricultural University, Ranchi during *Kharif* season of 2018 and 2019 with aromatic rice variety Birsa Vikas Sugandh-1 revealed that three foliar sprayings applied at 25,55 and 85 days after transplanting with tipronu 5SC @ 1200 ml ha^{-1} recorded lowest mean gundhi bug population (1.96) and highest polled mean yield (47.17 q ha^{-1}) followed by 2.9 and 46.13 q ha^{-1}, respectively recorded with fipronil 5SC @ 1000 ml ha^{-1}. Fipronil @ 1200 ml ha^{-1} recorded increased grain yield of 29.04 q ha^{-1} over the control. Alternate spray of fipronil and botanical insecticide neem (2.5 l ha^{-1}) also recorded appreciable reduction in gundhi bug population (4.21) and 23.5 per cent increase in rice grain yield over the control.

Key word: Gundhi bug, aromatic rice, Birsa Vikas Sugandh-1, integrated management

Rice gundhi or earhead bug, *Leptocorisa acuta* (Coreidae: Hemiptera) is one of the serious pests of rice in India causing reduction of rice yield to an extent of 30 per cent (Tiwari *et al*., 2014). Both nymphs and adults suck the sap from rice grains at milking stage when the starch formation within the grains are yet to be fully completed rendering in the grain to became chaffy (Sugimoto *et al*., 1995). During feeding, rice bugs also contaminate the grain endosperm with microorganisms (Shepard *et al*., 1995). The site of feeding/puncturing turns black leading to sterility in rice grain.

MATERIALS AND METHODS

The field experiment was conducted at Rice Research Farm of Birsa Agriultural University during *Kharif* season of 2018 and 2019 with aromatic rice variety Birsa Vikas Sugandh-1. Twenty one days old seedlings were transplanted from nursery to main field. All the locally recommended package of practices free from insecticide application were adopted. Field experiment was laid out in Randomized Block Design with nine treatments (including control) and three replications. Three foliar sprays with the respective insecticidal treatment(s) were applied at 25, 55 and 85 DAT using knap sack sprayer. A cloth screen around the plot was erected to check the insecticidal drift. The bunds around the sprayed plot were kept intact for 2-3 days to check the entry of the insecticide from one plot to another to maintain precision. Observations on gundhi bug population (no. of bugs 10 hills^{-1}) was recorded during milking stage at 7, 10 and 14 days coinciding with the third rounds of foliar sprays.

RESULTS AND DISCUSSION

The results presented in table-1 revealed that incidence of gundhi bug during both the year of experimentation was not only affected by third round spray of the insecticide (85 DAT) but was also affected by first (25 DAT) and second (55 DAT) round spray of the insecticides. However, incidence of gundhi bug was less with fipronil applied at a dose of 1200 ml ha^{-1} during third round of the spray. All the treatments were effective over the control. The lowest mean number of 3.17, 1.67, 1.33 and 1.67 gundhi bugs 10 hills^{-1} were found at 4, 7, 10 and 14 days after application with three foliar application of fipronil 5SC @ 1200 ml ha^{-1} (T_2). This was followed by three application of fipronil 5SC @ 1000 ml ha^{-1} (T_1) with overall mean 2.92. Alternate application of fipronil 5SC @ 1200 l ha^{-1} and neem oil @ 2.5 *l* ha^{-1} (T_4) recorded over all mean of 4.21 bugs 10 hills^{-1}. The highest gundhi bug numbers, 22.75 and 20.67 bugs 10 hills^{-1}, were recorded in untreated crop of rice during 2018 and 2019, respectively. Overall mean of two years observations followed almost similar trends of he lowest number of gundhi bug (1.96 bugs/ 10 hills^{-1}) with foliar spray of fipronil 5SC @ 1200 *l* ha^{-1}. This was followed by foliar spray of fipronil 5SC @ 1000 ml ha^{-1} at 85 DAT and neem oil @ 2.5 l ha^{-1}. It was interesting to note that need based foliar spray of fipronil 5SC @ 1200 l/ ha^{-1} or fipronil 5SC @ 1000 ml ha^{-1} alternated with botanical insecticides, the neem oil @ 2.5 l ha^{-1} significantly reduced the pest population and remained as effective as sole use of chemical insecticids.

Raghuraman *et al*., (2017) reported that a single spray of neem oil @ 3 per cent or neem kernel extract @ 5 per cent significantly reduced the population of gundhi bug. Application of quinalphos and phosphamidon reduced the

Table 1. Effect of botanical and chemical insecticides on the incidence of gundhi bug (*Leptocorisa spp.*) infesting aromatic rice (Var. BVS-1)

Treatment	No. of GB 10 hills^{-1} after 3rd spray														
	4 DAA			7 DAA			10 DAA			14 DAA			Overall mean		
	2018	2019	Pooled mean	2018	2019	Pooled mean	2018	2019	Pooled mean	2018	2019	Pooled mean	2018	2019	Pooled mean
T_1 A: Fipronil 5SC @1000 m ha^{-1}	4.67	3.33	4.00	2.67	3.00	2.83	1.67	2.33	2.00	3.67	2.00	2.83	3.17	2.67	2.92
B: Fipronil 5SC@1000 m ha^{-1}	(2.37)	(2.06)	(2.22)	(1.88)	(1.99)	(1.94)	(1.63)	(1.79)	(1.72)	(2.14)	(1.72)	(1.95)	(2.04)	(1.91)	(1.98)
T_2 A: Fipronil 5SC @ 1200 m ha^{-1}	4.00	2.33	3.17	2.00	1.33	1.67	2.00	0.67	1.33	2.00	1.33	1.67	2.50	1.42	1.96
B: Fipronil 5SC@1200 m ha^{-1}	(2.23)	(1.79)	(2.03)	(1.69)	(1.52)	(1.62)	(1.72)	(1.28)	(1.51)	(1.72)	(1.52)	(1.63)	(1.87)	(1.55)	(1.72)
T_3 A: Fipronil 5SC@1000 *l* ha^{-1}	6.33	6.00	6.17	5.00	5.67	5.33	4.67	4.33	4.50	8.33	6.33	7.33	6.08	5.58	5.83
B: Neem oil @ 2.5 *l* ha^{-1}	(2.70)	(2.64)	(2.67)	(2.44)	(2.56)	(2.51)	(2.38)	(2.29)	(2.34)	(3.05)	(2.70)	(2.88)	(2.66)	(2.56)	(2.61)
T_4 A: Fipronil 5SC @1200 *l* ha^{-1}	5.33	5.00	5.17	4.00	4.00	4.00	3.33	3.00	3.17	5.33	3.67	4.50	4.50	3.92	4.21
B: Neem oil @ 2.5 *l* ha^{-1}	(2.52)	(2.44)	(2.48)	(2.23)	(2.23)	(2.23)	(2.06)	(1.99)	(2.03)	(2.48)	(2.14)	(2.34)	(2.34)	(2.22)	(2.28)
T_5 A: Fipronil 5SC@1000 *l* ha^{-1}	7.00	6.33	6.67	6.00	5.33	5.67	6.00	5.00	5.50	10.00	5.33	7.67	7.25	5.50	6.38
B: Karanj oil @ 2.5 *l* ha^{-1}	(2.83)	(2.70)	(2.77)	(2.64)	(2.51)	(2.58)	(2.64)	(2.43)	(2.55)	(3.31)	(2.51)	(2.94)	(2.87)	(2.55)	(2.72)
T_6 A: Fipronil 5SC @1200 *l* ha^{-1}	5.67	5.67	5.67	5.00	3.33	4.17	4.00	3.33	3.67	6.00	4.00	5.00	5.17	4.08	4.63
B: Karanj oil @ 2.5 *l* ha^{-1}	(2.57)	(2.57)	(2.57)	(2.44)	(2.06)	(2.27)	(2.21)	(2.06)	(2.17)	(2.63)	(2.23)	(2.40)	(2.47)	(2.25)	(2.37)
T_7 A: Karanj oil @ 2.5 *l* ha^{-1}	14.67	14.67	14.67	12.33	11.33	11.83	15.33	15.33	15.33	19.67	17.00	18.33	15.50	14.58	15.04
B: Karanj oil @ 2.5 *l* ha^{-1}	(3.95)	(3.95)	(3.95)	(3.64)	(3.51)	(3.58)	(4.03)	(4.04)	(4.04)	(4.54)	(4.24)	(4.40)	(4.06)	(3.95)	(4.01)
T_8 A: Neem oil @ 2.5 *l* ha^{-1}	11.33	12.00	11.67	10.00	9.33	9.67	12.00	12.00	12.00	15.00	13.67	14.33	12.08	11.75	11.92
B: Neem oil @ 2.5 *l* ha^{-1}	(3.49)	(3.59)	(3.54)	(3.31)	(3.20)	(3.27)	(3.59)	(3.60)	(3.59)	(4.00)	(3.82)	(3.91)	(3.61)	(3.57)	(3.59)
T_9 Untreated control	20.00	18.67	19.33	21.67	20.00	20.83	23.67	21.00	22.33	25.67	23.00	24.33	22.75	20.67	21.71
	(4.58)	(4.43)	(4.51)	(4.76)	(4.58)	(4.67)	(4.96)	(4.69)	(4.83)	(5.16)	(4.90)	(5.03)	(4.87)	(4.66)	(4.76)
SE m(±)	(0.18)	(0.17)	(0.14)	(0.18)	(0.17)	(0.13)	(0.18)	(0.18)	(0.13)	(0.18)	(0.14)	(0.12)	(0.10)	(0.09)	(0.07)
CD (P=0.05)	(0.53)	(0.52)	(0.43)	(0.55)	(0.53)	(0.40)	(0.55)	(0.55)	(0.38)	(0.55)	(0.44)	(0.37)	(0.31)	(0.27)	(0.22)
CV (%)	(10.07)	(10.32)	(8.28)	(11.23)	(11.24)	(8.46)	(11.230	(11.81)	(7.97)	(9.85)	(8.71)	(7.00)	(5.86)	(5.45)	(4.25)

Figures in parentheses are square root transformed valueDAA:Day after application, DAT: Day after transplanting, GB: Gundhi bug
*Three foliar sprays with the respective treatment combination(s) were applied on need basis starting 1st spray at 25 DAT (days after transplanting) followed by 2nd and 3rd one at 55 and 85 DAT, respectively. As such, altogether spray of the each material contained in 'A' and 'B' were provided alternatively during the whole cropping season for protecting the crop against the major prevailing insect pests.

Table 2. Effect of botanical and chemical insecticides on grain yield of aromatic rice (Var. BVS-1)

Treatment Alternate spray of 'A & B' and so on.....on need based basis	Yield (q ha^{-1})			Additional yield over control (q ha^{-1})			Additional yield (%)		
	2018	2019	Pooled mean	2018	2019	Pooled mean	2018	2019	Pooled mean (%)
T_1 A: Fipronil 5SC @1000 m ha^{-1} B: Fipronil 5SC@1000 m ha^{-1}	44.97	47.30	46.13	12.74	12.60	12.67	28.33	26.64	27.44
T_2 A: Fipronil 5SC @ 1200 m ha^{-1} B: Fipronil 5SC@1200 m ha^{-1}	46.26	48.07	47.17	14.03	13.37	13.70	30.33	27.81	29.04
T_3 A: Fipronil 5SC@1000 m ha^{-1} B: Neem oil @ 2.5 *l* ha^{-1}	41.97	43.50	42.73	9.74	8.80	9.27	23.21	20.23	21.67
T_4 A: Fipronil 5SC @1200 m ha^{-1} B: Neem oil @ 2.5 *l* ha^{-1}	43.13	44.43	43.78	10.90	9.73	10.31	25.27	21.9	23.55
T_5 A: Fipronil 5SC@1000 m ha^{-1} B: Karanj oil @ 2.5 *l* ha^{-1}	39.52	41.23	40.38	7.29	6.53	6.91	18.45	15.84	17.11
T_6 A: Fipronil 5SC @1200 m ha^{-1} B: Karanj oil @ 2.5 *l* ha^{-1}	40.63	42.93	41.78	8.40	8.23	8.31	20.67	19.18	19.89
T_7 A: Karanj oil @ 2.5 *l* ha^{-1} B: Karanj oil @ 2.5 *l* ha^{-1}	35.03	37.70	36.37	2.80	3.00	2.90	8.69	7.96	7.97
T_8 A: Neem oil @ 2.5 *l* ha^{-1} B: Neem oil @ 2.5 *l* ha^{-1}	36.87	39.63	38.25	4.64	4.93	4.78	12.58	12.44	12.50
T_9 Untreated control	32.23	34.70	33.47	___	___	___	___	___	___
S.Em. (±)	2.17	2.10	1.29	___	___	___	___	___	___
CD (P=0.05)	6.57	6.33	3.89	____	___	___	___	___	___
C.V. (%)	9.39	8.61	5.42	____	___	___	___	___	___

*Three foliar sprays with the respective treatment combination(s) were applied on need basis starting 1st spray at 25 DAT (days after transplanting) followed by 2nd and 3rd one at 55 and 85 DAT, respectively. As such, altogether spray of the each material contained in 'A' and 'B' were provided alternatively during the whole cropping season for protecting the crop against the major prevailing insect pests

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population of gundhi bug up to 88.17 and 87.44 per cent, respectively (Verma and Gupta, 2010). Buprofezin 15 per cent + acephate 35 per cent WP @ 1500 ml ha^{-1} significantly suppressed the population of gundhi bug upto 1.66 per five sweeps (Choudhary and Raghuraman, 2014). The foliar spray with chemical insecticides remained more effective compared to botanicals insecticides against gundhi bugs and in turn realized significantly higher grains yield.

The data on grain yield of aromatic rice presented in table-2 showed highest pooled mean yield (47.17 q ha^{-1}) with three foliar sprays with fipronil 5 SC @ 1200 ml ha^{-1} applied at 25, 55 and 85 days after transplanting followed by three foliar sprays with fipronil 5 SC @ 1000 ml ha^{-1} (46.13 q ha^{-1}). Foliar sprays of fipronil 5 SC @ 1200 l ha^{-1} alternated with neem oil @ 2.5 l ha^{-1} gave 43.78 q ha^{-1} and fipronil 5 SC @ 1000 ml ha^{-1} alternated with neem oil @ 2.5 l ha^{-1} gave 42.73 q ha^{-1} rice yield. The lowest yield was obtained in the unprotected crop (33.47 q ha^{-1}). However all the treatments showed higher yield than the untreated control. The range of increase in yield over control among the treatments varied from 7.07 to 29.04 per cent.

This experimental finding is supported by Verma and Gupta (2001) who reported that the extent of per cent reduction of insect pest population in the respective treatment were responsible for the corresponding increase in yield of rice. Karanj and neem cake (applied as organic manure as basal application) was reported effective in reducing the incidence of major insect pests in Birsamati (an aromatic rice variety) and realized substantially higher grain yield of rice[4]. More recently, Jeer *et al*., (2017) conducted field experiment during *Kharif* and *Rabi* 2014-15 and reported highest grain yield with acephate 50 + imidacloprid 1.8 per cent SP @ 621.6 g a.i. ha^{-1} (50.14 & 51.02 q ha^{-1}) followed by acephate 50 + imidacloprid 1.8 per cent SP @ 518 g a.i. ha^{-1} (47.36 & 48.22 q ha^{-1}) with the lower incidence of the insect pest[2].

CONCLUSION

It was concluded that sole spray of fipronil 5 SC @ 1200 ml ha^{-1} or alternated spray of fipronil 5SC @ 1200 ml ha^{-1} and fipronil 5SC @ 1000 ml ha^{-1} provided excellent control of gundhi bug and increased grain yield upto 29 per cent. Alternate spray of fipronil with botanical insecticides also had significant effect. Judicious application of recommended insecticides (fipronil, neem oil or karanj oil) which proved next best in terms of reduction of gundhi population and percent increase in rice grain yield over the control could effectively be used in reducing gundhi bug infestation in rice.

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Short communication

Effectiveness of botanical insecticides in the management of rice gall midge (*Orseolia oryzae* Wood Mason)

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ABSTRACT

The experiment conducted to evaluate the efficacy of ten botanical insecticides against rice gall midge, carried out at Rice research farm, B.A.U, Kanke, Ranchi, Jharkhand during kharif, 2016 and kharif, 2017, showed that the crop treated with (Aza.0.03% EC) recorded the lowest incidence of silver shoot (1.76% SS) and proved superior over all the insecticides tested during both the seasons of experimentation. It was followed by rynaxypyr 20 SC (2.54% SS), dinotefuran 20 SG (2.80% SS) and neem baan (Aza.1% EC) (3.14% SS). Similar result were found with the overall pooled mean values. The untreated control plot recorded highest incidence of silver shoot (12.05%). However, all the botanical insecticides remained significantly superior over the untreated crop of rice.

Key words: *Gall midge, Incidence, Botanical insecticides, Management and environmental concern*

Rice (*Oryza sativa* L.) is the staple food for over half the world's population. It is widely distributed all over the world with a highly growing in Asia. Out of nearly 1000 insect pest species recorded on paddy, over two dozen insects & mites are the key pests in different rice ecologies in India (Prakash *et al*., 2000). Amongst these, the rice gall midge (*Orseolia oryzae* Wood Mason) is one of the most important pests capable of causing huge losses leading to low productively in Jharkhand. The yield loss in the state of Jharkhand under the favorable agro-climatic conditions ranges from 10-25 per cent (Prasad and Prasad, 2006). Seasonal occurrence of gall midge commences from 2nd week of August and continue till October with the peak activity observed in the last week of September. Parasite activity of *Platygaster oryzae* that started in the last week of August at a minimum host infestation of 2.0 per cent reached to its peak during second week of October resulting in 42 per cent of gall midge mortality (Mardi et al., 2009). In the past overview, the emphasis of using synthetic insecticides for the control of gall midge was found unsafe to environment, human health, biodiversity and the natural enemy of the rice pest. Besides, the conventional products are expensive and dangerous to users. Much attention has therefore been given to use of botanicals pesticides in the recent decades (Sahani et al. 2019). Botanicals leave greatest impact on agricultural regions in developing countries, where the source plants are readily available, It is applied from the ancient time when or where farmers may not be able to afford synthetic insecticides and the traditional use of plants and plant derivatives for protection of stored products is long established (Isman 2020; Reddy 2021). Pyrethrum and neem are well established commercially. Pesticides, based on plant essential oils have recently entered the marketplace and the use of rotenone appears to be waning. Botanical insecticides viz., neem oil (5%) and neem based products applied either as foliar or mixed with fertilizers has significantly reduced the incidence of gall midge in rice (Mahalingam 1984; Parija 1988). Therefore, selective bio-pesticides that aid in managing gall midge without harming the natural enemies is need of the day. Keeping this in view, the efficacy of some of the botanical insecticides, considered relatively environment friendly, were evaluated as an alternative to synthetic insecticides in the management of gall midge under field conditions.

The experiment using rice variety IR-64 Drt at Rice research farm, B.A.U, Kanke, Ranchi, Jharkhand during kharif, 2016 and kharif, 2017 was conducted in Randomized block design with three replications the spacing between plant to plant and row to row was kept at 15 and 20 cm, respectively Data on percent incidence of silver shoot (SS) caused by gall midge were recorded before one day & 4th, 7th

and 10th days after insecticidal application at 45 DAT (days after transplanting) using the below mentioned formula.

$$SS\ (\%) = \frac{\text{Total no. of silver shoot (SS) in 10 hills}}{\text{Total no. of tillers (SS + healthy tillers) in 10 hills}} \times 100$$

The results summarized in table 1 showed that the overall mean values of silver shoot recorded on three dates of observations (4th, 7th & 10th DAA) during 2016 and 2017, were significant. Minimum damage in terms of silver shoot (1.76%) was found in achook (Aza.0.03% EC) showing superiority over all other insecticides tested followed by rynaxypyr 20 SC (2.54% SS), dinotefuran 20 SG (2.8% SS) and neem baan, (Aza.1 per cent EC (3.14% SS). Similar results (2.77% SS) with Achook (Aza. 0.03% EC) were observed in the subsequent season again which was superior among all the test insecticides. It was at par with rynaxypyr 20 SC (3.60% SS) followed by dinotefuran 20 SG (3.87% SS), neem baan (Aza.1% EC) (4.23% SS) and neemazal (Aza.1% EC) (4.65% SS). The highest percent incidence of silver shoot (11.46 and 12.45%) was observed during 2016 and 2017, respectively in the untreated crop.

The effect of botanical insecticides on over all mean of silver shoot (SS) recorded at three dates of observations (4th, 7th & 10th DAA) showed significant results. Achook (Aza.0.03% EC) recorded the lowest incidence of silver shoot (2.26% SS). It was followed by rynaxypyr 20 SC (3.07% SS) and dinotefuran 20 SG (3.33% SS). The highest incidence of silver shoot (12.05%) was observed in the untreated control plot. However, all the botanical insecticides remained significantly superior over the untreated crop of rice the. These findings are in close agreement with the results of Mahalingam (1984). Neem oil (5.0%) used as foliar spray could cause significant reduction of gall midge infestation in rice. Similar observations were reported in AICRIP 2015, 2016. Among the botanicals used, the nemazal recorded the

Table 1. Effect of botanical insecticides on the incidence of gall midge (*Orseolia oryzae* Wood mason) in terms of silver shoots (SS %)

				Silver shoot (SS) caused by gall midge, recorded after 2nd spray* at											
				4 DAA			7 DAA			10 DAA			Overall mean		
S.N	Treatment	Formulations (a.i.) or Aza content %	Dose g or ml/ha	2016	2017	Pooled Mean	2016	2017	Pooled Mean	2016	2017	Pooled Mean	2016	2017	Pooled Mean
T1	Neem Baan	1.00	1000 ml	2.21	2.62	2.42	2.98	3.35	3.16	4.24	6.71	5.48	3.14	4.23	3.68
				(8.32)	(9.11)	(8.72)	(9.25)	(9.95)	(9.60)	(11.6)7	(14.72)	(13.19)	(9.91)	(11.55)	(10.73)
T2	Neemazal	1.00	1000 ml	2.63	3.06	2.84	3.32	3.70	3.51	4.72	7.20	5.96	3.55	4.65	4.10
				(9.18)	(9.93)	(9.55)	(10.08)	(10.72)	(10.40)	(12.37)	(15.32)	(13.84)	(10.67)	(12.23)	(11.45)
T3	Nimbecidine	0.03	2500 ml	3.36	3.82	3.59	3.65	4.05	3.85	5.71	8.23	6.97	4.24	5.37	4.81
				(10.40)	(11.11)	(10.76)	(10.68)	(11.30)	(10.99)	(13.68)	(16.46)	(15.07)	(11.70)	(13.18)	(12.44)
T4	Multineem	0.03	2500 ml	4.62	5.12	4.87	4.13	4.54	4.33	6.25	8.79	7.52	5.00	6.15	5.57
				(12.30)	(12.97)	(12.64)	(11.58)	(12.17)	(11.88)	(14.35)	(17.05)	(15.70)	(12.80)	(14.21)	(13.50)
T5	Neemoil	-	2500 ml	4.87	5.38	5.12	4.29	4.71	4.50	6.93	9.50	8.22	5.36	6.53	5.95
				(12.65)	(13.31)	(12.98)	(11.80)	(12.39)	(12.09)	(15.16)	(17.78)	(16.47)	(13.27)	(14.66)	(13.97)
T6	Achook	0.03	2500 ml	1.01	1.39	1.20	1.70	1.94	1.82	2.56	4.97	3.77	1.76	2.77	2.26
				(5.51)	(6.56)	(6.04)	(6.61)	(7.07)	(6.84)	(8.76)	(12.41)	(10.58)	(7.17)	(9.13)	(8.15)
T7	Pongamia oil	-	2500 ml	3.17	3.62	3.39	3.49	3.88	3.68	5.17	7.67	6.42	3.94	5.06	4.50
	(karanj oil)			(10.10)	(10.81)	(10.46)	(10.43)	(11.05)	(10.74)	(13.00)	(15.86)	(14.43)	(11.27)	(12.78)	(12.02)
T8	Dinotefuran	20 SG	200 g	1.90	2.31	2.10	2.70	3.06	2.88	3.79	6.24	5.02	2.80	3.87	3.33
				(7.68)	(8.51)	(8.10)	(8.72)	(9.46)	(9.09)	(10.9)3	(14.10)	(12.52)	(9.27)	(10.98)	(10.13)
T9	Rynaxypyr	20SC	150 ml	1.76	2.16	1.96	2.45	2.81	2.63	3.41	5.84	4.63	2.54	3.60	3.07
				(7.35)	(8.21)	(7.78)	(8.19)	(8.99)	(8.59)	(10.24)	(13.54)	(11.89)	(8.76)	(10.54)	(9.65)
T10	Untreated	Water spray	500 lit.	10.11	10.81	10.46	11.69	12.90	12.29	12.57	14.24	13.41	11.46	12.65	12.05
	control			(18.10)	(18.76)	(18.43)	(19.92)	(20.98)	(20.45)	(20.33)	(21.48)	(20.90)	(19.51)	(20.49)	(20.00)
	SEm±			(0.59)	(0.59)	(0.37)	(0.71)	(0.68)	(0.44)	(0.70)	(0.84)	(0.51)	(0.46)	(0.51)	(0.31)
	CD 5%			(1.74)	(1.76)	(1.06)	(2.11)	(2.01)	(1.25)	(2.07)	(2.51)	(1.46)	(1.35)	(1.51)	(0.89)
	CV %			(9.99)	(9.38)	(9.67)	(11.47)	(10.29)	(10.86)	(9.23)	(9.22)	(9.27)	(6.91)	(6.77)	(6.85)

Figures under the parenthesis are angular transformed values. SS- Silver shoot caused by gall midge
DAT-Days after transplanting; DAA-Days after application of insecticidal treatment
*2nd foliar spray of the insecticidal treatments was applied at 40 DAT.

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lowest mean infestation (8.4%SS). However, the efficacy was on par with insecticides and significantly superior to control (14.70%SS). This finding also endorsed the results of the present studies. Similarly, botanical insecticides like gardi leaves, neem and karanj cake used to manage rice gall midge in the varying doses of 0.5, 1.0 and 2.5 t ha-1 proved significantly effective in reducing the incidence of rice gall midge and yellow stem borer infestation resulting in substantially higher yield (Borkar and Sarode 2008; Prasad et al., 2018).

According to Seni 2019; Sahani and Mondal 2020, the experimental results showed that both botanical and synthetic insecticides are effective in minimizing the infestation of gall midge as compared to the untreated control. The gall midge infestation in terms of silver shoot in insecticide treated plots ranged from 5.55 to 6.09 per cent in 2017 as against 10.76 per cent in the control. Whereas in 2018, the silver shoot ranged from 15.03 to 20.42 per cent as against 34.22 per cent in untreated control. All the botanical treatments reduced gall midge infestation from 7.42 to 9.13 per cent in *kharif* 2017 and 15.62 to 24.19 per cent in 2018, Among them the cedarwood oil @ 1000 ml ha^{-1} showed better efficacy than other treatments (7.42% and 15.62% in *kharif* 2017 and 2018, respectively). Neem baan (Aza. 1.0 per cent EC) @ 1000 ml ha^{-1} was most effective (4.71% SS) with maximum net profit with reducing SS (%). Neem baan (Aza. 1.0% EC) could be responsible for realization of the highest grains yield (34.03 q ha^{-1}). However, chlorpyriphos (@ 2000 ml ha^{-1}) when compared in the context of yield of grains took the lead with (38.95 q ha^{-1} Alka and Rabindra 2020).

CONCLUSION

It was concluded that the treatment of rice crop with Achook (Aza.0.03% EC) that recorded lowest incidence of silver shoot (1.76% SS) and is relatively environment friendly could satisfactorily be used to protect rice crop from gall midge infestation.

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Bio-efficacy of botanicals against pulse beetle, *Callosobruchus chinensis* (L.) in stored chickpea

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ABSTRACT

The laboratory trial revealed that neem leaf powder (6%) followed by melia (6%) and datura leaf powder (8%), recording 31.86, 40.17 and 46.06 per cent mean seed damage and 6.27, 8.61 and 10.84 per cent mean weight loss, respectively proved the best in protecting chickpea from pulse beetle under storage. Neem leaf powder also registered highest Cost-Benefit Ratio (1:2.09) followed by melia leaf powder (1:1.97). Although the synthetic insecticide showed the highest C: B ratio (1:6.17), the botanicals being cheap, locally available, biodegradable, non-hazardous to human as well as animal life and the environment could be considered as seed protectants against storage pests.

Key words: *Callosobruchus chinensis*, Chickpea, Botanicals, Weight loss, Seed damage, Cost benefit ratio.

Chickpea (*Cicer arietinum* L.), an important legume crop, grown and consumed in India since long, suffers qualitative and quantitative losses due to bruchid's (pulse beetle) attack at post-harvest level. The pest attacks the crop both under the field and storage conditions. However, the attack is more in the storage (Bhalla *et al*., 2008). It was found to damage 50-60 per cent of stored grains even after 6 months of traditional storage (Caswell, 1973). In severe cases of infestation, the damage can reach up to 100 per cent (Pruthi and Singh, 1950) and the infested grains become unfit for human consumption. The stored grain insects are controlled by the application of fumigants and synthetic insecticides. However, the indiscriminate use of these leads to many problems like insect resurgence, environmental pollution, toxic residue of food grains and increased cost of application (Fishwick, 1988). There is therefore need to replace these with some suitable alternatives. Considering these, the bio-efficacy of some botanicals against pulse beetle in chickpea was studied and their cost benefit ratio was calculated.

MATERIALS AND METHODS

The study was carried out in the Post Graduate laboratory of Department of Entomology, Uttar Banga Krishi Viswavidyalaya, Pundibari, Coochbehar, West Bengal during 2018-2020. The mother culture of *Callosobruchus chinensis* was raised on the chickpea seed (variety: Anuradha) collected from the farm at Pulses and Oilseeds Research Station (PORS), Department of Agriculture, Government of West Bengal, Berhampore, Murshidabad, West Bengal. The seeds after sterilization in hot air oven at 80ÚC for 2 hrs were disinfected with 1 per cent formalin to destroy insect-pests and micro-organisms present in the seed. The *Callosobruchus chinensis* adults collected with the aspirator were later released in the glass jar having sterilized seeds. The mouth of the glass jar was covered with muslin cloth and tied with rubber band. After 7 days, the old adults were removed from the container and emergence of the fresh adults was awaited. Newly emerged adults were used for the experiments.

The different plant parts of neem (*Azadirachta indica* A. Juss), dharek (*Melia azedarach* L.), datura (*Datura stramonium* L.) and tulsi (*Ocimum sanctum* L.) were used for assessing their efficacy against pulse beetle, *Callosobruchus chinensis*. The fresh leaves of neem, dharek, datura, tulsi, bark of neem and dharek and the ripened fruits and matured perianth of dhatura and tulsi were used. These plant parts were dried under sunlight and powdered in mixer grinder. Fine powders

Table 1: Treatment details for the management of pulse beetle, *Callosobruchus chinensis*

Treatment	Name of the botanicals	Parts used	Conc. (%)
T1	Neem *(Azadirachta indica)*	Leaves	6
T2	Neem (*Azadirachta indica)*	Bark	2
T3	Neem *(Azadirachta indica)*	Leaves + Bark	6 + 2
T4	Dharek *(Melia azedarach)*	Leaves	6
T5	Dharek *(Melia azedarach)*	Bark	2
T6	Dharek *(Melia azedarach)*	Leaves + Bark	6 + 2
T7	Datura *(Datura stramonium)*	Leaves	8
T8	Datura *(Datura stramonium)*	Seeds	8
T9	Datura *(Datura stramonium)*	Leaves + Seeds	8 + 8
T10	Tulsi *(Ocimum sanctum)*	Leaves	5
T11	Tulsi *(Ocimum sanctum)*	Seeds	2
T12	Tulsi *(Ocimum sanctum)*	Leaves + Seeds	5 + 2
T13	Standard Check (Deltamethrin 2.5 WP)	-	0.04
T14	Untreated control	-	-

passed through 60 mesh sieves at different concentrations (table 1) were utilized for the bio-efficacy study (Kinchi *et al*., 2017).

The bio-efficacy of botanicals was examined in terms of percent weight loss and percent chickpea seeds damaged due to pulse beetle. This was studied by using 25 g of sterilized chickpea seeds treated with different botanical treatments in plastic containers. Five pairs of adult pulse beetles were released in each container. Observations on percent seed weight loss were recorded at weekly intervals for about 6 months until complete weight loss in case of untreated control. The no. of infested and uninfested grains as well as the weight of infested and uninfested grains was recorded with the help of electronic weighing balance. The percent loss in seed weight and number of seeds damaged was worked out by using the following formulae (Adams and Schulton, 1978) Pathania, 2013).

$$\% \text{ Weight loss} = \frac{[(UND) - (DNU)]}{[U(ND + NU)]} \times 100$$

$$\text{Percent seed damage} = \frac{\text{Number of damaged seeds}}{\text{Total number of seeds used}} \times 100$$

Where u, NU, D and ND are the weight of uninfested grains (g), number of uninfested grains, weight of infested grains (g) and the number of infested grains, respectively.

The cost: benefit ratio of botanicals compared to conventional insecticides was studied considering the cost involved in collection, drying and processing of the botanicals, of seed used and lost due to pest's damage and the pesticidal treatment. Labour wages were computed as per the rate of unskilled agricultural labour wages of the Government of West Bengal during 2019. The net benefit was obtained by subtracting the total cost of plant protection from total income. Benefit over the control for each treatment was calculated by subtracting the income of the control treatment from that of each treatment. Cost benefit ratio of the treatments were derived by comparing the cost of plant protection against benefit over untreated control (UC). The data were statistically analysed using Duncan multiple range test (DMRT).

RESULTS AND DISCUSSION

Efficacy of different botanicals in terms of per cent reduction in seed weight and seed number

The data (table-2) showed that the percentage loss in seed weight during March-June, 2019 increased from fourth week onwards with the emergence of adults from the seeds and continued up to 14th week when tell all the seeds in untreated control were damaged. The lowest percent loss in seed weight (6.57%) was recorded when the seeds were treated with 6 per cent neem leaf powder (T1) after 14th week and this was found most effective among all the botanicals and also statistically different from the other treatments. However, no loss in chickpea seed weight was found in standard check (deltamethrin). Melia leaf powder @6 per cent (T4) and datura leaf powder @8 per cent (T7) recording 8.97 and 11.82 per cent losses in seed weight on 14th week were the next best. All the treatments were significantly different from each other and performed statistically better than the untreated control (81.78% weight loss). The experiment repeated during July-October, 2019 confirmed the superiority of neem powder over other treatments. It recorded minimum percent weight loss (6.45%) during 15th week followed by melia @6 per cent (T4) leaf powder (8.77%) and with datura leaf of powder 8 per cent (10.50%). All the treatments in July-October trials were also significantly different with each other and performed better than the untreated control.

The pooled mean of the two different observations (March-June and July-October, 2019) shown in table 3 concluded that neem powder at 6 per cent concentration was the best recording lowest percentage weight loss during different weeks (1.15 to 6.27%) in (T1). All the treatments resulted significantly less seed damage than the untreated control. However, the standard check, deltamethrin recorded zero per cent loss. Melia leaf powder with 1.72 to 8.61 per cent weight loss and datura leaf powder at 8 per cent with 2.59 to 10.84 per cent loss were rated the next best treatments after neem powder.

These findings are in agreement with Jadhav *et al*. (2015) & Khalequzzaman and Osman (2009) who reported that dried leaf powder of neem was much effective in reducing the weight loss caused by pulse beetle. Kaur (2017) also reported that neem leaf powder and melia leaf powder at 6% concentration caused weight loss of 7.67 and 9.83 per cent, respectively and were effective in preventing the weight loss. Besides, Tabu *et al.* (2012) reported that datura leaf powder was effective against pulse beetle.

Efficacy of different botanicals in terms of reduction in percent seed damage

The per cent chickpea seed damaged by *Callosobruchus chinensis* under different botanicals treatments during March-June and July-October, 2019 trials, presented in the table– 3, revealed lowest percentage of seed damage in case of neem leaf powder applied at 6 per cent (T1) during 4th to 14th week (7.08 to 31.86 per cent in March-June and 3.54 to 32.59 per cent in July-October 2019 trials), respectively. Melia leaf

Table 2: Bio-efficacy of different botanicals in terms of loss in chickpea seed weight due to *Callosobruchus chinensis* during March- June and July-October 2019.

Treatments	Dose	Percent loss in seed weight during 2019							
		March-June				July-October			
		4th week	8th week	12th week	14th week	4th week	8th week	12th week	14th week
T1-neem LP #	6%	1.46^{j}*	3.97^{g}	5.83^{gh}	6.57^{hi}	0.84^{ij}	3.98^{e}	5.39^{hi}	6.45^{k}
T2-neem BP	2%	8.19^{bc}	15.32^{bc}	25.39^{b}	39.04^{bc}	6.93^{bc}	13.68^{b}	19.90^{bc}	29.74^{c}
T3-neem LP+BP	6%+2%	3.87^{gh}	6.72^{efg}	11.13^{efg}	14.41^{fghi}	2.95^{g}	5.52^{e}	9.03^{fgh}	12.91^{hi}
T4-melia LP	6%	1.93^{ij}	4.60^{fg}	7.53^{fg}	8.97^{ghi}	1.50^{hi}	4.03^{e}	6.79^{gh}	8.77^{jk}
T5-melia BP	2%	9.34^{b}	17.39^{b}	25.60^{b}	44.58^{b}	7.74^{b}	15.16^{b}	22.03^{b}	40.49^{b}
T6-melia LP+BP	6%+2%	5.71^{ef}	8.63^{ef}	16.90^{cde}	21.22^{defgh}	4.78^{f}	7.43^{de}	11.91^{efg}	17.19^{fg}
T7-datura LP	8%	3.14^{hi}	5.71^{fg}	9.67^{fg}	11.82^{fghi}	2.04^{h}	4.76^{e}	8.50^{fgh}	10.50^{ij}
T8-datura SP	8%	6.58^{de}	11.06^{cde}	18.86^{cd}	26.14^{cdef}	5.76^{de}	9.59^{cd}	14.64^{cdef}	20.36^{ef}
T9-datura LP+SP	8%+8%	4.67^{fg}	7.58^{efg}	13.25^{def}	17.82^{efgh}	3.20^{g}	6.57^{de}	9.68^{fgh}	14.83^{gh}
T10-tulsi LP	5%	7.14^{cd}	13.64^{bcd}	21.61^{bc}	31.02^{bcde}	6.19^{cd}	11.82^{bc}	17.78^{bcde}	21.62^{e}
T11-tulsi SP	2%	7.32^{cd}	14.87^{bc}	22.82^{bc}	35.25^{bcd}	6.44^{cd}	13.45^{b}	18.55^{bcd}	25.80^{d}
T12-tulsi LP+SP	5%+2%	6.09^{de}	10.16^{de}	17.56^{cd}	24.23^{cdefg}	5.21^{ef}	8.99^{cd}	13.75^{def}	19.09^{ef}
T13-deltamethrin 2.5 WP (standard check)	0.04%	0.00^{k}	0.00^{h}	0.00^{h}	0.00^{i}	0.00^{j}	0.00^{f}	0.00^{i}	0.00^{l}
T14- Untreated control	-	11.76^{a}	24.19^{a}	62.29^{a}	81.78^{a}	9.55^{a}	23.31^{a}	57.29^{a}	79.48^{a}
SEm (±)	-	0.45	1.36	2.04	4.93	0.30	1.08	1.89	1.22
CD (at 5%)	-	1.30	3.94	5.90	14.28	0.87	3.12	5.47	3.53
CV (%)	-	14.08	22.93	19.10	32.95	11.52	20.33	21.26	9.61

*Data followed by same letters are statistically at par by DMRT, # LP- leaf powder; BP- bark powder; SP- seed powder

Table 3: Pooled mean data on bio-efficacy of different botanicals in terms of chickpea seed weight loss (%) due to *Callosobruchus chinensis* (4th to 14th week of March-June and July-Oct 2019 trials)

Treatments	Dose	Pooled per cent seed weight loss due to pulse beetle			
		4th week	8th week	12th week	14th week
T1-neem LP #	6%	1.15^{i}	3.98^{i}	5.61^{i}	6.27^{ij}
T2-neem BP	2%	7.56^{c}	14.50^{bc}	22.64^{b}	31.59^{bc}
T3-neem LP+BP	6%+2%	3.41^{g}	6.12^{ghi}	10.08^{gh}	12.91^{ghi}
T4-melia LP	6%	1.72^{i}	4.32^{i}	7.16^{hi}	8.61^{hi}
T5-melia BP	2%	8.54^{b}	16.28^{b}	23.81^{b}	35.86^{b}
T6-melia LP+BP	6%+2%	5.25^{f}	8.03^{efg}	14.41^{ef}	18.62^{efg}
T7-datura LP	8%	2.59^{h}	5.24^{hi}	9.09^{ghi}	10.84^{hi}
T8-datura SP	8%	6.17^{de}	10.32^{de}	16.75^{cde}	22.50^{def}
T9-datura LP+SP	8%+8%	3.94^{g}	7.07^{fgh}	11.47^{fg}	15.63^{fgh}
T10-tulsi LP	5%	6.66^{d}	12.73^{cd}	19.69^{bcd}	26.12^{cde}
T11-tulsi SP	2%	6.88^{cd}	14.16^{bc}	20.69^{bc}	29.61^{bcd}
T12-tulsi LP+SP	5%+2%	5.65^{ef}	9.57^{ef}	15.66^{de}	20.70^{ef}
T13-deltamethrin 2.5 WP (standard check)	0.04%	0.00^{j}	0.00^{j}	0.00^{j}	0.00^{j}
T14- Untreated control	-	10.65^{a}	23.75^{a}	59.79^{a}	76.87^{a}
SEm (±)	-	0.27	0.87	1.39	2.50
CD (at 5%)	-	0.76	2.46	3.93	7.07
CV (%)	-	13.18	21.85	20.10	27.09

(*Data followed by same letters are statistically at par by DMRT).
(# LP- leaf powder; BP- bark powder; SP- seed powder)

powder @6 per cent (T4) and datura leaf powder @8 per cent (T7) resulting 9.61 to 41.14 per cent and 13.94 to 48.48 per cent seed damage, respectively during March-June and 6.76 to 40.39 and 10.18 to 44.55 per cent in July-October 2019 were found the next best effective botanicals.

The pooled data (table–5) showed minimum per cent damaged seeds (5.31 to 31.86 %) in case of neem leaf powder (6%), which was most effective in reducing the seed damage compared to all botanicals used in the experiment except insecticide deltamethrin. The best treatments registered were the melia at 6 per cent and datura leaf powder at 8 per cent recording as 8.19 to 40.17 and 12.05 to 46.06 per cent, respectively. Least damaged seeds per cent damage of the chickpea seeds with neem and melia leaf powder may be attributable to their high anti-feedant activity against pulse beetle (Dhaliwal and Koul, 2007 and Hossain et al., 2014). The findings are in accordance with Yadava and Bhatnagar (1987) who reported that mixing of dried neem (*Azadirachta indica)* leaves with chickpea grains under storage effectively protects the stored grain seeds from pulse beetle.

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Table 4: Bio-efficacy of different botanicals in terms of per cent seed damaged by *Callosobruchus chinensis* on chickpea seeds during March- June and July-October 2019

Treatments Done (%)	Dose	Per cent loss in seed number during, 2019							
		March-June				July-October			
		4th week	8th week	12th week	14th week	4th week	8th week	12th week	14th week
T1- neem LP #	6%	7.08 *(15.43)^{l}	17.41 (24.66)^{l}	29.06 (32.61)^{k}	31.86 (34.36)^{k}	3.54 (10.83)^{k}	15.04 (22.82)^{l}	27.58 (31.68)^{j}	32.59 (34.81)^{m}
T2-neem BP	2%	39.85 (39.14)^{c}	64.39 (53.40)^{bc}	80.46 (63.81)^{bc}	87.73 (69.72)^{bc}	32.88 (34.99)^{c}	57.43 (49.28)^{c}	77.27 (61.53)^{c}	85.00 (67.21)^{c}
T3-neem LP+BP	6%+2%	20.18 (26.68)^{i}	32.88 (34.97)^{ij}	49.03 (44.44)^{hi}	56.50 (48.76)^{hi}	17.94 (25.05)^{h}	25.86 (30.55)^{j}	41.70 (40.22)^{h}	51.42 (45.82)^{j}
T4-melia LP	6%	9.61 (18.06)^{k}	24.17 (29.42)^{k}	37.09 (37.52)^{j}	41.14 (39.90)^{jk}	6.76 ƨ(15.03)^{j}	20.42 (26.85)^{k}	33.78 (35.53)^{i}	40.39 (39.46)^{l}
T5-melia BP	2%	44.28 (41.72)^{b}	68.16 (55.69)^{b}	83.72 (66.28)^{b}	90.91 (72.87)^{b}	39.30 (38.82)^{b}	61.72 (51.79)^{b}	81.08 (64.28)^{b}	92.09 (73.69)^{b}
T6-melia LP+BP	6%+2%	27.70 (31.75)^{g}	42.01 (40.39)^{gh}	61.64 (51.76)^{fg}	68.65 (56.02)^{fg}	24.20 (29.46)^{f}	36.38 (37.10)^{h}	55.40 (48.10)^{f}	65.45 (54.00)^{h}
T7-datura LP	8%	13.94 (21.90)^{j}	27.27 (31.44)^{jk}	43.79 (41.43)^{ij}	48.48 (44.13)^{ij}	10.15 (18.57)^{i}	22.27 (28.16)^{k}	39.24 (38.79)^{h}	44.55 (41.87)^{k}
T8-datura SP	8%	33.18 (35.16)^{ef}	52.25 (46.30)^{ef}	69.22 (56.34)^{e}	76.57 (61.15)^{def}	28.68 (32.38)^{d}	42.04 (40.42)^{f}	63.51 (52.85)^{e}	73.58 (59.07)^{f}
T9-datura LP+SP	8%+8%	24.74 (29.81)^{h}	37.86 (37.96)^{hi}	55.81 (48.35)^{gh}	63.20 (52.69)^{gh}	21.57 (27.67)^{g}	32.58 (34.81)^{i}	47.81 (43.74)^{g}	59.43 (50.43)^{i}
T10-tulsi LP	5%	34.99 (36.26)^{de}	57.16 (49.13)^{de}	72.85 (58.64)^{de}	80.54 (64.04)^{cde}	30.17 (33.31)^{d}	45.10 (42.19)^{e}	68.18 (55.68)^{d}	76.62 (61.08)^{e}
T11-tulsi SP	2%	37.82 (37.94)^{cd}	60.84 (51.27)^{cd}	77.13 (61.53)^{cd}	84.30 (66.90)^{bcd}	32.14 (34.54)^{c}	51.87 (46.07)^{d}	72.05 (58.11)^{d}	80.72 (63.96)^{d}
T12-tulsi LP+SP	5%+2%	30.33 (33.41)^{fg}	46.85 (43.19)^{fg}	66.16 (54.45)^{ef}	73.27 (59.00)^{ef}	25.98 (30.64)^{e}	39.04 (38.67)^{g}	57.96 (49.58)^{f}	68.62 (55.94)^{g}
T13-deltamethrin 2.5 WP (Standard Check)	0.04%	0.00 (1.28)^{m}	0.00 (1.28)^{m}	0.00 (1.28)^{l}	0.00 (1.28)^{l}	0.00 (1.28)^{l}	0.00 (1.28)^{m}	0.00 (1.28)^{k}	0.00 (1.28)^{n}
T14- Untreated control	-	51.9 (46.09)^{a}	77.78 (61.89)^{a}	95.47 (77.77)^{a}	98.97 (84.21)^{a}	42.25 (40.54)^{a}	71.34 (57.64)^{a}	93.86 (75.76)^{a}	98.68 (83.47)^{a}
SEm (±)	-	0.67	1.39	1.41	2.02	0.40	0.53	0.87	0.54
CD (at 5%)	-	1.93	4.03	4.08	5.85	1.17	1.55	2.51	1.57
CV (%)	-	3.90	6.02	4.90	6.48	2.61	2.55	3.19	1.79

*Figures in parenthesis are angular transformed values. Parentheses followed by same letters are statistically at par by DMRT.

Economics (cost and benefit ratio) of different botanicals

The cost-benefit ratio of different treatment against pulse beetle (Table–6) indicated highest ratio under pesticidal treatment (1: 6.17). Among the botanicals, the neem leaf powder with (1: 2.09) followed by melia and datura leaf powder with 1.1.97 and (1: 1.38), respectively registered the best C:B ratio. The cost: benefit ratio is an indicator of relative economic performance of the treatments used in the experiment. Although, the standard check involving pesticides register highest cost benefit ratio as compared to different treatments, their indiscriminate use are highly hazardous to mankind, animals and the environment. Hence, the botanicals, because of being non-persistent, safer to users, consumers, animals and the environment and besides being cost effective appears to be the best alterative to pesticides.

In several parts of the developing world, resource limited farmers don't have financial capacity to purchase synthetic insecticides, but they have adequate and free labours to prepare and use botanicals. As such local farmers still find the use of locally prepared botanicals more convenient (Amoabeng *et al*., 2014).

CONCLUSION

It is concluded that the mixing of 6 per cent neem leaf powder with chickpea seeds before storage, that recorded least per cent damage in seed weight, seed number and was cost effective, could be considered as an alternative to synthetic insecticides in protecting chick pea seeds from pulse beetle under storage conditions.

Table 5: Pooled mean data on bio-efficacy of different botanicals in terms of seed damage (%) by *Callosobruchus chinensis* on chickpea seeds (Pooled data from 4th to 14th week of March-June and July-Oct, 2019

Treatments dose (%)		Per cent seed damage by Pulse beetle, *C. chinensis* after			
		4th week	**8th week**	**12th week**	**14th week**
T_1- neem LP #	6%	5.31 (13.13)^{k}	16.23 (23.74)^{l}	28.32 (32.14)^{k}	31.86 (34.36)^{l}
T_2-neem BP	2%	36.37 (37.06)^{c}	60.91 (51.34)^{c}	78.87 (62.67)^{c}	85.16 (67.55)^{bc}
T_3-neem LP+BP	6%+2%	19.06 (25.87)^{h}	29.37 (32.76)^{j}	45.37 (42.33)^{i}	52.17 (46.26)^{i}
T_4-melia LP	6%	8.19 (16.55)^{j}	22.30 (28.14)^{k}	35.44 (36.53)^{j}	40.17 (39.33)^{k}
T_5-melia BP	2%	41.79 (40.27)^{b}	64.94 (53.74)^{b}	82.40 (65.28)^{b}	88.20 (70.25)^{bc}
T_6-melia LP+BP	6%+2%	25.95 (30.61)^{f}	39.20 (38.74)^{h}	58.52 (49.93)^{g}	65.45 (54.06)^{g}
T_7-datura LP	8%	12.05 (20.24)^{i}	24.77 (29.80)^{k}	41.52 (40.11)^{i}	46.06 (42.74)^{j}
T_8-datura SP	8%	30.93 (33.77)^{d}	47.15 (43.36)^{f}	66.37 (54.60)^{f}	73.87 (59.34)^{ef}
T_9-datura LP+SP	8%+8%	23.16 (28.74)^{g}	35.2**2** (36.38)^{i}	51.81 (46.05)^{h}	59.88 (50.73)^{h}
T_{10}-tulsi LP	5%	32.58 (34.79)^{d}	51.13 (45.66)^{e}	70.52 (57.16)^{e}	77.45 (61.81)^{de}
T_{11}-tulsi SP	2%	34.98 (36.24)^{c}	56.36 (48.67)^{d}	74.59 (59.82)^{d}	81.54 (64.74)^{cd}
T_{12}-tulsi LP+SP	5%+2%	28.16 (32.03)^{e}	42.95 (40.93)^{g}	62.06 (52.02)^{g}	69.22 (56.41)^{fg}
T_{13}-deltamethrin 2.5 WP (Standard Check)	0.04%	0.00 (1.28)^{l}	0.00 (1.28)^{m}	0.00 (1.28)^{l}	0.00 (1.28)^{m}
T_{14}- Untreated control	-	47.08 (43.32)^{a}	74.56 (59.76)^{a}	94.67 (76.76)^{a}	97.88 (81.96)^{a}
SEm (±)	-	0.39	0.75	0.83	1.04
CD (at 5%)	-	1.10	2.11	2.34	2.96
CV (%)	-	3.39	4.78	4.19	4.90

Rate of chickpea seed (MSP) - Rs.50 kg^{-1}
Rate of deltamethrin 2.5WP- Rs.80/ 100gm
Agricultural labour wages: Rs.245 labour^{-1} during 2019 as per Govt. of West Bengal

Table 6: Economic analysis (C: B ratio) of the botanicals used against pulse beetle, *Callosobruchus chinensis* in chickpea during 2019

Treatments	Undamaged chickpea seeds (Kg qt^{-1})	Cost of undamaged chickpea seeds (total income)	Cost of plant protection\ qt. seed^{-1}	Net Benefit	Benefit over untreated control	Cost: Benefit ratio
T1-neem LP	94.04	4702.00	1225.00	3477.00	2565.83	1: 2.09
T2-neem BP	60.96	3048.17	1715.00	1333.17	422.00	1: 0.25
T3-neem LP+BP	85.59	4279.33	2695.00	1584.33	673.17	1: 0.25
T4-melia LP	91.03	4551.33	1225.00	3326.33	2415.17	1: 1.97
T5-melia BP	55.42	2771.17	1715.00	1056.17	145.00	1: 0.08
T6-melia LP+BP	78.78	3939.00	2450.00	1489.00	577.83	1: 0.24
T7-datura LP	88.18	4409.17	1470.00	2939.17	2028.00	1: 1.38
T8-datura SP	73.86	3693.00	2205.00	1488.00	576.83	1: 0.26
T9-daturaLP+SP	82.18	4109.17	2940.00	1169.17	258.00	1: 0.09
T10-tulsi LP	68.98	3449.00	1470.00	1979.00	1067.83	1: 0.73
T11-tulsi SP	64.75	3237.33	1960.00	1277.33	366.17	1: 0.19
T12-tulsi LP+SP	75.77	3788.67	2695.00	1093.67	182.50	1: 0.07
T13-deltamethrin 2.5 WP	100.00	5000.00	570.00	4430.00	3518.83	1: 6.17
T-14 untreated control	18.22	911.17	0.00	911.17	-	-

*Figures in parenthesis are angular transformed values.
Parentheses followed by same letters are statistically at par by DMRT.

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Impact of abiotic factors on incidence of rice leaf folder (*Cnaphalocrocis medinalis* Guenee) in agro-climatic condition of Ranchi, Jharkhand

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ABSTRACT

The field trial conducted to evaluate the effect of weather parameters on incidence and abundance of rice leaf folder revealed that leaf damage due to leaf folder (LDLF) had positive correlation with temperature (both minimum and maximum), morning humidity and sunshine hours i. e. r = 0.180, 0.196, 0.338 and 0.483*, while negative correlation with evening humidity (r = -0.048), wind speed (r = -0.096), rainfall (r = -0.144) and no. of rainy days (r = -0.071). The climatic factors together were able to explain the variation in LDLF (%) during experimental period to the extent of 76.30 per cent the coefficient of determination (R^2) was found to be significant i.e. 0.7630.

Keywords: Leaf folder, rice, weather

Rice (*Oryza sativa* L.), belonging family of grasses (Poaceae), is one of the most important cereal crops worldwide. It is the major staple food in Asian countries as well as in India and Jharkhand. It occupies third position in global cereals production after wheat and maize, (Tiwari *et al*., 2014) and its consumption outside Asia has increased recently (Orthoefer, 2005). *Cnaphalocrocis medinalis* Guenee (Pyralidae: Lepidoptera) is the most widespread and important species of leaf folder (Bhatti 1995). It is a predominant and one of the most destructive pests affecting rice ecosystems in Asia resulting in rice yield losses upto 30 to 80 per cent under epidemic condition (Raveeshkumar, 2015). Dyck, 1976 stated that weather factors are the major regulating factors for the insect pest populations under field circumstances. Abiotic factors such as temperature, relative humidity, rainfall and wind velocity may affect the distribution, growth, survival, behaviour, movement, reproduction, population dynamics, and outbreaks of insect pests of rice. Balasubramanian *et al*., (1982) reported maximum leaf folder damage at Coimbatore during the last week of April, October and second week of November. Balasubramani *et al*. (2000) also reported that the incidence of leaf folder in rice was very common in the month of September and October. Ahmad *et al.* (2010) reported that the incidence of leaf folder larvae on rice leaves commenced in 27 SMW with 0.33 larvae 10 hills^{-1} and reached to its maximum with 19 larvae 10 hills^{-1} in 36th SMW. Kumar *et al*. (2013) reported that its population reached at its peak level on 35th SMW in Haryana during in *Kharif* season 2013. It was positively and negatively correlated with maximum temperature and rainfall, respectively. Netam and Dupta (2015) reported regression equation for maximum temperature [Y = 2.149x-47.42; R^2 = 0.288] and evening relative humidity [y = -0.491+42.14; R^2 = 0.273].

MATERIALS AND METHODS

The field experiment was carried out to monitor the incidence and abundance of leaf folder in rice crop during *kharif* 2019 and 2020 at Rice Research Farm of BAU, Ranchi through fixed plot technique. Rice variety TN-1 were transplanted from nursery to the main field and crop was grown according to the recommended agronomical packages and practices. The size of each plot was 5.0m × 3m. The distance from row to row and plant to plant was kept at 20 and 10 cm, respectively. No insecticide either in soil or as seed treatment or foliar application was applied. This was done to allow development of natural population of leaf folder on the rice crop. Date of sowing was 27th June, date of transplanting was 17th July and date of harvesting was 27th December for both the season.

Meteorological observations with regard to ambient (maximum and minimum) temperature (^oC), per cent relative humidity (RH%), wind speed, rainfall (mm), no of rainy days and sunshine hours prevailing at the Research Farm of Birsa Agricultural University, RAC (Ranchi Agriculture College) Kanke, Ranchi during *Kharif* season of 2019 and 2020 were obtained from the Department of Agricultural Physics and Meteorology, Birsa Agricultural University, Ranchi (Table-1).

Ten rice plants (hills) were randomly selected for recording observation on presence of leaf damage (LD) by counting total no. of leaves (i.e . damaged + healthy leaves)

Table 1: Weekly meteorological data during the period of investigation

SMW*	Period	Temperature(°C)						R. H.(%)						Wind speed (km hr^{-1})			SS(hr)			Rain (mm)			Rainy days		
		Max.			Min.			7:00 AM			2:00 PM														
		2019	2020	Pooled Mean	2019	2020	Pooled Mean	2019	2020	Pooled Mean	2019	2020	Pooled Mean	2019	2020	Pooled Mean	2019	2020	Pooled Mean	2019	2020	Pooled Mean	2019	2020	Pooled Mean
31	30th Jul-5th Aug	30.4	33.1	31.7	23.4	24.0	23.7	84.9	85.6	85.2	68.3	69.0	68.6	3.2	3.2	3.2	14.4	28.8	21.6	18.2	31.4	24.8	2	2	2.0
32	6th Aug-12th Aug	30.0	33.3	31.6	23.1	23.7	23.4	85.6	85.1	85.4	69.6	69.1	69.4	3.6	2.6	3.1	5.5	26.7	16.1	161.8	92.8	127.3	6	4	5.0
33	13th Aug-19th Aug	29.6	31.2	30.4	22.3	22.1	22.2	85.7	85.3	85.5	69.1	69.7	69.4	2.7	2.5	2.6	8.5	16.8	12.7	96	48.8	72.4	3	4	3.5
34	20th Aug-26th Aug	29.9	32.4	31.2	23.6	23.2	23.4	85.0	86.3	85.6	68.6	70.1	69.4	3.2	3.0	3.1	26.5	39.6	33.1	47.8	177	112.4	4	4	4.0
35	27th Aug-2nd Sep	32.1	32.2	32.2	23.6	23.3	23.4	85.9	86.1	86.0	68.3	69.3	68.8	2.6	3.9	3.2	42.9	41.2	42.1	26.7	91.8	59.3	3	3	3.0
36	3rd Sep-9th Sep	30.1	31.6	30.9	22.2	23.3	22.8	85.9	85.4	85.6	68.4	69.1	68.8	3.0	2.6	2.8	28.2	49.7	39.0	62.7	0	31.4	6	0	3.0
37	10th Sep-16th Sep	30.7	33.6	32.1	23.7	24.1	23.9	86.7	86.3	86.5	68.0	69.6	68.8	2.3	2.8	2.5	21.9	58.0	40.0	32.6	6.2	19.4	2	1	1.5
38	17th sep-23rd Sep	31.8	31.3	31.6	22.9	22.5	22.7	87.4	83.7	85.6	68.4	69.7	69.1	2.7	3.0	2.8	54.4	33.1	43.8	36.8	120.6	78.7	2	4	3.0
39	24th Sep-30th Sep	28.8	32.1	30.5	21.3	22.1	21.7	88.0	85.1	86.6	70.0	69.9	69.9	4.0	2.8	3.4	9.4	52.6	31.0	152.2	5.2	78.7	7	1	4.0
40	1st Oct-07th Oct	28.9	31.1	30.0	21.7	21.7	21.7	85.9	86.1	86.0	69.4	69.0	69.2	2.2	2.3	2.3	48.5	26.9	37.7	0	12.2	6.1	0	2	1.0
41	8th Oct-14th Oct	28.3	31.1	29.7	21.2	22.3	21.7	86.3	83.7	85.0	68.4	69.3	68.9	2.7	2.2	2.4	39	50.0	44.5	60	2	31.0	3	0	1.5
42	15th Oct-21st Oct	29.1	30.7	29.9	20.7	21.2	20.9	85.6	87.0	86.3	66.4	68.9	67.6	2.2	2.3	2.3	51.6	63.7	57.7	30.6	0	15.3	2	0	1.0
43	22nd Oct-28th Oct	26.2	28.8	27.5	17.8	17.9	17.9	88.0	83.4	85.7	70.0	68.4	69.2	3.5	2.1	2.8	22.7	61.9	42.3	170.6	0	85.3	3	0	1.5
44	29th Oct-4th Nov	26.3	30.0	28.1	14.3	18.7	16.5	87.0	86.0	86.5	69.0	69.6	69.3	2.6	2.7	2.7	57.9	63.5	60.7	0	0	0.0	0	0	0.0

*SMW-Standard meteorological week

and no. of damaged leaves due to the pest. Per cent damaged leaves caused by leaf folder were calculated applying formula suggested by SES of IRRI Philippines:

$$\% \text{ leaf damage (LDLF\%)} = \frac{\text{Total no. of damaged leaves/10 hills}}{\text{Total no. of leaves (damaged+healthy)/ 10 hills}} \times 100$$

Weekly data on LDL.F per cent in rice variety TN-1 were co-related with weekly meteorological parameters viz. maximum temperature (X_1) and minimum temperature (X_2), morning relative humidity (X_3), evening relative humidity (X_4), wind speed (X_5), sunshine hours (X_6), rainfall (X_7) and number of rainy days (X_8). It was calculated to find out strength of relationship between these abiotic factors and the incidence of the leaf folder. Correlation coefficient (r) with positive and negative sign indicates that change in population and weather parameters was in same (increases or decrease in both) and opposite direction (increase in one and decrease in other), respectively. The multiple regression equation were established to find out change in dependent variable *i.e.* incidence of leaf folder (LDLF% per 10 hills) for a unit change in independent variable *i.e.* weather parameters. Regression coefficient represents amount of change in dependent variable for a unit change in corresponding independent variables. The sign (+ or -) of regression coefficient represents the direction of change for dependent variable *i.e.* positive sign indicate increases and negative sign denote decrease in dependent variable. The coefficient of determination (R^2) was also calculated for each multiple regression equation to interpret proportion of variance in the dependent variable that is predicted from the independent variable so that risk bearing capacity of model may be determined. In simple word R^2 indicates per cent variation in dependent variable by all the independent variable of multiple regression equation.

The regression co-efficients (b's) were also worked out for drawing the conclusion by using the formula of correlation.

$$\text{Correlation coefficient (r)} = \frac{\Sigma xy - \frac{(\Sigma x)(\Sigma y)}{n}}{\sqrt{\left[\Sigma x^2 - \frac{(\Sigma x)^2}{n}\right]\left[\Sigma y^2 - \frac{(\Sigma y)^2}{n}\right]}}$$

Regression coefficient (a & b; for linear regression equation y=a + bx)

$$a = \frac{(\Sigma y)(\Sigma x^2) - (\Sigma x)(\Sigma xy)}{n(\Sigma x^2) - (\Sigma x)^2}$$

$$b = \frac{n(\Sigma xy) - (\Sigma x)(\Sigma y)}{n(\Sigma x^2) - (\Sigma x)^2}$$

RESULTS AND DISCUSSION

The data on incidence of yellow stem borer and leaf folder during present experiment indicate that abiotic factors *viz;* maximum and minimum temperature, morning and evening relative humidity, wind speed (km h^{-1}), sunshine hour (hrs), rainfall and number of rainy days played an important role in oscillating the insect population in rice ecosystem during the *kharif* season 2019 and 2020. Monitoring operation of the fluctuation in the incidence and abundance of leaf folder infesting the popular rice varieties viz., TN-1 was taken up by regular observations recorded at weekly intervals. Population was recorded either in terms of number of damaged leaf caused by leaf folder.

The data on incidence of leaf folder recorded in terms of leaf damage due to lead folder (LDLF) during both the year of experiments (*Kharif* 2019 and 2020) are presented in the table -2. The incidence of LDLF in variety TN-1 was observed to occur at a very low level from 34th SMW (standard meteorological week) with density 12.55 and 13.98 per cent and reached at peak level at 38th SMW with 30.20 and 31.00 per cent LDLF, respectively in the year 2019 and 2020. Later the pest incidence began to decline from 28.44 and 28.88 in 37th SMW to the minimum level of 5.00 and 5.40 per cent LDLF in 45th SMW in the first and second year of the experiment, respectively. After 41st and 42nd SMW, the incidence of LDLF disappeared from the field. Pooled mean of the two years data revealed that incidence of LDLF started from 34th SMW with 13.27 per cent LDLF and reached at its peak level on 38th SMW with 30.60 per cent LDLF. After attainment of peak level of the incidence of LDLF, it was

Table 2: Population dynamics of leaf folder associated with rice var. (TN-1)

SMW	Period	TN1		
		LDLF (%)		
		2019	2020	Pooled mean
31	30th Jul-5th Aug	---	---	---
32	6th Aug- 12th Aug	---	---	---
33	13th Aug- 19th Aug	---	---	---
34	20th Aug- 26th Aug	12.55	13.98	13.27
35	27th Aug-2nd Sep	16.88	17.33	17.11
36	3rd Sep- 9th Sep	23.55	22.54	23.05
37	10th Sep-16th Sep	26.76	27.65	27.21
38	17th sep-23rd Sep	30.20	31.00	30.60
39	24th Sep- 30th Sep	28.44	28.88	28.66
40	1st Oct-07th Oct	25.33	25.21	25.27
41	8th Oct-14th Oct	21.20	21.56	21.38
42	15th Oct- 21st Oct	18.65	18.76	18.71
43	22nd Oct- 28th Oct	14.42	15.20	14.81
44	30th Jul-5th Aug	10.43	11.21	10.82
45	6th Aug- 12th Aug	5.00	5.40	5.20

LDLF: Leaf damaged by leaf folder

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Table 3: Simple correlation coefficients between LDLF % in rice varietyTN-1 and associated weather factors:

Year	Temperature		R.H.(%)		Wind speed (Km/hr^{-1})	Sunshine hr.	Rainfall (mm)	No of rainy days
	Max	Min	7:00 AM	2:00 PM				
2019	0.206	0.195	0.508*	-0.159	-0.124	0.376	-0.091	0.094
2020	0.108	0.188	-0.160	0.202	0.032	0.418	-0.106	-0.229
Pooled	0.180	0.196	0.338	-0.048	-0.096	0.483*	-0.144	-0.071

*Significant at P 5 per cent

observed to declined continuously and disappear from the field after 45th SMW with pest density 5.20 per cent LDLF. This experimental finding is almost in the agreement with the result of Balasubramanian *et al.,* (1982) who reported maximum leaf folder damage at Coimbatore during the last week of April, last week of October and second week of November. Balasubramani *et al*. (2000) reported that the incidence of leaf folder in rice was very common in the month of Sep and Oct. Ahmad *et al.* (2010) revealed that the incidence of leaf folder larvae on rice leaves commenced in 27 SMW with 0.33 larvae 10 hills^{-1} and reached its maximum with 19 larvae 10 hills^{-1} in 36th SMW.

Table 4: Multiple regression equation between weather parameters (X) and LDLF % (Y) of rice (Var. TN-1)

Year	Multiple regression equation	Coefficient of determination (R^2)
2019	Y = -920.6-5.213 (X1) + 4.61 (X2) +11.042 (X3) +0.714 (X4) -7.877 (X5) +0.277 (X6) - 0.146 (X7) +4.23 (X8)	0.9644
2020	Y = -342.538-8.558 (X1) + 6.404 (X2) -1.184 (X3) +8.129 (X4) +0.483 (X5) +0.524 (X6) - 0.066 (X7) +2.27 (X8)	0.4978
Mean	Y = -1,211.06-5.221 (X1) + 6.023 (X2) +5.765 (X3) +10.817 (X4) -6.69 (X5) +0.816 (X6) - 0.059 (X7) +2.328 (X8)	0.7630

X1 - Maximum temperature (°C)
X2 - Minimum temperature (°C)
X3 - Relative humidity (%) at 7:00 AM
X4 - Relative humidity (%) at 2:00 PM
X5 - Wind speed (km h-1)
X6 - Sunshine hour (hrs)
X7 - Rainfall
X8 - No. of rainy days
LDLF: Leaf damaged by leaf folder

The data presented in Table-3 revealed that, the LDLF per cent in case of rice variety TN-1 had non-significant correlation with all the weather parameters except in morning at relative humidity (r=0.508*) in the year 2019. LDLF showed non-significant positive correlation with maximum temperature (r = 0.206, 0.108), minimum temperature (r = 0.195, 0.188) and sunshine hours (r = 0.376, 0. 418) while negative correlation with rainfall (r = -0.091, -0.106) in the year 2018 and 2019, respectively. LDLF had positive and negative correlation with morning relative humidity (r= 0.508, -0.160) and number of rainy days (=0.094, -0.229) in the first and second year of the experiment, respectively while it had negative and positive correlation with evening humidity (r= -0.159, 0.202), wind speed (r = -0.124, 0.032) in the first and second year of the experiment, respectively. Pooled mean of two years data followed almost similar pattern of correlation as in the first year of experiment. The value of correlation coefficient between LDLF and weather parameters *viz*; maximum temperature, minimum temperature, morning humidity, evening humidity, wind speed, sunshine hours, rainfall and number of rainy days in rice variety TN-1 remained to be 0.180, 0.196, 0.338, -0.048, -0.096,0.483, -0.144 and -0.071, respectively. These findings are in corroboration with the results of Kumar *et al*. (2013) who reported the effect of weather parameters on the larval population of leaf folder at Haryana during *Kharif* season 2013. He reported that the population of leaf folder reached at its peak level on 35th SMW. It was positively and negatively correlated with maximum temperature and rainfall, respectively.

The multiple regression equation between LDLF per cent as dependent variable and weather parameters as independent variable were drawn and presented in Table-4 which revealed that unit change in weather parameters i.e. maximum temperature, minimum temperature, morning humidity, evening humidity, wind speed, sunshine hours, rainfall and number of rainy days was responsible for (-5.213, -8.558 and -5.221), (4.61, 6.404 and 6.023), (11.042, -1.184 and 5.765), (0.714, 8.129 and 10.817), (-7.877, 0.483 and -6.69), (0.277, 0.524 and 0.816), (-0.146, -0.066 and -0.059) and (4.23, 2.27 and 2.328) unit change in LDLF per cent during the first, second year of the experiment and their pooled mean, respectively.

The value of coefficient of determination (R^2) was 0.9644, 0.4978 and 0.7630 for the year 2019, 2020 and their pooled values, respectively indicated that 96.44, 49.78 and 76.30 per cent of the variation in the dependent variable can be predicted by the independent variable while 3.56, 50.22 and 23.70 per cent, respectively remained to be unexplained (Table-3). These findings are in partial agreement with the results of Netam and Dupta (2015) reporting the regression equation for maximum temperature [Y = 2.149x-47.42; R^2 = 0.288] and evening relative humidity [y = -0.491+42.14; R^2 = 0.273].

CONCLUSION

The study concludes that the lead damage due to leaf folder (LDLF) had positive correlation with temperature (both minimum and maximum), morning humidity and sunshine

hours while there was negative correlation with evening RH (%), wind speed, rainfall, and no. of rainy days. The climatic factors together were able to explain the variation in LDLF (%) during experimental period to the extent of 76.30 per cent while remaining 23.70 per cent depended on biotic factors.

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Biodegradation of imidacloprid by microbial consortium in sterile and non-sterile soil under *in vitro* conditions

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Abstract

Investigation on degradation of imidacloprid at 5 and 10 mg L^{-1} concentrations by microbial consortium in sterile and non-sterile soil was undertaken under *in vitro* conditions. HPLC analysis of imidacloprid residues done at 10 days interval for a period of 50 days showed that the rate of degradation of imidacloprid by microbial consortium was 52.63 and 51.85 per cent in the non-sterile and 72.69 and 73.21 per cent in the sterile soil at 5 and 10 mg L^{-1} doses, respectively. Imidacloprid degraded significantly faster in both treated sterile and non-sterile soil compared to control soil. The rate of degradation followed first-order and pseudo first-order kinetics in non-sterile and sterile soil, respectively. The study concluded that microbial consortium could effectively be used for degrading imidacloprid in sterile and non-sterile soil under *in vitro* conditions.

Key words: Imidacloprid, degradation, microbial consortia, sterile soil, non-sterile soil

Pesticides are an integral part of modern agricultural ecosystem. They help in protecting desired crops from various pests and diseases and thereby increasing the food production. However, their indiscriminate use may cause severe environmental pollution and their entry into food chain can lead to many human ailments. Soil is an important component of the environment and act as a sink for the pesticides used in crop protection. Only 0.1-2 per cent of the total pesticides applied reaches to the target organisms and rest (99%) enters into various parts of environment like soil, water, plant causing severe environmental problems (Miller, 2004). Carcinogenicity, mutagenicity, reproductive sterility, endocrine disorders, neurological and behavioral disorders especially among children and other health hazards are some of the problems of human beings occurred due to pesticide residues (Agnihotri, 1999). Biological removal of chemical pollutants becomes the method of choice recently since microbes can use a variety of xenobiotic compounds including pesticides for their food source and in the process they mineralize and detoxify them (Li *et al*., 2012).

Neonicotinoid insecticides are widely used in the chemical based control of sucking insect-pests in many agricultural and horticultural crops and imidacloprid is the most popular insecticide from this group because of its wider range of activity as seed dressing, soil treatment and foliar spray (Jeschke *et al*., 2011). Due to indiscriminate use of these insecticides, there rise serious concerns because these insecticides are neurotoxic in nature, exhibit prolonged persistence in the soil and also kill beneficial insects eventually affecting the whole ecosystem (Bhattacherjee *et al*., 2020a). Imidacloprid [1-(6-chloro-3-pyridinyl) methyl) N-nitro-2-imidazolidinimine] is found effective against soil insects, sucking insects, termites and some biting insects in various crops including rice, cotton, cereals, maize, sugar beet, vegetables, fruits, etc. (Elbert *et al*., 1998). Imidacloprid is used extensively in mango orchard to control hopper (*Amritodus atkinsoni* and *Idioscopus niveosparsus*) which are the major insect pests of mango. This being is a persistent insecticide, its residue can persist in the soil for more than one year depending on soil type environment and the soil conditions. It persisted in sandy loam soil of Australia with half-life values of 990-1230 days (Baskaran *et al*., 1999), in sandy loam and silty clay loam soil in Spain with 455-518 days and 233-366 days, respectively (Fernandez-Bayo *et al*., 2009), in alluvial, lateritie and coastal alkaline soil in India with 29-48 days (Sarkar *et al*., 2001), in sandy loam soil in India with 17.5 days (Bhattacherjee *et al*., 2019), in black sandy loam, clay and red soil in India with 50.10, 42.74 and 45.69 days (Samnani *et al*., 2013). This scenario warrants eco-friendly cost effective degradation of such a persistent insecticide and natural microbial transformation seems to be an effective and most viable approach. Several microorganisms were isolated and identified from contaminated soil which is capable of degrading imidacloprid. These include *Leifsonia* sp., *Pseudomonas* sp., *Bacillus* sp., *Ochrobactrum* sp., *Enterobacter* sp., *Burkholderia* sp. and so on (Anhalt *et al*., 2007; Pandey *et al*., 2009; Sharma *et al*., 2014; Hu *et al*., 2013; Sharma T. *et al*., 2014; Gopal *et al*., 2011). However, very few studies have been conducted on simultaneous microbial biotransformation of imidacloprid in sterile as well as non-sterile soil and that too using microbial consortium (Sharma S. *et al*., 2014; Vineyard and

Stewart, 2017). Sharma S. *et al*. (2014) have reported that two bacterial isolates of *Bacillus* sp. could degrade imidacloprid in clay loam soil under both autoclaved and unautoclaved conditions after 56 days. Vineyard and Stewart (2017) have suggested that the presence of soil microbes could help to degrade imidacloprid in cotton field soil after 25 days as sterilization of soil significantly reduced the concentrations of imidacloprid metabolites which were formed due to microbial degradation. Therefore, the present investigation was carried out to isolate different microbes from imidacloprid contaminated mango orchard soil and their capability as consortium to degrade imidacloprid in both sterile and non-sterile soil under *in vitro* conditions.

MATERIALS AND METHODS

Technical grade standard of imidacloprid (98.9% purity) was procured from Sigma-Aldrich India branch, Mumbai. Analytical and HPLC grade solvents were procured from the local market. Culture media were prepared according to Bergey's manual. The work has been done at the pesticide residue laboratory of ICAR-CISH, Rehmankhera, Lucknow. Soil samples were collected from mango field using aseptic technique for preventing contamination. Ten gram soil samples in petriplates were treated with two concentrations of imidacloprid (5 and 10 ppm), maintained in triplicate and kept at room temperature up to 50 days along with control (without microbes but with pesticide). Sampling was done at 10 days interval (0, 10, 20, 30, 40 and 50 days after treatment) to analyze imidacloprid residues by HPLC.

Carbohydrate utilization broth and agar with 1 per cent imidacloprid formulation as carbon source were used to isolate and purify microbial cultures with imidacloprid degradation potential. After autoclaving at 121°C and 15 psi for 15 min, imidacloprid formulation (Media® 17.8 SL, 1%) and suspension from mango orchard soil (1 mL) were added aseptically to cooled broth. All the composites were then mixed properly and incubated at 30 °C for 3 days. The colonies were isolated after pour-plating 1 mL of culture broth on carbohydrate utilization agar with the insecticide. Further purification of isolates was done by streaking on same agar plates. Nutrient agar slants were used for the maintenance of pure cultures.

A total of 16 microbes (12 bacteria + 4 fungi) were isolated from imidacloprid containing mango orchard soil. All the bacteria were found gram negative after gram staining. DNAs were extracted from microbial cultures and evaluated on 1.2 per cent agarose gel. Amplification of isolated DNAs for bacteria was done with *16S rRNA* Specific Primer (8F and 1492R) and a single discrete PCR amplicon band of 1500 bp was observed. Amplification for fungal isolates was done with ITS region Specific Primer (ITS1 and ITS4) and a single discrete PCR amplicon band of 700 bp was recorded. After enzymatic purification of PCR amplicon, bi-directional DNA sequencing reaction was done with 8F and 1492R primers for bacteria and ITS1 and ITS4 primers for fungi, using BDT v3.1 Cycle sequencing kit. According to the method mentioned by Altschul *et al.* (1997), BLAST alignment search of NCBI Gene bank database was carried out using *16S rDNA* sequence. Distance matrix was developed using RDP database on the basis of maximum identity score and Phylogenetic tree was prepared using MEGA7 software.

Soil samples were crushed to powder with the help of pestle and mortar after sieving to remove exogenous materials. Ten gram soil (three replications for each treatment) was taken in 50 mL culture tube and 25 mL of AR grade acetonitrile was added to it followed by extraction by vortexing for 5 min. Sonication for samples in an ultrasonic cleaner was then done for 20 min and the supernatant was filtered through whatman No. 42 filter paper. After repeating the process with 25 mL acetonitrile, pooled extract was evaporated in a rotary vacuum evaporator to near dryness. The residues were immediately dissolved in 5 mL of HPLC grade acetonitrile for HPLC analysis. Imidacloprid stock solution of 400 mg L^{-1} was prepared by dissolving precisely weighed 10 mg of technical grade insecticide (Sigma-Aldrich, Switzerland; > 98% pure) in 25 mL of HPLC grade acetonitrile. Working solutions of 1, 2, 4, 5 and 10 mg L^{-1} were prepared by subsequent dilution in acetonitrile.

Residues of imidacloprid were analyzed using HPLC (Shimadzu, Japan make model LC 10 ATVP) combined with photodiode array detector and rheodyne injector as per the method mentioned in literature (Bhattacherjee, 2013). Reverse phase consisted of µBondapak™ C-18 column (300 mm × 3.9 mm id with 125 Å porosity and 10 µm film thickness) and mobile phase was acetonitrile:water (35:65, v/v) with a flow-rate of 0.8 mL min^{-1}. Maximum wavelength and injection volume were set at 270 nm and 20 µL, respectively. Before injecting to HPLC, samples were filtered using a nylon membrane filter (Millipore, 0.45 mm thickness and 13 mm diameter) held tightly in a filter holder attached to a glass syringe.

RESULTS AND DISCUSSION

Identification of microbes

Among, sixteen isolated microbes, two bacteria (*Pseudomonas mosselii* strain NG1 with NCBI accession no. MN227542 and *Sphingobacterium mucilagenosum* strain NG 201 with NCBI accession no. MN818683, and three fungi

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(*Penicillium oxylatum* strain NG 201, *Aspergillus aculeanatum* strain NG 202 and *Aspergillus oryzae* strain NG 203 with NCBI accession nos. MT023713, MT023714 and MT023715 respectively were identified on the basis of growth promoting and enzymatic activities, using 16S rDNA gene sequencing technique.

Leifsonia sp. strain PC-21, having ability to degenerate imidacloprid in soil, has been isolated and identified by 16S rRNA method through PCR amplification of 500 bp sequence (Anhalt *et al*., 2007). *Pseudomonas sp.* strain 1G has also been isolated from soil as imidacloprid degrading bacteria (Pandey *et al*., 2009). *Burkholderia cepacia* strain CH9, an aerobic bacterium identified by 16S rRNA gene sequence method, has been isolated from agriculture field soil by enrichment culture which was found capable of removing imidacloprid residues (Gopal *et al*., 2011). Hu *et al*. (2013) have identified *Ochrobacterium* sp. strain BCL-1, a gram negative rod shaped bacterium, from tea rhizosphere soil with the help of 16S rRNA gene sequence having capability to degrade imidacloprid. From agriculture field soil of Uttrakhand, India, three bacterial strains (*Achromobacter* sp. GB 5, *Pseudomonas* sp. GB 35 and *Microbacterium* sp. GB 78) have been identified to degenerate imidaclopid by using 16S rDNA blast method (Negi *et al*., 2014). Sharma T. *et al*. (2014) have reported that *Enterobacter* sp. strain ATA1, isolated from paddy field soil at Punjab, had the ability of degrading imidacloprid. Similarly, *Bacillus aerophilus* showed maximum potential to degrade imidacloprid in clay loam soil as mentioned (Sharma *et al*., 2016). Recently in mango orchard soil, *Pseudomonas mosselii* strain NG1 has been identified with imidacloprid degradation potential using 16S rRNA gene sequencing technique (Bhattacherjee *et al*., 2020b). In NCBI data base there are five reports of the presence of imidacloprid degrading genes in *Pseudomonas* sp. and one of *Marinobacter salarious*. However, no report is available for the isolation of both bacteria and fungi in the earlier investigations.

Degradation of imidacloprid in non-sterile soil

Ten gram non-sterile (unautoclaved) soil samples in petriplates were fortified with 5 and 10 mg L^{-1} concentrations of imidacloprid and kept at room temperature up to 50 days after inoculating with microbial consortium. In non-sterile soil, degradation of imidacloprid by microbial consortium was almost at par in both lower (5 mg L^{-1}) and higher (10 mg L^{-1}) doses. Imidacloprid degraded from 0.76 mg g^{-1} at 0 day to 0.36 mg g^{-1} after 50 days of inoculation at 5 mg L^{-1} concentration resulting in 52.63 per cent degradation, whereas in case of 10 mg L^{-1} concentration, the degradation of imidacloprid was found to be 51.85 per cent (from 1.62 mg g^{-1} at 0 day to 0.78 mg g^{-1} after 50 days of inoculation) (Table 1). The degradation was slow in control soil samples without any microbe. For 5 mg L^{-1} concentration, it degraded from 0.81 mg g^{-1} at 0 day to 0.46 mg g^{-1} after 50 days of treatment (43.21% degradation). In case of 10 mg L^{-1} concentration, the degradation (41.01% - from 1.78 mg g^{-1} at 0 day to 1.05 mg g^{-1} after 50 days of treatment) is slightly slower than that in 5 mg L^{-1} concentration. Compared to control samples, degradation of imidacloprid was significantly better in treated soil samples. These results suggest that microbial degradation of imidacloprid in non-sterile soil does not depend on its dose. The degradation followed first-order rate kinetics in all four treatments.

Table 1. Degradation of imidacloprid (98.9% pure) by microbial consortium in non-sterile soil under *in vitro* conditions

Sampling period (Days)	Imidacloprid residues (µg/g) ± SD*			
	C1 (5 µg/g)	T1 (5 µg/g)	C2 (10 µg/g)	T2 (10 µg/g)
0	0.81 ± 0.025	0.76 ± 0.206	1.78 ± 0.15	1.62 ± 0.11
10	0.68 ± 0.07	0.63 ± 0.015	1.43 ± 0.09	1.27 ± 0.049
20	0.59 ± 0.036	0.55 ± 0.047	1.32 ± 0.011	1.16 ± 0.061
30	0.53 ± 0.025	0.48 ± 0.041	1.21 ± 0.006	1.05 ± 0.01
40	0.49 ± 0.021	0.43 ± 0.036	1.11 ± 0.015	0.92 ± 0.041
50	0.46 ± 0.036	0.36 ± 0.026	1.05 ± 0.015	0.78 ± 0.05
Degradation after 50 days (%)	43.21	52.63	41.01	51.85

*SD = Standard deviation
T1 – 5 mg/g imidacloprid with consortium
C1 – 5 mg/g imidacloprid without consortium
T2 – 10 mg/g imidacloprid with consortium
C2 – 10 mg/g imidacloprid without consortium

Degradation of imidacloprid in sterile soil

Significant degradation of both the concentrations of imidacloprid by microbial consortium was recorded in sterile (autoclaved) soil under laboratory concentrations. Imidacloprid degraded from 2.82 and 5.86 mg g^{-1} at 0 day to 0.77 and 1.57 mg g^{-1} after 50 days of inoculation at 5 and 10 mg L^{-1} level of concentrations, respectively. Whereas in control soil, the degradation of imidacloprid was recorded as from 2.94 and 6.74 mg g^{-1} at 0 day to 1.42 and 3.12 mg g^{-1} at 50 days after inoculation in 5 and 10 mg L^{-1} concentrations, respectively. Though the variation in level of degradation among two concentrations was minimal (72.70% in 5 mg L^{-1} and 73.21% in 10 mg L^{-1}) in treated samples, but it was significantly higher than that in control samples (51.70 and 53.71% for 5 and 10 mg L^{-1}, respectively) during the study period. The residues of imidacloprid in control soil samples were higher than their respective inoculated soil samples throughout the study period (Table 2). In all four treatments the rate of degradation followed pseudo first-order kinetics – after faster degradation initially it became slower at later stages. Better degradation in sterile soil indicated that microbial consortium is quite capable of degrading

Table 2. Degradation of imidacloprid (98.9% pure) by microbial consortium in sterile soil under laboratory conditions.

Sampling period (Days)	Imidacloprid residues (µg/g) ± SD*			
	C1 (5 µg/g)	T1 (5 µg/g)	C2 (10 µg/g)	T2 (10 µg/g)
0	2.94 ± 0.03	2.82 ±0.101	6.74 ± 0.21	5.86 ± 0.246
10	2.16 ± 0.102	1.97 ± 0.026	4.47 ± 0.107	3.16 ± 0.114
20	1.77 ± 0.053	1.44 ± 0.119	3.93 ± 0.076	2.76 ± 0.121
30	1.66 ± 0.05	1.24 ± 0.137	3.57 ± 0.165	2.53 ± 0.02
40	1.55 ± 0.085	1.05 ± 0.058	3.31 ± 0.046	2.16 ± 0.01
50	1.42 ± 0.041	0.77 ± 0.069	3.12 ± 0.076	1.57 ± 0.234
Degradation after 50 days (%)	51.70	72.69	53.71	73.21

*SD = Standard deviation
T1 – 5 mg/g imidacloprid with consortium
C1 – 5 mg/g imidacloprid without consortium
T2 – 10 mg/g imidacloprid with consortium
C2 – 10 mg/g imidacloprid without consortium

imidacloprid at room temperature under *in vitro* conditions.

Higher degradation of imidacloprid by microbial consortium in sterile soil than non-sterile soil might be due the better activities of study microorganisms in sterile soil where other microbes do not interfere in their degradation potential. The consortium of two soil isolated bacteria (*Bacillus aerophilus* and *Bacillus alkalinitrilicus*) has been found capable of degrading imidacloprid under both autoclaved and unautoclaved conditions. Imidacloprid degraded from 50, 100, and 150 mg kg^{-1} concentration to 3.18, 5.83 and 10.31 mg kg^{-1} under unautoclaved conditions after 56 days of inoculation (Sharma S. *et al*., 2014). Sharma *et al.* (2016) have reported that *B. aerophilus* could degrade imidacloprid in clay loam soil up to 93.45, 95.41 and 95.02 per cent under autoclaved (sterile) conditions and up to 80.93, 87.57 and 85.95 per cent under unautoclaved (non-sterile) conditions after 56 days of imidacloprid application at 50, 100 and 150 mg kg^{-1} doses, respectively. Our results are in contrast with that of Liu *et al*. (2011) where imidacloprid degradation was reported to be only 22.5 per cent in unsterilized soil after 25 days in contrast to almost nil degradation in sterilized soil within the same period, which indicated that soil microbes played an important role in imidacloprid degradation. Pooja (2017) have mentioned that two soil isolated bacterial strains *Bacillus pumilus* strain NFB1 and *Bacillus aryabhattai* were capable of degrading 100 mg kg^{-1} imidacloprid up to 73 and 76 per cent in sterilized soil which is in sync with our study and up to 80.9 and 82.2 per cent in unsterilized soil after 60 days of treatment, which was much higher than our results. Bacterial isolates *Ochrobactrum thiophenivorans* and *Sphingomonas melonis,* isolated from cotton field soil, individually and as mixture could degrade imidacloprid completely in soil within two weeks period (Erguven and Demirci, 2019). Our recent findings suggested that imidacloprid could influence bacterial community diversity in mango orchard soil where some bacterial populations were increased and some decreased / disappeared at various levels while no changes were observed in some populations, which could be exploited for developing microbial bioremediation consortium (Garg *et al*., 2021).

CONCLUSION

The study concludes that the consortium of bacteria and fungi are capable of degrading imidacloprid in both sterile and non-sterile soil under laboratory conditions. Therefore, utilization of this consortium in farmers' fields can be used in minimizing imidacloprid residues from the contaminated soil.

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Potential of moth bean (*Vigna aconitifolia*) in effective bee keeping of *Apis mellifera ligustica* Spinola colonies (Hymenoptera: Apidae) during autumn season

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ABSTRACT

Investigations carried out to assess the effectivity of moth bean (*Vigna aconitifolia*) through various combinations with millets (*Pennisetum glaucum*), imported pollens (mixed), honey and powdered sugar, used as suitable substitute of natural food (pollen and rector) showed that feeding bee colonies with pollen diet alone (treatment *P*) resulted in superior capped workers brood area, unsealed honey stored area, pollen (bee bread) stored area and number of frames occupied by bees. Among other combinations with pollen, the bean combination *PB* (pollen+ moth bean flour + powder sugar + honey) was the second followed by *PMB* (pollen+ pearl millet = moth bean +). When used without pollens the moth bean was found most effective in producing capped workers brood area, the unsealed honey stored area, pollen stored area and a number of frames occupied by bees.

Keywords: Supplements, substitutes, pollen, capped workers broad area, unsealed honey stored area, diet.

Beekeeping is highly dependent on the environmental fluctuations including flexibility in availability of natural food (pollens and nector) that serves as a primary source of nutrients (carbohydrates, proteins, lipids, vitamins and minerals) required for proper growth and development of honeybees (Di Pasquale *et al*., 2013; Naug, 2009). For carbohydrates and proteins, bees are reliant on pollen grains from various floral resources (Javaheri *et al*., 2000). These floral resources are harvested by worker bees to collect pollen grains and nectar (the energetic food resource) and feed the colony members including immature ones (Vaudo *et al*., 2015). Not merely for growth and development, this nutrition is also required by the worker bees in large amounts to activate glands for producing honey, royal jelly and propolis (Standifer *et al*., 1977; Keller *et al*., 2005; Saffari *et al*., 2010; Vasquez and Olofsson, 2009; Brodschneider and Crailsheim, 2010).

Availability of natural food gets significantly reduced during the dearth periods of the year (dry season) resulting in drastic decline in the number of worker bees in the hive. Further, it leads to reduction in the egg laying potential of the queen and decreases the survival rate of individuals that ultimately makes the colony vulnerable to disease and higher abandonment rates (Hoover *et al*., 2006; Morais *et al*., 2013). Moreover, honey and pollen grains stored by honey bees in the combs are exhausted very quickly and such conditions often lead to cannibalism among colony members (Schmickl and Crailsheim, 2001).

In today's time, the bee keepers feed bee colonies with supplements and substitutes of pollen and nectar to maintain the strength of colonies, honey production and other activities (Herbert, 1992). Such additional feeding in the form of artificial diets help in improving the strength of bee colonies and ultimately colony's ability to withstand both biotic (parasites and pathogens) and abiotic (temperature) stresses (Michener, 2007; Annoscia *et al*., 2017; Frizzera *et al*., 2020). Beekeepers usually manage colonies with additional carbohydrate sources with starch syrups or high-fructose corn syrup (HFCS) and homemade inverted sugar syrups (Brodschneider *et al*., 2010; Brodschneider and Crailsheim, 2010; Krainer *et al*., 2016). A mixture of sucrose and water is widely used to feed colonies of honey bees that have enough to stores (Semkiw and Skubida, 2016). Meanwhile, in temperate areas, additional feeding typically occurs during the autumn season, when honey bees suffer from low nectar flow from floral resources and bad weather. The inverted sugar syrups are the most common diet being used for supplementary feeding of bees. It is obtained by mixing sugar and water in 2:1 ratio and further adding acidifying agents to this sugar solution (Frizzera *et al*., 2020; Genc and Aksoy, 1993). Beside this sugar, additional sources of proteins are also required by bees to discharge their duties efficiently (Standifer *et al*., 1977). Eventually, the addition of such additional nutrition (carbohydrate and proteins) has become a common practice particularly during the fall period (Semkiw and Skubida, 2016).

In the past, the use of several foodstuffs rich in nutrition along with pollen (18-27 kg colonys) have been proved

effective in producing high number of vigorous broods and workers (Farrar, 1968; Standifer *et al*., 1973; Shelly and Villalobos, 2000). It is further evident from the literature that pollen substitutes (soybean, wheat, maize, horse gram, chickpea, green gram, field pea etc.), honey and sugar solution had a positive impact on overall activities of honey bee colonies (Al-Eitby, 2009; Al-Maktary, 2009; Nowar, 2011; Sena *et al*., 2012; Ghazala and Nowar, 2013; Shehata, 2016; Goodwin *et al*., 1994). However, there is dire need to find out more useful food supplements for better bee keeping during incongruous environmental conditions. In the present investigation, the effect of unexplored supplemental feeding by millet, bean and pollen based diets in the autumn season on honeybee colony status, the activity of brood rearing, honey storage, pollen storing, number of frames occupied by bees and diet consumption have been evaluated.

MATERIALS AND METHODS

The experiment was carried out at the apiary located in the Department of Plant Protection, Faculty of Agricultural Sciences, Aligarh Muslim University, Aligarh, U.P., India, during the autumn seasons of the year 2017-18. Langstroth beehives (modern hives/ Alexander Briones Gomez (Peru) of *Apis mellifera ligustica* Spinola were used in the experiment. Each hive contained six bees occupied frames comprising broods of different ages, pollen frame, honey frame and comb wax (Kasangaki *et al*., 2020).

A total of 45 honey bee colonies, procured from local beekeepers of Aligarh district, were maintained during autumn months. The selected colonies were reared in Langstroth modern beehive. The colonies were standardized and balanced in terms of brood frames, honey and pollen at the beginning of the experiment with the queens of equal age.

Seven diets comprising of pollen, pearl millet *P. glaucum* flour, moth bean (*V. aconitifolia*) flour and sugar were evaluated with supplements and substitutes. Pollen and nectar substitute (sugar solution) were prepared in different combinations. In these nine treatments, three pollen substitutes (without natural pollen) and four supplements (with natural pollen) were prepared. The feeds (amount of the diet to be given at one time) were prepared in the ratio 2:1:1 of supplements and substitutes pollen in seven different combinations with different ratios as shown in table 1 and 2. The feeds prepared in the patty form was provided @ 400 gm cooony^{-1} after every 12 days of interval. The sugar solution prepared in the ratio 1:1 (w/v) of sugar and water was given

Table1. Composition supplement and substitutes of pollen

Pollen supplements/ substitutes	Diet	Components	Source	Mode of application	Percentages (%)
Pollen supplements	P	Pollen	Imported	powder	50%
		Sugar	Local market	powder	25%
		Honey	beekeeper	Liquid	25%
	PM	Pollen	Imported	powder	25%
		Pearl millet	Local market	powder	25%
		Sugar	Local market	powder	25%
		Honey	beekeeper	Liquid	25%
	PB	Pollen	Imported	powder	25%
		Moth bean	Local market	powder	25%
		Sugar	Local market	powder	25%
		Honey	beekeeper	Liquid	25%
	PMB	Pollen	Imported	powder	25%
		Pearl millet	Local market	powder	12.50%
		Moth bean	Local market	powder	12.50%
		Sugar	beekeeper	powder	25%
		Honey	beekeeper	Liquid	25%
Pollen substitutes	M	Pearl millet	Local market	powder	50%
		Sugar	Local market	powder	25%
		Honey	beekeeper	Liquid	25%
	B	Moth bean	Local market	powder	50%
		Sugar	Local market	powder	25%
		Honey	beekeeper	Liquid	25%
	MB	Pearl millet	Local market	powder	25%
		Moth bean	Local market	powder	25%
		Sugar	Local market	powder	25%
		Honey	beekeeper	Liquid	25%

Table 2. Treatments used for honey bee feeding during autumn season

Treatments	Combination of diets
P	200 gm Pollen + 100 gm powder sugar + 100 ml honey
PM	100 gm Pollen + 100 gm pearl millet +100 gm powder sugar + 100 ml honey
PB	100 gm Pollen + 100 gm Moth bean + 100 gm powder sugar + 100 ml honey
PMB	100 gm Pollen + 50 gm Pearl millet + 50 gm Moth bean + 100 gm powder sugar + 100 ml honey
M	200 gm Pearl millet flour + 100 gm powder sugar + 100 ml honey
B	200 gm Moth bean flour + 100 gm powder sugar + 100 ml honey
MB	100 gm Pearl millet + 100 gm Moth bean +100 gm powder sugar + 100 ml honey
SS	500 ml Sugar solation
C	Without feeding - Control

twice/week @ 500 ml colony^{-1} through a plastic feeder kept next to the last frame of the hive. A control treatment without feeding was also kept last for comparison. For each treatment, five colonies were assigned.

The effect of feeding with supplements, pollen substitutes and nectar on honey bee activities was monitored through recording brood area, unsealed honey stored area, pollen stored area and a number of frames occupied by bees. The capped workers brood area, unsealed stored area and pollen stored area were measured at 12-day intervals using an empty standard frame with grid wire divided into square inches, and the overall seasonal areas were determined. The data were converted from square inch to cm² (1 inch = 2.54 cm). The number of frames occupied by bees from the two sides was taken in to account to determine the number of frames occupied by bees reared in each colony.

Percentage increase or decrease in different parameters was calculated using the following equation:

$$\%\text{Increase or decrease in area of brood, honey pollen and no. of bees frames} = \frac{\text{Average area of brood, honey and pollen area after 12 days} - \text{Average area of brood, honey, pollen and no. of bees frames at beginning of experiment}}{\text{Average area of brood, honey, pollen and no. of bees frames at beginning of experimer}} \times 100$$

Complete Randomized Design (CRD) was used for calculating Analysis of Variance (ANOVA) ($p < 0.05$). Further, Duncan Multiple Range and Least Significant Difference (LSD) tests were utilized using Statistical Analysis Software (SAS version 6.12) to compare means and estimate the results.

RESULTS AND DISCUSSIONS

Capped workers brood area

All the treatments exhibited positive impact on the mean capped workers brood area over the control with a significant (f=56.79, $p<0.05$, df=8; f=26.65, $p<0.05$, df=8) difference between each other. The largest capped workers brood area (1403.14 and 1804.29 cm²/colony) was seen in the colonies fed with pollen (treatment *P*). It was 65.37 and 91.64 per cent larger than those in control treatment during both years (2017 & 2018), respectively followed by treatments *PB* (60.37%), *PMB* (54.17%), *PM* (49.61%), *B* (43.82%), *M* (39.41%), *MB* (38.00%) and *SS* (26.61%) in the year (2017) and by treatments PB (87.63%), *PM* (79.08%), *PMB* (72.22%), *B* (62.44%), *MB* (50.69%), *M* (49.36%) and *SS* (25.56%) in the year (2018) (Table 3).

Table 3. Impact of feeding on capping of workers brood cells (area in cm²) during the autumn season (2017 & 2018)

Treatments	Before feeding		After feeding			
	2017	2018			Increase/decrease (%)	
			2017	2018	2017	2018
P	1220.00 ±11.55a	1183.33 ±36.32a	1403.14 ±a173.35	1804.29 ±372.42a	15.01 (65.37)	52.48 (91.64)
PM	1230.00 ±17.32a	1210.00 ±23.09a	1220.81 ±241.72cd	1593.62 ±260.07ab	-0.75 (49.61)	39.92 (79.08)
PB	1220.00 ±14.43a	1200.00 ±57.74a	1342.14 ±138.62ab	1718..67 ±262.47ab	10.01 (60.37)	48.47 (87.63)
PMB	1225.00 ±14.43a	1210.00 ±11.55a	1271.67 ±124.60bc	1610.00 ±323.93ab	3.81 (54.17)	33.06 (72.22)
M	1258.33 ±57.74a	1225.00 ±25.00a	1120.52 ±171.96e	1350.00 ±283.69c	-10.95 (39.41)	10.20 (49.36)
B	1250.00 ±28.87a	1141.67 ±100.00a	1168.24 ±176.86de	1407.48 ±264.35c	-6.54 (43.82)	23.28 (62.44)
MB	1233.33 ±18.19a	1208.33 ±82.07a	1080.86 ±241.72e	1347.62 ±417.35c	-12.36 (38.00)	11.53 (50.69)
SS	1225.00 ±14.43a	1208.33 ±150.23a	933.95 ±259.17f	1044.05 ±380.70d	-23.75 (26.61)	-13.60 (25.56)
C	1216.67 ±57.74a	1191.67 ±115.77a	603.95 ±313.97g	725.00 ±264.35e	-50.36	-39.16
LSD	93.38	224.91	89.73	185.25		
f	0.21	0.10	56.79	26.65		

The means that have different small letters have significant differences in all of column.

P=diet (Pollen + powder sugar + honey), PM= diet (Pollen + Pearl millet flour + powder sugar + honey), PB= diet (Pollen + Moth bean flour + powder sugar + honey), PMB=diet (Pollen +Pearl millet flour + Moth bean flour + powder sugar+ honey), M= diet (Pearl millet flour + powder sugar + honey), B= diet (Moth bean flour + powder sugar+ honey), MB= diet (Pearl millet flour + Moth bean flour + powder sugar+ honey), SS= Sugar solution and C= Without feeding (Control).

*The values above in parenthesis in % increase or decrease in comparison to observation before feeding.

* The values in parenthesis in % increase or decrease in comparison to values in control.

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Unsealed honey stored area

All the treatments exhibited positive significant (*f*=40.50, p<0.05, df=8; *f*=5.50, p<0.05, df=8) impact on the mean unsealed honey stored area. The biggest unsealed honey area was observed again in the colonies fed with pollen based diet (treatment *P*) (289.57 and 353.57 cm²/ colony) that increased 487.72 and 304.76 per cent over the control during both years (2017 & 2018), respectively followed by treatments *PB* (398.38%), *PMB* (393.34%), *MB* (390.25%), *PM* (375.82%), *B* (257.25%), *M* (257.25%), *SS* (221.54%) in the year (2017) and the treatment *PB* (205.67%), *PM* (221.38%), *B* (179.38%), *M* (176.43%), *PMB* (168.14%), *SS* (83.43%) and *MB* (81.90%) in the year (2018) (Table 4).

Table 4. Impact of feeding on production of unsealed honey stored cells (area in cm²) during the autumn (2017 & 2018)

Treatments	Before feeding		After feeding			
	2017	2018	Total mean		Increase/decrease (%)	
			2017	2018	2017	2018
P	50.00 ±11.55a	0.00 ±0.00a	289.57 ±178.65a	353.57 ±91.89a	479.14 (487.72)	353.57 (304.76)
PM	50.00 ±5.77a	0.00 ±0.00a	233.62 ±157cb	270.19 ±32.92ab	367.24 (375.82)	270.19 (221.38)
PB	50.00 ±2.89a	.00 ±0.00a	244.90 ±142.74b	254.48 ±28.49ab	389.80 (398.38)	254.48 (205.67)
PMB	50.00 ±5.77a	0.00 ±0.00a	242.38 ±148.99b	216.95 ±20.03bc	384.76 (234.40)	216.95 (168.14)
M	58.33 ±8.66a	0.00 ±0.00a	190.05 ±110.47d	225.24 ±37.35bc	225.82 (257.25)	225.24 (176.43)
B	58.33 ±8.33a	0.00 ±0.00a	203.38 ±122.31cd	228.19 ±25.05bc	248.67 (257.25)	228.19 (179.38)
MB	41.67 ±5.77a	0.00 ±0.00a	200.71 ±125.45d	130.71 ±14.07cd	381.67 (390.25)	130.71 (81.90)
SS	50.00 ±2.89a	0.00 ±0.00a	156.48 ±150.66e	132.24 ±22.37cd	212.96 (221.54)	132.24 (83.43)
C	50.00 ±8.66	0.00 ±0.00a	45.71 ±26.64f	48.81 ±12.22d	-8.58	48.81
LSD	21.96	0.00	30.60	107.63		
f	0.46	0.00	40.5	5.50		

Pollen (bee bread) stored area

All the diets (treatments) proved significantly (*f*=15.18, p<0.05, df=8; *f*=10.08, p<0.05, df=8) different with each other in terms of bee bread collection and storage. The highest pollen stored area (141.52 and 195.95 cm²/colony) was found in colonies supplemented with pollen (treatment *P*) which increased (66.13 and 654.84%) over the control, during both years (2017 & 2018), respectively followed by treatments *PB* (65.15%), *PMB* (55.32%), *PM* (54.81%), *B* (48.81%), *MB* (39.81%), *M* (31.56%), *SS* (10.16%) in the year (2017) and by treatments *PB* (611.80%), *PMB* (557.72%), *PM* (499.24%), *B* (492.00%), *M* (472.72%), *MB* (418.64%), *SS* (242.48%) in the year (2018) (Table 5).

Table 5. Impact of feeding on storage of pollen in cells (area in cm²) during the autumn season (2017 & 2018)

Treatments	Before feeding		After feeding			
	2017	2018	Total mean		Increase/ decrease (%)	
			2017	2018	2017	2018
P	150.00 ±17.32a	25.00 ±14.43a	141.52 ±94.90a	195.95 ±171.08a	-5.65 (66.13)	683.80 (654.84)
PM	140.00 ±5.77a	25.00 ±2.89a	116.24 ±50.22ab	157.05 ±101.68a	-16.97 (54.81)	528.20 (499.24)
PB	135.00 ±2.89a	25.00 ±1.15a	126.05 ±46.79ab	185.19 ±121.31a	-6.63 (65.15)	640.76 (611.80)
PMB	140.00 ±5.77a	25.00 ±2.31a	116.95 ±43.30ab	171.67 ±119.69a	-16.46 (55.32)	586.68 (557.72)
M	150.00 ±8.66a	25.00 ±0.00a	89.67 ±50.47c	150.48 ±17.97a	-40.22 (31.56)	501.92 (472.72)
B	133.33 ±5.77a	25.00 ±14.43a	102.71 ±49.60bc	1155.24 ±18.49a	-22.97 (48.81)	520.96 (492.00)
MB	150.00 ±5.77a	25.00 ±14.43a	102.05 ±51.81bc	136.90 ±115.82ab	-31.97 (39.81)	447.60 (418.64)
SS	141.67 ±5.77a	25.00 ±14.43a	54.38 ±41.26d	92.86 ±92.24b	-61.62 (10.16)	271.44 (242.48)
C	133.33 ±5.77a	25.00 ±0.00a	37.62 ±26.01d	32.24 ±13c	-71.78	28.96
LSD	25.25	28.85	24.192	52.85		
f	0.69	1.00	15.18	7.19		

Frames occupied by bees

All the diets significantly (*f*=28.55, p<0.05, df=8; *f*=45.11, p<0.05, df=8) influenced the frame occupation by honey bees (Table 4). The highest number of frames were occupied by bees (5.90 and 6.43 frames/colony) when fed on pollen based diet (treatment P). The frame occupation was 36.50 and 50.00% more as compared to control during both years (2017 & 2018), respectively. It was followed by treatments *PB* (35.34%), *PM* (34.17%), *B* (32.17%), *M* (30.17%), *MB* (29.50%) and *SS* (18.67%) in the year (2017) and by treatments *PMB* (45.66%), *PB* (44.50%), *PM* (44.00%), *B* (39.33%) *MB* (37.16%), *M* (34.50%) and *SS* (13.50%) in the yea (2018) (Table 6).

Table 6. Impact of feeding on frame occupation by bees (No.) during the autumn season (2017 & 2018)

Treatments	Before feeding		After feeding			
			Total mean		Increase/decrease (%)	
	2017	2018	2017	2018	2017	2018
P	6.00 ±0.00a	6.00 ±0.00a	5.90 ±0.52a	6.43 ±0.46a	-1.67 (36.50)	7.17 (50.00)
PM	6.00 ±0.00a	6.00 ±0.00a	5.76 ±0.74a	6.07 ±0.43abc	-4.00 (34.17)	1.17 (44.00)
PB	6.00 ±0.00a	6.00 ±0.00a	5.83 ±0.46a	6.10 ±0.60abc	-2.83 (35.34)	1.67 (44.50)
PMB	6.00 ±0.00a	6.00 ±0.00a	5.90 ±0.62a	6.17 ±0.78abc	-1.67 (36.50)	2.83 (45.66)
M	6.00 ±0.00a	6.00 ±0.00a	5.52 ±0.68a	5.50 ±0.47d	-8.00 (30.17)	-8.33 (34.50)
B	6.00 ±0.00a	6.00 ±0.00a	5.64 ±0.42a	5.79 ±0.37bcd	-6.00 (32.17)	-3.50 (39.33)
MB	6.00 ±0.00a	6.00 ±0.00a	5.48 ±0.60a	5.67 ±0.43d	-8.67 (29.50)	-5.67 (37.16)
SS	6.00 ±0.00a	6.00 ±0.00a	4.83 ±0.66b	4.24 ±0.11f	-19.50 (18.67)	-29.33 (13.50)
C	6.00 ±0.00a	6.00 ±0.00a	3.71 ±1.46c	3.43 ±1.83f	-38.17	-42.83
LSD	0.00	0.00	0.37	0.42		
f	0.00	0.00	28.55	45.11		

The apiculturists generally use sugar solution to maintain bee colonies during odd periods and to enhance the productivity during favorable conditions. However, use of grains viz., pulses, cereals, millets etc., can improve the honey bee colonies in all aspects. Past studies have proven this hypothesis. Gemeda (2014) found a significant difference in honey yield in colonies fed with pea flour and sugar solution. He pointed out that feeding colonies with pollen substitute during the dry seasons was better as compared to sugars. Similar findings were also reported by Goodwin et al. (1991), Goodwin (1997), Somerville and Collins (2007), Gemeda (2014), Gemeda et al. (2018), Al-Zubaidi (1998), Wongsiri (1999), Al-Hammadi (2001), Goodwin and Houten (1991), Goodwin et al. (1991), Goodwin (2015), Gemeda et al. (2018), Al-Zubaidi (1998), Wongsiri (1999), Al-Hammadi (2001) and Singh (2003).

It is clearly evident from the present finding that feeding bee colonies only with the pollen based diet (treatment *P*) resulted in superior capped workers brood area, unsealed honey stored area, pollen (bee bread) stored area and number of frames occupied by bees. However, among the other combinations with pollen, the bean combination *PB* (pollen+ moth bean flour + powder sugar + honey) was found second best in producing capped workers brood area, the unsealed honey stored area, pollen stored area and a number of frames occupied by bees followed by *PMB* (pollen+ pearl millet = moth bean +). The combination of pollen with millet (*PM*) was found fourth best diet however still inferior to *PB* combination. When the grains without pollens were provided, the bean has been found most effective in producing capped workers brood area, the unsealed honey stored area, pollen stored area and a number of frames occupied by bees. When the moth bean was combined with pearl millet the results were comparatively inferior to diet prepared only with beans but superior to millets used alone.

The superiority of pollen among all diet components is because they are the rich source of protein in the diet of the honey bee, also containing majority of lipids, vitamins and minerals necessarily required for the normal growth, development and vigor of the colony (Pernal and Currie 2000; Roulston and Cane 2000; Ghosh and Jung 2017). Presence of high nutrition in the pollen has probably stimulated the colonies that discharged a positive impact on honey bee colonies in terms of various parameters (Singh, 2003; Mattila and Otis, 2006; Gemeda, 2014; Kumar and Agrawal, 2014; Mahfouz, 2016). The probable reason behind second best performance by treatment *PB* (pollen + moth bean + powder sugar + honey) may be due to the reduction in total amount of protein when pollen and beans were combined in a diet form. However, this combination was still capable of stimulating the performance of honey bees to a more than required level. Similarly, Mahmood et al. (2013) also noted that, feeding honey bee colonies with soybean, gram, maize and sugar only, the treatment of the pollen with gram was higher in terms of the amount of honey extracted than other diets.

In commercial bee keeping, the pollens are very costly, if purchased from the market. They may also become a source of infection in the colonies during dearth period. In a study, Singh et al. (2010) showed that the pollen collected directly from healthy foraging bees from vegetation are sometimes carrying infective viruses *viz*., deformed wing virus (DWV), sac brood virus (SBV) and black queen cell virus (BQCV). This discovery raised concerns about the possible role of pollen in spreading the virus. Further, it is costly and sometimes it is difficult to obtain in the areas further from the main city.

The diets prepared with moth bean alone have been superior in simulating the performance of honey bees over moth bean and millet combination and millet alone. This shows that the bee colonies can sustain adequately even in absence of pollen with moth bean based diets as the colonies continued to raise brood at a good rate during the autumn and passed the season successfully in a good condition. Combining bean with pollen can reduce the amount of cost of cultivation for bee keepers as pollens are very costly and

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Table 7: Nutritional quantification of moth bean (*V. aconitifolia*) and pearl millet (*P. glaucum*) from the literature.

Parameter	Concentration (%)	
	Moth bean	Pearl millet
Crude protein (g $100g^{-1}$)	14.06-24.30	11.00-12.99
Crude fibre	0.33-3.90	2.30-11.30
Crude fat	1.60-4.20	4.30-7.96
Ash	2.81-4.30	2.20-3.00
Carbohydrate	57.90-68.0	67.0-72.8
Vitamins (mg $100g^{-1}$)		
Vitamin (Retinol)A	14.65-16.00	22.00
Thiamin B1	0.23-0.50	0.30-0.38
Riboflavin B2	0.45-0.10	0.15-0.29
Niacin B3	0.47-28.08	2.00-4.72
Pantothenic acid B5	0-0.50	0.85
Vitamin B6	0-0.40	0.38
Ascorbic C	42.25-59.10	1.60
Vitamin E	0-0.25	1.90
Minerals (mg $100g^{-1}$)		
Sodium	37.20-34.06	10.90-112.12
Calcium	133.00-244.10	8.00-50.07
Magnesium	183.00-214.04	114.00-177.45
Potassium	1191.00-2256.68	195.00-325.36
Phosphorus	174.26-489.00	285.00-399.23
Iron	7.46-11.00	3.00-11.0
Zinc	1.41-2.48	1.70-6.43
Amino acid (g $100g^{-1}$)		
Lysine	5.60-6.34	3.10
Leucine	7.00-7.42	10.70
Isoleucine	5.10-4.16	4.40
Valine	3.30-5.16	4.90
Methionine	1.00-1.62	1.10
Cystine	0.50-0.64	1.50
Phenylalanine	4.70-5.48	4.40
Tyrosine	3.14	3.00
Threonine	3.96	4.00
Tryptophan	0.70-1.24	1.40
Tannine	0	0
Histidine	2.76	2.30
Arginine	6.14	4.60
Aspartic acid	10.64	8.50
Glutamic acid	16.12	23.00
Glycine	3.08	2.70
Proline	3.33	5.80
Serine	4.36	5.20
Alanine	3.68	8.70

Sources: Opara et al., 2017; Hajjagana et al., 2014; Deshmukh and Pawar, 2020; Ramashia et al., 2019; Kanmani et al., 2018; Saleh et al., 2013; Vanga et al., 2018; Kaleem et al., 2020; Dias-Martins et al., 2018; Adsule, 1996.

not available readily in the market whereas moth bean is comparatively cheaper and good source of nutrition. The dietary components (bean, *V. unguiculata* and pearl millet, *P. glaucum*) used in this investigation contains an adequate proportion of protein, fat, ash, carbohydrate, ascorbic acid, Vitamin A, Thiamin, Riboflavin, Niacin and Vitamin E, potassium, Sodium, Phosphorus, Magnesium, Calcium, Zinc and Iron in an adequate amount naturally in them (Marsh et al., 2013; Hajjagana et al., 2014; Deshmukh and Pawar, 2020; Ramashia et al., 2019) (Table 7).

Moreover, the diets used in this study were a combination of single or multiple grains along with sugar solution. Sugar helps in feeding honeybees and its presence in the diet do not cause the moist cake to become hard when exposed to the warm and comparatively dry environment of the brood nest. Sucrose with protein supplements produces cakes that maintain their consistency for longer periods. Cakes prepared with honey maintain their consistency for a prolonged period (Standifer, 1980), Further, Macicka (1987) found that when the honey bee colonies were fed with a semi-solid diet (more than 20% protein), the bee consumption was very slow compared to the carbohydrate added to the protein.

CONCLUSION

The experiment concludes that honey bee colonies accept all the diets and sugar solution provided during the dearth period. Pollen supplement and its substitutes made from pollen, pearl millet, moth bean flour, sugar powder and honey led to continued colonies in brood production and compensated the shortage of pollen and nectar during the period of dearth.

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Impact of insecticides used in controlling desert locust (*Schistocerca gregaria* Forskal) and on crawling arthropods in Shieb, Eritrea

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ABSTRACT

The experiment conducted at Shelshela, sub-region Shieb – Eritrea during November 2015 to February 2016 showed that the use of fenitrothion 96 per cent ULV and chlorpyrifos 45 per cent ULV to control locust led to significant reduction arthropod population after 48 hrs of post-spray than the control plot. The fenitrothion treated plots generally showed more reduction in population count than chlorpyrifos. Fenitrothion treated plots recorded maximum reduction (89%) of formicids population. Whereas, chlorpyriphos recorded the lowest of carabids (52%). Population count of Gryllidae and Tenebrionidae showed slight improvement after a month of spray than others. Generally, a population reduction of 86.2 per cent in fenitrothion and 81.1 per cent in chlorpyrifos was recorded in first post-spray count of D3 as compared to pre-spray counts of D1 and D2. From the study it is also observed that Formicidae and Gryllidae can be used as bio-indicators during insecticide control of desert locust in the study areas.

Keywords: f*enitrothion, chlorpyrifos, non-targeted arthropods, Desert Locust, bio-indicator.*

The Desert Locust (*Schistocerca gregaria* Forskål, 1775) belonging to class – Insecta, Order – Orthoptera, and family Acrididae is one of the most serious and important agricultural pests, especially in low rainfall and marginal parts of the world during plague years. Extent of damage is due to its nature of damage, rapid increase in number or population and its capability for long distance migration. It damages crops and other vegetation from Atlantic coast of West Africa to India during outbreak/plague years. During plague it covers about 64 countries representing 20 per cent of land surface of earth, which is about 30 million Km^2 that extends from Mauritania in West Africa to Pakistan and India in Asia (Steedman, 1990). However, during recession period, when population occurs at low density, infestation area is reduced to 16 million Km^2 mostly distributed in arid areas that covers about 30 countries of North and East Africa, Middle East and Northwest of India (Steedman, 1990 and PAN-UK, 1998).

Most of the insecticides used in the control of locust are broad spectrum that may kill non–targeted organisms such as natural enemies, birds, rodents and can also be washed to nearest water sources and affect water dwelling organisms. From the sprayed insecticide only about 0.1 per cent reach target organisms and the remaining bulk (99.9%) contaminate the surrounding environment (Gill and Garg, 2014). Generally, the disadvantage of chemical insecticides, currently under use, arise from their contamination to environment, toxicity and hazard effect on non-target organisms (predators, parasitiods, pollinators), harmful residues on food, development of resistance by the pest against to insecticides and bio-accumulation (bio-magnification) in food chain (Borror *et al*., 1992).

Until very recently, environmental impact of insecticides sprayed for control of desert locust was not given much attention and investigation. Studies on negative impact of insecticides go back to early 1800s that mainly focused on effect of natural enemies and its importance on agriculture (Khalaf, 2004). Around 26 bird species and over 100 insect taxa are known to be natural antagonists (parasites, parasitiod and predators) of locusts and grasshoppers and that chemical control threatens them by secondary poisoning and food deprivation (Everts, 1990).

Eritrea is one of the frontline countries for Desert Locust breeding in North-east Africa and Arabian Peninsula. Its long Coastal lines (about 1200 km) are Desert which are favorable for locust winter breeding especially in sub-regions of Shieb, Afabet, Qurora, Massawa, Gelalo and Foro. During summer season, the locusts breed in the arid areas of Gash Barka and some part of Anseba. During winter when there is rainfall and floods along Wadis and Red Sea coast, there is always locust problem in the country. Natural habitats for breeding and recession areas of Desert Locust usually is treated with synthetic insecticide to prevent outbreak of the pest so as to secure food production for humans and animals in the country. During application of insecticide, many non-target arthropods that play key role in function and structure of arid ecosystem, share a common habitat with a desert locust become victims of the operations.

During outbreak of desert locust in Red Sea coast of Eritrea, control operations were taken up between October to March i.e. winter season. Over 219 insecticide control operations of the pest were taken from 1980s to 1997 (Wiktelius *et al*., 2003). Sometimes plague comes from neighbour countries like Sudan. For example, there was an invasion of tree locust in 1995 in Eritrea where control operation took place in Hazemo, Ala, and Deki-seb. During the operation, 26,243 liters of liquid formulation and 664 kg of dusts/powder were applied using aerial and ground application in 51,897 ha (Table 1). Maximum application was done during winter season of 2006/2007 in eastern lowland of Eritrea (Table 1). The operation was done both through ground and aerial application consuming 68,706 liters of spray liquid applied over 85,806 hectares of land that costed over 14 million Nakfa (MoA, 2007). Since 1986, in Eritrea more than 158,213.25 liters and 664 Kg of dust of different chemical formulations were applied against Desert Locust and tree locust over 469,059.5 hectare using ground and aerial application (aerial spray in 1992/93, 1995, 1997, and 2006/07). In many different ecological sensitive areas, many non-targeted arthropods were killed. Role of crawling arthropods in arid ecosystem and unavailability of data on the impact of insecticide control operations on non-targeted organisms in Desert Locust habitat of Eritrea, are alerting factors to conduct a study on effect of frequent application of insecticides to non-target terrestrial arthropods. The study with this objective and also to identify bio-indicators was therefore carried out. This will help us to understand the impact of insecticides applied to non-target arthropods and thereby develop an efficient application method which will protect natural biodiversity of terrestrial and aquatic habitat in areas of arid ecosystems.

MATERIAL AND METHODS

The experiment was carried out during winter breeding season in Shieb-Mensheb (Sheshela) area is, well known as hot spot of the pest incidence and outbreak in winter season, situated 12 Km North of the small town of Shieb-Mensheb, at $15^0$55′ 54.2" North and $39^0$04′ 40.2" East, with an elevation of 195 meter mean above sea level. It has arid climate with annual rainfall of 40 mm and an average annual temperature of 33.5^{0}C. The area is plain with clustered bunches of sand dunes and sand gravels. It was designed in a Randomized Complete Block Design (RCBD) with three replications in total area of 25 ha with 9 plots. Each plot had an area of 1 ha with a buffer zone of 100 meter between experimental plots to minimize spray drift between plots during the spraying (Dobson, 1991).

Three treatments namely: fenitrothion, chlorpyrifos and unsprayed plots (control) were used. The two insecticides were applied at recommended dose i.e. fenitrothion at 450 g a.i. ha^{-1} and chlorpyrifos at 240 g a.i. ha^{-1} Each treatment was replicated three times. Data were collected from each plot using pitfall trap for ground arthropods in both before and after spray. The days for population count were D1, D2, D3, D4, D5, D6, and D7. Where 'D' represents 'observation day'. The first two observation was taking before spray (D1 and D2), and the rest five observation were taken in post-spray count (D3, D4, D5, D6, and D7) (Table 1).

Table 1. Days of population count

Days	Time interval from spray
D1	10 days before spray
D2	3 days before spray
Spray	0
D3	2 days after spray
D4	9 days after spray
D5	16 days after spray
D6	24 days after spray
D7	34 days after spray

Five pitfall traps of 20cm x 25cm x 10cm size were used in each plot to catch ground and walking species in the experimental plots. The traps were placed at four corners of the fields fixed at about 20 m toward center of plot from the corner and the fifth trap was placed in the center of each plot. The traps were fixed into the ground, labelled so as the normal walking organisms may easily slip into the trap. Each trap was filled with water and detergents to deter the organisms not walking out from the trap. All the trapped organisms were collected every day in vials containing formaldehyde (40%) solution for preservation and species identification. The collected samples (preserved catches) were identified only up to family level.

The collected samples/specimens were identified at Hamelmalo Agricultural College using keys, manuals, books and comparing them with the already collected samples kept in the department.

For proper spraying of ULV formulations, hand-held sprayer (AULV+) was used. The calibration was done according to the method described by Dobson (2001). Treatments were applied using a hand-held ULV sprayer (ULVA+) having eight batteries with high emission height. The spray was done during between morning hrs start from 7:00 to 10:30 a.m., when the wind speed was 2-6 mile/hr, at temperature of 25 - 28^{0}C and relative humidity of 76 - 68%. During the application the sprayer having 8 batteries was held high (Dobson, 1991).

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Spray drifts were measured in 10 points as described by Dobson (1991) using oil sensitive paper of approximately 1.5 cm width and 7 cm long. The paper was attached on the top of 50 cm height of quadrant woodblock faced up the wind to catch the drifted droplets. The sticks were placed at an interval of 0, 2, 4, 7, 10, 15, 20, 25, 35, 50 m from the border of a treated plot to the direction of downwind in straight line. For each plot, two line of measuring drifts paper kept 40 meter apart and 30 meters from the corner toward centre of the line were used. As soon as the operation completed, the oil sensitive papers were collected and the droplets drifted on from the spray were counted using a magnifying hand lens per cm^2 (Dobson, 1991).

The effect of each treatment on individual families was calculated after the application of the chemicals as compared with mean of pre-spray count as proposed by Peveling and Weyrich, (1991).

$$\mathbf{R_p\ (\%) = [D_{pr}/D_{ps}] \times 100}$$

Where; D_{pr} = Mean of pre-spray count (D1 and D2 counts)

R_p =Population count reduction in %

D_{ps} = Population count of post-spray

Proportion of each family count/catches as relative abundance was used as described by Peveling and Weyrich (1991).

$$\mathbf{R_a\ \% = [F_c / T_c] \times 100}$$

R_a = Relative abundance of family in all count

F_c = Individual Family count/catch in all observations

T_c = Total families count

The data so collected were tested for significant differences using analysis of variance (ANOVA) for RCBD. The population count of each family throughout the study period (D1 to D7) was analysed statistically using a computer software Genstat® discovery fourth edition. Furthermore, the analysed data were used for the comparing pre- and post–spray count.

RESULTS AND DISCUSSION

Population count of arthropod families with respect to application of treatment

The study was focused on 4 insect families and order Arenae (spider), predominantly found in all the plots, throughout study period and were analyzed statistically. However, the insect families that were less and irregular in abundance were not analyzed. The identified and analyzed families were Formicidae (Hymenoptera), Tenebrionidae (Coleoptera), Carabidae (Coleoptera), Gryllidae (Orthoptera) which belonged to class Insecta and order Arenae to class Arachinidae.

Arenae

The population count of order Arenae was done in two pre- and five post-spray observations using pitfall trap (Figure 1). In pre-spray counts (D1 and D2), the population count did not show significant difference among treatments. The treatments of fenitrothion and chlorpyrifos count taken after 48 hrs of spray (D3) gave significantly lower population ($P \leq 0.05$) than the control plots. The lowest population of Arenae pitfall trap count was recorded in D3. The result revealed that the population of the Arenae increased at a decreased rate in pitfall trap counts of D4 to D7 which is significantly lower in population count than the pre-spray counts and the control plots. The result of D6, was significantly not different in population count from the control plot. The reduction of Arenae population in fenitrothion treated plots was 81 per cent whereas in chlorpyrifos treated plots it was 69 per cent in the pre-spray counts (D1 and D2). This may probably be due to the effectivity of the spider net used to hold insecticides, which might have increased the exposure time to the applied insecticides leading to high mortality of Arenae. The control plot gave significantly higher ($P \leq 0.05$) pitfall trap counts than fenitrothion and chlorpyrifos treatments. There was no significant difference in pitfall trap count between the plots treated with the fenitrothion and chlorpyrifos insecticides. The present result corroborates with result of Khalaf (2004) who reported that the spray of chlorpyrifos, fipronil and carbofuran reduced the relative abundance of spiders after 48 hrs of post-spray.

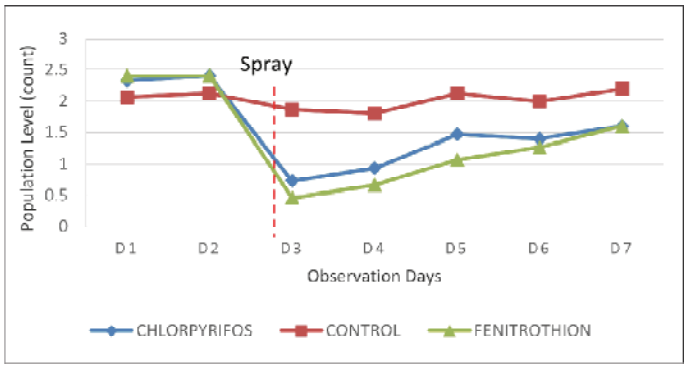

Fig. 1. Population count of Arenae (Arachnida) from pitfall trap

Formicidae

Population counts of family formicidae (Hymenoptera) from pitfall trap in pre-spray (D1 and D2) was non-significant

(P≤0.05). The mean population counts were 81.2 and 77.9 for D1 and D2, respectively (figure 2). However, the population in formicidae was significantly reduced (P≤0.05) after 48 hrs of post-spray in D3. The reduction in chlorpyrifos sprayed plot was 85%, whereas in fenitrothion sprayed plots was 89% from a mean value of pre-spray count (D1 and D2). After that the count remained almost constant in the next observation periods D4 to D6. The last observation (D7) showed an increase at decreased rate of population when compared with the pre-spray count. In general, the formicid population was significantly lower (P≤0.05) in the post-spray count in all the plots.

This finding is in agreement with the result of Khalaf (2004) in Sudan who reported significant reduction in ants population after the application of fenitrothion for control of Desert Locust. This also coincides with the finding of Peveling, *et al*. (1999) who reported reduction of ants population by 75 per cent in Madagascar using pitfall trap after spraying fenitrothion for control of red locusts. Similarly, finding of Childebaev (2001) are also in agreement with the current result, thus, the population of ants reduced after application of chlorpyrifos (180 g a.i./ha) to control rangeland grasshoppers and it took 5 weeks to reach pre-spray level in Kazakistan. Ants are easily affected non-target organism to the sprays of acetyl cholinesterase (AChE) inhibitors (OP) and IGR diflubenzuron (Sanchez-Bayo, 2012). Hence due to their abundance and sensitivity to insecticides applied for control of locust, Ants can be used as bio-indicator in locust insecticide control operations (FAO, 1988).

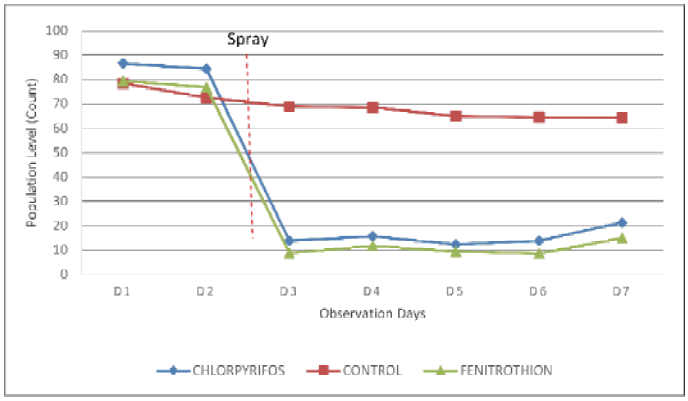

Figure 2. Population count of Formicidae (Hymenoptera) from pitfall trap

Gryllidae

The pre-spray pitfall trap population count of the family Gryllidae (Orthoptera) in the first two observations (D1 and D2) were 16.27 and 14.93, respectively. The data in the pre-spraying also showed no significant difference among plots (figure 3). Pitfall traps count 48 hrs after spray (D3) gave a significant reduction in the population of the cricket as compared to the pre-spray and control plot. Plots treated with fenitothion and chlorpyrifos had significantly lower population than the pre-spray plots in the pitfall trap population count of the fenitrothion and chlorpyrifos it was reduced by 82 per cent and 77 per cent from the mean of pre-spray count (D1 and D2), respectively. The pitfall trap population count of gryllidae in D4, D5, D6, and D7 steadily increased in both insecticides treated plots and it reached 19 and 22 per cent in D7 in fenitrothion, and chlorpyrifos treated plots as compared to the pre-spray count of D1 and D2 (Figure 17). There was no significant difference between fenitrothion and chlorpyrifos treated plots. The control plot had significantly higher population than the treated plots throughout out the pitfall trap of the Gryllidae.

The present finding is similar to the study conducted by Quinn *et al.* (1990) who reported that malathion (693 g a.i. ha^{-1}) and carbaryl (1.5 kg ha^{-1} of 5%) bran bait spray recorded, a reduction of 49-89 per cent of field cricket and other ground dwelling insects. This reduction in the population of Gryllidae and others is due to inhibiting action of AChE of the non-target insects. Khalaf (2004), reported that the relative abundance of crickets was adversely affected after the application of fipronil, which is in line with this study.

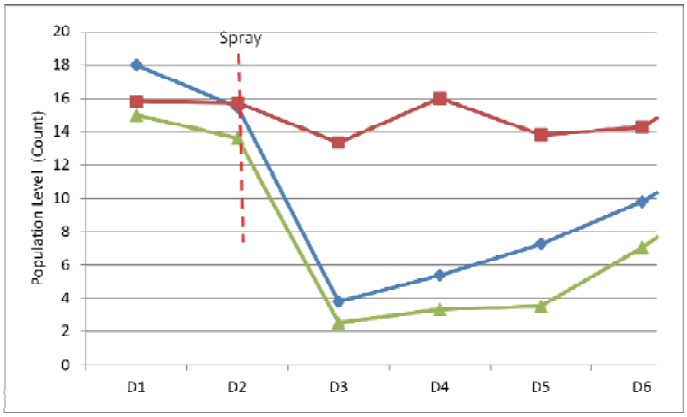

Figure 3. Population count of Gryllidae (Orthoptera) from pitfall trap

Tenebrionidae

Population count of Tenebrionidae (Coleoptera) from pitfall trap during pre-and post-spray period is presented in figure 4. It was in general low with a mean of 3.6. The population count in the pre-spray period (D1 and D2) was uniform in all the plots. Forty eight hours after the spray (D3), the population count in chlorpyrifos and fenitrothion treated plots was significantly lower (P≤0.05) than the pre-spray and the control plots. There was a reduction of 74 and

76 percent in population of tenebrionid beetle in post-spray observations in chlorpyrifos and fenitrothion treated plots, respectively. The population gradually increased in D4, D5, D6, and D7. Reduction of 50 per cent population in chlorpyrifos and 53 per cent in fenitrothion treated plots in the last observation (D7) was recorded as compared with the mean of pre-spray counts (D1 and D2). The study however, revealed no significant difference between the two insecticides used. The control plot gave significantly higher population than insecticide treated plots throughout the study period.

Most of the studies carried out on the effect of chemical control of Desert Locust and its impact on tenebrionids had similar results with the current study. Field studies conducted in Madagascar showed that fenitrothion (250g a.i ha^{-1}) a week after its application resulted in 69 per cent reduction of tenebrionidae beetles (Peveling *et al.*, 1999). It was also reported that four weeks post- spray count showed that the beetle population was still reduced by 51 per cent. Khalaf, (2004) reported that the application of fiprinol (4.2-13.4g a.i ha^{-1}) for control of Desert Locust caused 99 per cent mortality of tenebrionid beetles. In agreement with current study Tingle and McWilliams (1998) reported that tenebrionidae were more affected among the coleopteran families when exposed to fipronil sprays that is used for control of locust. Similarly, In Mauritania from field trial using fipronil at 13.4 and 4.2 g a.i ha^{-1} for the control of locust resulted in non-targeted mortality of tenebrionds to an extant of 99 and 85 per cent respectively (Tingle *et al.*, 2000). The result also showed population recovery of tenebriondas after 1-2 weeks of post spray count. Furthermore, a study on the effect of malathion (693 g a.i. ha^{-1}) and carbaryl 5 per cent bran bait spray on non-target arthropods carried out by Quinn *et al.* (1990) using a pitfall trap reported a reduction of 49-89 per cent of darkling beetle and others.

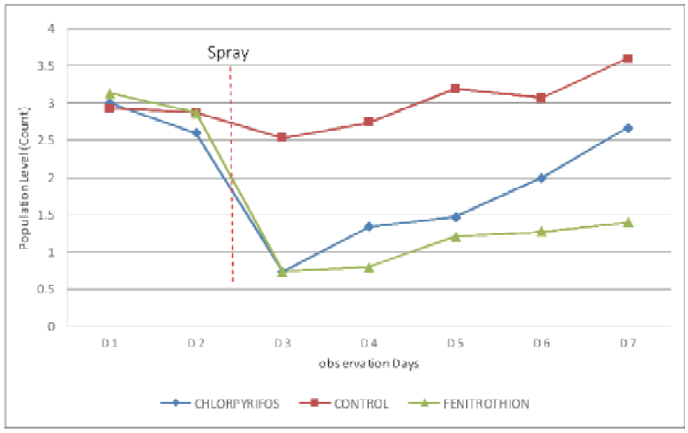

Figure 4. Population count of family tenebrionidae (Coleoptera) from pitfall trap

Carabidae

The population of Carabidae (Coleoptera) from pitfall trap was observed in pre- and post-spray period. In the first two days of pre-spray (D1 and D2), no significant difference among plots (Figure 5) was observed. However, after the application of the fenitrothion and chlorpyrifos in D3, D4, D5, D6 and D7, the treated plots recorded significantly lower ($P \leq 0.05$) count than the pre-spray and unsprayed (control) plots. Plots treated with chlorpyrifos shoved reduction in population count by 52 per cent whereas, in fenitrothion treated plots, the counts were reduced by 67 per cent than pre-spray period (D1 and D2). Even though the insecticide sprayed plots had a significant lower population count than control, the chemical treated plots recorded a slight and uniform increase in population count of pitfall trap from D4 to D7. The population level shows slight recovery. Thirty three percent thus of reduction in population was observed in both insecticide treated plots as compared with pre-spray observations (D1 and D2). The result of current study coincides with the finding of Smith *et al.* (2006) who reported that plots treated with fenitrothion and combination of esfenvalerate (245 g a.i. ha^{-1}) resulted in Carabidae population reduction up to 61 per cent. The author also reported reduction in the population of Carabidae by 69 per cent after one week of post-sprayed count with fenitrothion at a rate of 250 g a.i. ha^{-1} in Madagascar. Childebaev (2001) studied the impact of chlorpyrifos spray for control of rangeland grasshopper in Kazakhstan using pitfall. The author reported that population of carabids was significantly reduced than pre-sprayed counts and population of the beetle was even very low after five weeks of post-spray count. Furthermore, Sokolov (2000) reported that the application of chlorpyrifos at 205g a.i. ha^{-1} for control of range land grasshopper had a significant impact on population of carabid beetle and the population reached to its pre-spray level after three weeks of the spray. Generally, the insecticide treated plots showed slight improvement but did not reach the pre - spray level.

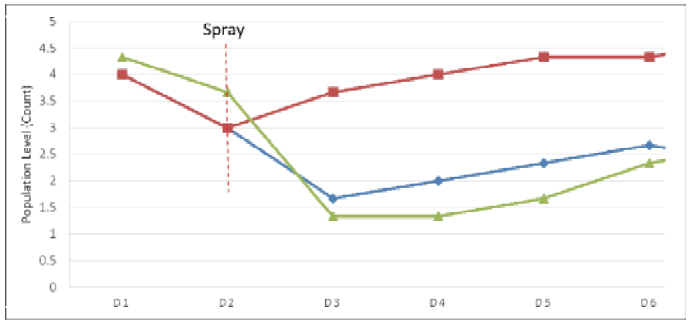

Figure 5. Population count of Carabidae (Coleoptera) from pitfall trap

Over all pitfall trapped arthropod species

The highest arthropod counts/catch recorded during the first two days of observation in pre-sprays counts (D1 and D2) was 24.4 and 23 per cent from the total population catches, respectively (table 2), whereas the lowest catches were collected in D3 with total catches of 9.5 per cent followed by 10.3 in D4.

Table 2. Total arthropod species trapped by pitfall throughout the experimental time

Trap Type		Observation Days							Total
		D1	D2	D3	D4	D5	D6	D7	
Pitfall trap	Count	965.4	910.6	374.8	408.2	390.2	414.8	495	**3959**
	(%)	24.4	23	9.5	10.3	9.9	10.5	12.4	**100**

Arthropod species which can work as bio-indicator

Some trapped arthropod families can serve as bio-indicators for monitoring the impact of insecticide on Desert Locust insecticide control operations. Those that can serve as bio-indicators are Formicidae and Gryllidae, These were found in abundance and were sensitive to the applied insecticide playing a key role in structure and function of the ecosystem. Therefore, these fulfill the criteria stated by Helena (2008), that the species which are abundant, representative of the arid ecosystem play a key role in structure and function of an ecosystem, are essential for monitoring program during insecticide control of Desert Locust. Thus, they can serve as bio-indicator. Besides, when the frequency and occurrence of the above mentioned families are certain, they can be used as bio-indicator in locust prune areas as described by McGeoch *et al.* (2002). Furthermore, according to FAO (1988) due to their abundance and sensitivity to insecticides applied for control of locust, Ants can be used as bio-indicator in locust insecticide control operations.

The study revealed highest catches of formicids (Hymenoptera) with a total trap count of 2,819 (71.2%) ants, followed by Gryllidae (crickets) with 709.4 (18%). This indicates that the population of these two families are highly abundant in arid ecosystem of Desert Locust habitats (Table 3). In the contrary, the lowest population count was observed for Arenae (spiders) 104.8 (2.6%).

Impact of treatments on non-target population

The result on impact of pesticides on non-targeted arthropod population show 86.2 per cent reduction in fenitrothion treated plots (table 4). Whereas in chlorpyrifos treated plots, the population count of non-target arthropods was reduced to 81.1 per cent as compared with mean of pre-spray count (D1 and D2). Fenitrothion was found to be most poisonous insecticide to non-targeted arthropods. Moreover, the reduction at non-target arthropods in all the post-spray population count (D3 – D7) due to fenitrothion was 78.8 per cent. Likewise, chlorpyrifos treated plots 72.7 per cent of reduction as compared with mean of pre-spray count (D1 and D2). However population reduction in control plots in post-spray spray period was only 9 per cent. The reduction in control plots was probably due to weather fluctuations.

Droplet drift measurement

Maximum number of droplets were was recorded at a distance of 7 m from the boarder of the plots with droplets/ cm^2 (Figure 6). The sprayed insecticide drifted up to 25 m with 3.5 droplets/cm^2. As droplets are carried by the wind, they may further go up higher than recorded and affect non-target species in the area when accompanied by high wind

Table 3. Arthropod families consistently found in Desert Locust habitat

Order	Family	Common name	Total catch count	Relative abundance (%)
Hymenoptera	**Formicidae**	Ants	2819	**71.2**
Orthoptera	**Gryllidae**	Crickets	709.4	18
Coleoptera	Tenebrionidae	Darkling Beetles	138.4	3.5
Coleoptera	Carabidae	Ground Beetles	187	4.7
Arenaea	-	Spiders	104.8	2.6
	Total		**3959**	**100**

*R% = Relative abundance in percentage

Table 4. Impact of treatments on overall non-targeted population mean

Treatment	Observation days								Mean		
	Day 1	Day 2	Day 3	Population reduction in D3 (%)	Day 4	Day 5	Day 6	Day 7	Pre-spray	Post-spray	Pop. Red. (%)
Chllorpyrifos	114	108	21	81.1	25	25	30	41	111	30.25	72.7
Fenitrothion	104	99	14	86.2	18	17	20	31	101.5	21.5	78.8
Control	103	96	91	8.5	93	88	88	93	99.5	90.5	9

D = day of observation

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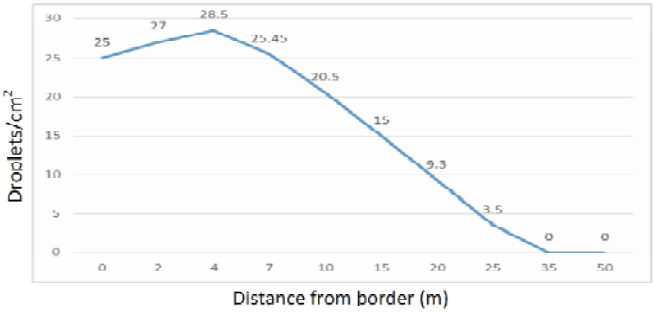

Figure 6. Droplets drift from the sprayed plots

speed and high emission height. Helena (2008) reported aerial application of chlorpyrifos in coastal area of Red Sea (Sudan) at low wind speed 2.7 m s^{-1} can move 1000 m from target area. Thus, it is important to understand proper weather condition and calibration method during the application of ULV formulations to minimize environmental hazard.

CONCLUSION

The study concluded that the insecticides used in the control of desert locust left highly significant impact at $P \leq 0.05$ on non-targeted arthropods as compared to the mean of pre-spray period. The use of fenitrothion 96 per cent ULV and chlorpyrifos 45 per cent ULV against Desert Locust, using an ordinary method of application, impacted non-targeted arthropods, especially the soil dwelling insects the most. However, fenitrothion showed relatively higher impact on arthropods.

The study further suggests that the two arthropods families, the formicidae (Hymenoptera) and gryllidae (Orthopteran) found in abundance and are sensitive to applied insecticides can be used as bio-indicators for monitoring the impact of insecticidal applications against the locust. Some of these findings can be used in the integrated pest management programme along with the fungus *Beauveria bassian Metarhizium anisoplae,* neem oil the neem leaf extracts.

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Short communication

Comparative biology of invasive fall armyworm, *Spodoptera frugiperda* Smith (Lepidoptera: Noctuidae) on maize, castor and artificial diet

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ABSTRACT

The study on biology of fall armyworm, *Spodoptera frugiperda* (J. E. Smith) (Lepidoptera: Noctuidae), conducted on different natural hosts (maize and castor) and artificial diet under laboratory conditions (26 ± 1° C, 75 to 80% RH) revealed host and diet dependent variations in biological parameters (incubation period: mean ± SE, 2.60 ± 0.09, 2.50 ± 0.09 and 2.83 ± 0.13 days; larval duration: mean ± SE, 17.03 ± 0.33, 18.53 ± 0.34 and 21.14 ± 0.20 days; pupal period: mean ± SE, 10.13 ± 0.24, 10.63 ± 0.23 and 11.40 ± 0.25 days; male adult longevity: mean ± SE, 8.30 ± 0.20, 8.10 ± 0.14 and 7.73 ± 0.13 days and female adult longevity: mean ± SE, 10.33 ± 0.19, 9.80 ± 0.14 and 9.76 ± 0.13 days on maize, castor and artificial diet, respectively. Differences were also noticed in the moth's fecundity, pre-oviposition, oviposition, post-oviposition and the total life periods.

Key words: Fall armyworm, maize, castor, artificial diet and biology

The fall armyworm (FAW), *Spodoptera frugiperda* (J.E. Smith) (Lepidoptera: Noctuidae) is an extremely polyphagous pest native to the tropical as well as sub-tropical regions of America. The pest having a great migration ability has invaded 47 African countries, 18 Asian countries and now Australia in the past three years and dangerously threatens crop production (https://www.cabi.org/isc/datasheet/29810). Recently, the occurrence of fall armyworm on maize has been reported in Karnataka, India. The pest has further moved to Indian states of Andhra Pradesh, Gujarat, Madhya Pradesh, Maharashtra, Tamil Nadu and Telangana (Sharanabasappa *et al.*, 2018; Ganiger *et al.*, 2018; Shylesha *et al.*, 2018). The FAW has recorded exceptionally wide host range (>353 host plants; Montezano *et al.*, 2018) and the most frequently damaged host plants are field maize, sorghum, rice, millet, soybean, peanut, cotton, sudan grass and other fodder grasses (CABI, 2018; Montezano *et al.*, 2018; Casmuz *et al.*, 2010). At present in India, the pest is predominantly found feeding on maize (Sharanabasappa *et al.*, 2018) and had been reported as pest on sorghum (Jaba *et al.*, 2020) and sugarcane (Chormule *et al.*, 2019). In India, during the non-existence of the maize crop, the pest may shift to other available host plants infusing economic damage and thus, upholding the residual population throughout the year as the climate is congenial for its development. Understanding the biology of *S. frugiperda* on different host plants is essential as it helps to better understand the bio-ecology of this pest and to develop sustainable integrated pest management strategies. Hence, the present study was undertaken.

Studies on natural host maize and artificial diet were initiated with the larvae collected from the maize fields at Central Research Station, Odisha University of Agriculture and Technology (OUAT), Bhubaneswar, Odisha and the study on host plant, castor was conducted at the Department of Entomology and Nematology, ICAR-IIHR, Bengaluru, Karnataka during January – March, 2020 and 2021, respectively. The culture of *S. frugiperda* on maize and castor was maintained as explained in the protocol of Sharanabasappa *et al.* (2018) and the emerged adults were used to establish a laboratory colony. The larvae were reared on the cut leaf bits of maize/castor and artificial square shaped diet pieces (5 g; prepared from the composition mentioned in the table 1, placed in plastic cell well boxes with closed lid) and maintained at ambient conditions (26 ± 1° C, 75 to 80% RH). The emerged adults (one pair) were placed in oviposition cages (30 x 45 cm) lined with paper towels as oviposition substrate and covered with nylon mesh on upper side. The moths were fed on10 per cent sucrose solution soaked on cotton pads offered in small plastic caps inside the cages and replaced daily. Eggs laid were collected and kept in a circular insect breeding dish until hatching.

The eggs were then examined at an interval of 12 h and the emerging larvae (n = 30) were reared individually and examined at an interval of 24 h until pupation.

Observations on larval, pupal, incubation, pre-oviposition, oviposition, post-oviposition periods, fecundity and adult moth's (B& and @&) longevity were observed at an interval of 24 h and the data were subjected to mean data analyses.

Table 1: Artificial diet for fall armyworm, *Spodoptera frugiperda* (Jaba *et al.*, 2020)

Ingredients	Quantity (g or ml)
Fraction A	
Ascorbic acid	7.5 g
Carbendazim (Bavistin)	1.0 g
Chickpea flour	265.0 g
Formaldehyde	5.0 ml
Maize leaf powder	75.6 gm
Methyl-p hydroxy benzoate	6.0 gm
Multi vitamin	2 Capsules
Sorbic acid	3.9 g
Vitamin E	5 Capsules
Water	1210.0 ml
Yeast	68.0 g
Fraction B	
Agar agar	38.0 g
Water	1210.0 ml

The results revealed significant differences in the fecundity and incubation period of *S. frugiperda* when reared on maize, castor and diet. Gravid female laid eggs in clusters which were dorso-ventrally flattened and covered with a layer of scales showing moldy like appearance. The number of eggs (Mean ± SE) laid were 977.93 ± 14.99, 941.83 ± 13.09 and 925.83 ± 20.05 when the moths were reared on maize, castor and artificial diet, respectively. An incubation period (Mean ± SE) of 2.60 ± 0.09, 2.50 ± 0.09 and 2.83 ± 0.13 were recorded on maize, castor and diet, respectively (Table 2). Similar results were reported earlier by Sharanabasappa *et al.* (2018) and Jaba *et al.* (2020) on maize and diet.

Newly hatched caterpillars are green in colour during their first and second instars and turn brown to black from third to sixth instars. Caterpillars have a dark head with a pale, upside down Y-shaped mark on the frontal side of head capsule. Each larva passed through six distinct instars over a period of 17.03 ± 0.33, 18.53 ± 0.34 and 21.14 ± 0.20 days on maize, castor and diet, respectively (Table 2). These results are similar to previous studies made by Pitre and Hogg (1983) and Sharanabasappa *et al.* (2018), who reported the larval period as 14-30 days on maize. Whereas Jaba *et al.* (2020) reported 20 days on the diet, which is in line with our current study.

Table 2: Biology of fall armyworm on maize, castor and artificial diet

Parameters (Mean ± SE)	Maize	Castor	Artificial diet
I Duration (Days)			
a. Incubation period	2.60 ± 0.09	2.50 ± 0.09	2.83 ± 0.13
b. Larval period	17.03 ± 0.33	18.53 ± 0.34	21.14 ± 0.20
c. Pupal period	10.13 ± 0.24	10.63 ± 0.23	11.40 ± 0.25
d. Pre oviposition period	3.60 ± 0.13	3.56 ± 0.10	3.53 ± 0.09
e. Oviposition period	2.43 ± 0.09	2.50 ± 0.09	2.73 ± 0.08
f. Post oviposition period	4.90 ± 0.12	3.63 ± 0.12	3.53 ± 0.15
g. Male adult longevity	8.30 ± 0.20	8.10 ± 0.14	7.73 ± 0.13
h. Female adult longevity	10.33 ± 0.19	9.80 ± 0.14	9.76 ± 0.13
i. Total life cycle (♂)	38.07 ± 0.52	39. 77 ± 0.43	43.10 ± 0.34
j. Total life cycle (♀)	40.10 ± 0.49	41.46 ± 0.44	45.13 ± 0.38
II Fecundity/♀ (No)	977.93 ± 14.99	941.83 ± 13.09	925.83 ± 20.05

During the pre-pupal period, the full-grown larva stopped feeding, turned greenish and looked bright brown colour. Duration of the pupal period (Mean ± SE) was about 10.13 ± 0.24, 10.63 ± 0.23 and 11.40 ± 0.25 days on maize, castor and diet, respectively (Table 2). Similar results were reported by Sharanabasappa *et al.* (2018) on maize and by Debora *et al.* (2017) and Jaba *et al.* (2020) on diet.

The forewings of adult male are shaded with grey and brown, with triangular white patches at the apical region and circular spot at the center of the wing. The forewings of female have uniform greyish brown to a fine mottling of grey and brown. The hind wing is silver-white with a narrow dark border in both male and female. These morphological characters are similar as reported in earlier studies (Oliver and Chapin, 1981; Sharanabasappa *et al.*, 2018 and Jaba *et al.*, 2020).

The mean pre-oviposition, oviposition and post-oviposition periods of *S. frugiperda* on maize was 3.60 ± 0.13, 2.43 ± 0.09 and 4.90 ± 0.12, respectively on castor (3.56 ± 0.10, 2.50 ± 0.09 and 3.63 ± 0.12, respectively on castor and 3.53 ± 0.09, 2.73 ± 0.08 and 3.53 ± 0.15, respectively on the artificial diet recording minimal variations across the treatments. The lifecycle of male and female moths was found to be maximum with artificial diet (43.10 ± 0.34 and 45.13 ± 0.38 respectively) followed by castor (39. 77 ± 0.43 and 41.46 ± 0.44 respectively) and was found to be the least with maize (38.07 ± 0.52 and 40.10 ± 0.49 respectively). The adult female moths survived longer (10.33 ± 0.19, 9.80 ± 0.14 and 9.76 ± 0.13 days) compared to male moths (8.30 ± 0.20, 8.10 ± 0.14 and 7.73 ± 0.13) on all the treatments studied viz., maize, castor and diet, respectively (Table 2). These findings corroborate with those of Sharanabasappa *et al.* (2018) and Jaba *et al.*, (2020) on maize as host and diet as well. Further, it was noted that minimal oviposition period, incubation period, larval period, pupal period and adult longevity (@&

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and B&) were recorded in the maize which is a natural host of FAW followed by alternate host, castor and artificial diet. Besides that, oviposition period, post oviposition period and fecundity were greater in maize followed by castor and artificial diet.

CONCLUSION

The information on biology of *S. frugiperda* studied on different host plants and artificial diet may be utilized in developing sustainable integrated management strategies for fall army worm.

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Short communication

On farm trails on management of whitefly, *Bemisia tabaci* in cotton

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ABSTRACT

The on farm trial (OFT) conducted to manage whitefly Krishi Vigyan Kendra, Lunkaransar of Bikaner (Rajasthan) for the two consecutive years during *Kharif,* 2019 and 2020 indicated maximum per cent reduction of 82.70 and 88.75 during *Kharif* 2019 and 2020, respectively in whitefly population at recommended practices of spray with pyriproxyfen 10 EC (@ 01 litre ha^{-1}) . Minimum percent reduction of whitefly population (73.26 and 75.58 per cent) during *Kharif* 2019 and 2020, respectively was recorded under the farmers practices (T_2 spray of acetamiprid 20 SP @0.4 g $litre^{-1}$). The maximum benefit cost ratio, 3.19 and 4.15, was obtained in the recommended practices while it was minimum in farmer's practice 2.41 and 3.29 during *kharif* 2019 and 2020, respectively.

Key Words : Cotton**,** whitefly, pyriproxyfen, acetamiprid , benefit cost ratio

Cotton, popularly known as "White gold" is an important fibre crop cultivated in India. It is mainly grown for seed products including lint, hulls and oil etc. The production and productivity of this crop is affected by the several biotic constraints, among this the insect pests are one of the important factor. Many pests attacks the crop from germination to harvesting. Patil (1998), reported 30-80 per cent yield losses in cotton due to insect pests. According to Choudhary (2000), 184 insect pests in India have been recorded on cotton and the major among these is the whitefly, *Bemisia tabaci* (Hemiptera: Aleyrodidae) which is widely distributed sucking pest of this crop. Whitelfy suck the phloem sap of more than 500 host plant species (Hunter and Polston, 2001), causing damage by the excretion of large amounts of honeydew which serves as a medium for black sooty mold fungi. The mold reduces the photosynthesis rate and tin turn results in lowering the flower production, a smaller number of bolls and low-quality end product. Whitefly can transmit more than 90 types of plant virus (Jorge and Mendoza, 1995; Hunter and Polston, 2001) including the tomato yellow leaf curl virus (Ghanim and Czosnek, 2000), the sweet potato leaf curl virus (Lotrakul et al., 1998) and the tomato mottle virus (Hunter et al., 1998). In cotton it's also act as vector of cotton leaf curl disease, which causes heavy losses in cotton. In many agricultural ecosystems worldwide, it is well known that whitefly showed resistance against organophosphate and pyrethroid insecticides (Horowitz et al., 1998). Hence, a farm trial for the management of the whitefly in cotton through new group of insecticide was carried out.

The experiment was conducted by Krishi Vigyan Kendra, Lunkaransar, Bikaner (Rajasthan) OFT's at ten farmers' field in different villages during (*Kharif*) 2019 and 2020. The experiment material consisted of two treatments *i.e.* Farmers' practice spray with acetamiprid 20 SP @ 0.4g $litre^{-1}$ (T_1) and the recommended practice spray of pyriproxyfen 10 EC @ 01 litre ha^{-1} (T_2).

The percentage reduction in whitefly population was calculated by using the formula given by Henderson and Tilton (1955), which is the modification of Abott's (1925) formula.

$$\text{Per cent reduction in population} = 100\left[1 - \frac{\text{TaXCb}}{\text{TbXCa}}\right]$$

Where,

Ta = Number of insects after treatment

Tb = Number of insects before treatment

Ca = Number of insects in untreated check after treatment

Cb = Number of insects in untreated check before treatment

The avoidable loss and increase in seed cotton yield over control was calculated for each treatment by the following formula:

$$\text{Avoidable loss (\%)} = \frac{\text{Highest yield in treated plot} - \text{Yield in the treatment}}{\text{Highest yield in the treated plot}} \text{ X } 100$$

$$\text{Increase in yield (\%)} = \frac{\text{Yield in treatment} - \text{Yield in control}}{\text{Yield in control}} \text{ X } 100$$

The data presented in table 1 revealed highest percent reduction in whitefly population i.e. 82.70 and 88.75 per cent during *kharif* 2019 and 2020, respectively through use of recommended practices (T_2-spray of pyriproxyfen 10 EC @ 01 litre ha^{-1}). The minimum percent reduction in whitefly population (73.26 and 75.58 per cent during *kharif* 2019 and 2020, respectively) was recorded in farmers practices (T_2 spray of acetamiprid 20 SP @0.4g litre^{-1}). Highest cotton seed yield was obtained in the recommended practices during both the years *i.e.* 25.54 and 29.90 q ha^{-1} against the minimum of 20.79 and 24.92 q ha^{-1} in *Kharif* 2019 and 2020, respectively under the farmers practices. The per cent increase in cotton seed yield was observed 22.84 and 19.98 q ha^{-1} in the recommended farmers practice during *Kharif* 2019 and 2020, respectively. The per cent avoidable losses in seed cotton yield was observed maximum (18.59 and 16.65 per cent during *Kharif* 2019 and 2020, respectively) in farmer's practices while, avoidable losses were zero in recommended practices. The highest net return (Rs 107975 and Rs 140368 ha^{-1}) and benefit cost ratio (3.19 and 4.15) was obtained in the recommended practices during *Kharif* 2019 and 2020, respectively. This may be due to the higher yield obtained by the proper management of whitefly. Lowest net return (Rs 81613 and Rs 111350 Rs ha^{-1}) and benefit cost ratio (2.41 and 3.29) was obtained in the farmers practices during *Kharif* 2019 and 2020. This findings also corroborates with the findings of Ahmad *et al.*, (2014). They recorded maximum (80.14 %) reduction in whitefly population in the treatment of pyriproxyfen followed by imidacloprid (78.94%) and acetameprid (78.77). Swami *et al.*, (2018) also recorded highest reduction in whitefly population with pyriproxyfen 10 EC in chilli crop. These results were supported by Kumar *et al.*, (2014) who reported highest population reduction in whitefly population and the highest seed cotton yield with in the pyriproxyfen 10 EC treatment.

Table 1: Performance of OFT in the Management of whitefly on cotton during *Kharif*, 2019 and 2020

Year	Technology option	Percent reduction in whitefly population	Yield (q ha^{-1})	Per cent increase in yield over farmers practice	Total avoidable losses (q ha^{-1})	Total avoidable losses (%)	Net returns (Rs.)	B:C ratio
2019	T_1- Farmer practice: spray of acetamiprid 20 SP @0.4g/litre	73.26	20.79	-	4.75	18.59	81613	2.41
	T_2- spray of pyriproxyfen 10 EC @ 01 litre/ha	82.70	25.54	22.84	-	-	107975	3.19
2020	T_1- Farmer practice: spray of acetamiprid 20 SP @0.4g/litre;	75.58	24.92	-	4.98	16.65	111359	3.29
	T_2- spray of pyriproxyfen 10 EC @ 01 litre/ha	88.75	29.90	19.98	-	-	140368	4.15

CONCLUSION

It was concluded that spraying of cotton crop with pyriproxyfen (10 EC @ 1 lit ha^{-1}), that helps in regulating the transition of whitefly from one developmental stage to another, can be used in effective management of whitefly.

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Seasonal fluctuations in the population of *Tetranychus urticae* Koch on French marigold under screen house conditions

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ABSTRACT

The study carried out during summer, rainy and winter seasons showed prevalence of spider mite, *Tetranychus urticae* on marigold plants in the screen house throughout the year, the maximum population observed during third week of April (144.95 mites leaf^{-1}) and fourth week of November (130.59 mites leaf^{-1}). Grown up leaf of middle canopy harbored maximum population with an average of 85.86 mites leaf^{-1} followed by tender (60.78 mites leaf^{-1}) and older leaves (28.60 mites leaf^{-1}). During summer, an average number of 86.46 mites leaf^{-1} followed by 52.60 mites leaf^{-1} in winter and 36.18 mites leaf^{-1} during rainy season was recorded.

Key words: summer, winter, rainy, two spoted spider mite

French marigold (*Tagetes patula*) is an important commercial flower crop. The genus *Tagetes* comprises nearly 33 species of which, the french marigold is under commercial cultivation in India for loose flower production (Netam 2017). It is mainly cultivated in Maharashtra, Karnataka, Gujarat, Andhra Pradesh, Tamil Nadu, Madhya Pradesh and Haryana. Because of being highly suitable for cultivation under different argo-climatic conditions in Haryana, is grown year round in summer, rainy and winter seasons. The estimated area under marigold cultivation in India is about 64 thousand ha with a production of 608.97 thousand MT (Anonymous 2017).

However, its successful cultivation is threatened by a number of pests. Among these, the two-spotted spider mite, *Tetranychus urticae* Koch (Acari: Tetranychidae) has achieved a major pest status (Pateladiti *et al*. 2016). This is an economically important and ubiquitous agricultural pest of french marigold. It is known to attack nearly 1200 species of the plants of which 150 are economically important (Zhang 2003). This is attributed to its high fecundity birth rate, rapid population growth, long adult survival and short developmental time (Clotuche *et al*. 2011).

Two spotted spider mite feeds on leaf by puncturing the cells with its stylets and draining the sap content, thereby producing a characteristic yellow specking on the leaf surface. The chlorotic damage and webbings produced by deutonymphs, protonymphs and adults hampers the plant's ability to carry out photosynthesis resulting in reduction of the total crop yield. Its infestation represents a potential biotic stress to its host plant. Heavy damage may cause leaves to dry and drop (Abdel-Wali *et al*. 2012). The silvering of marigold leaves is the result of mite feeding on the leaf cells. *T. urticae* causes reduction in the economic yield of crops ranging from 20-45 per cent depending upon agro-climatic conditions and cropping season.

With a view to develop its life table, the fluctuation in the population of *Tetranychus urticae* in different seasons and also on different leaf stages (*viz*., older, tender and grown) of marigold was studied for its effective management.

MATERIALS AND METHODS

The study was carried out in the screen house of Department of Zoology, College of Basic Sciences and Humanities, Chaudhary Charan Singh Haryana Agricultural University, India during summer (March to May, 2018), rainy (June to September, 2018) and winter (October, 2018 to March, 2019) season. Seeds of french marigold were grown in screen house under replicated conditions. Standard agronomical practices were followed to raise the crop and were kept free from any insecticidal contamination. *T. urticae* were reared to have pure stock culture on few french marigold plants and to accomplish it, unsprayed infested marigold leaves with live *T. urticae* mites were placed on the leaves of fresh potted marigold plants. These mites were allowed to naturally migrate to the uninfested plant from the infested plants by placing 7-10 days old healthy seedling near the infested plants.

The observations were recorded at weekly intervals. Ten plants were randomly selected and mobile stages (mixed population) of *T. urticae* were counted per leaf from three leaf stages covering from the top (tender leaves), middle (grown up leaves) and bottom (older leaves). Two leaves from each stage of marigold plant were taken. These were bought to acarology laboratory for recording the mite population under the stereo zoom binocular microscope. Also, mobile stages at the screen house itself were noted with the help of hand

lens. Population of mite was recorded from both the dorsal as well as the ventral surface of the leaf. To study the spatial distribution of mite, symptoms of mite infestation on leaf were also examined. Data was recorded up to the time of harvesting in each season.

'OPSTAT', an online statistical analysis tool developed at the Computer Centre of College of Basic Sciences and Humanities, CCS Haryana Agricultural University, Hisar, was used for the data analysis. To record the effect of observation period, leaf stage and season on the population dynamics of *T. urticae*, critical difference was calculated by applying ANOVA (Analysis of variance) under two factor Completely Randomized block Design (CRD) and the statistical significance of data was assessed.

RESULTS AND DISCUSSION

T.urticae incidence in the screen house was observed in all the three seasons on dorsal and ventral surfaces of leaves flowers. Its population increased to its maximum in third week of April, 2018 (Table 1) at maximum temperature, morning relative humidity, sunshine hours and evaporation of 33.66°C, 72 per cent, 7.01 hours, 5.40 mm, respectively. The re-after, mite population gradually and significantly declined to 90.12 in the fourth week of May. Grown up leaves harboured significantly higher mite population (121.90 mites leaf^{-1}) than the tender (91.33) and older (46.16) leaves. The analysis of data revealed a significant effect of the observation period on population buildup of *T. urticae* (CD = 1.53; p = 0.05) (Table 1).

Table1. Effect of leaf stage on *T.urticae* population during summer season

Observation period	Average number of *T. urticae* leaf^{-1}			
	Tender leaf	Grown up leaf	Older leaf	Mean
3rd Week March, 2018	3.59	7.65	6.63	5.96
4th Week March, 2018	13.24	23.40	11.20	15.95
1st Week April, 2018	29.49	44.73	26.95	33.73
2nd Week April, 2018	75.21	86.39	38.64	66.75
3rd Week April, 2018	160.05	190.53	84.26	144.95
4th Week April, 2018	152.43	186.97	73.69	137.70
5th Week April, 2018	145.83	178.84	65.56	130.08
1st Week May, 2018	123.98	170.72	59.97	118.22
2nd Week May, 2018	110.27	163.10	54.39	109.25
3rd Week May, 2018	99.60	150.40	45.24	98.41
4th Week May, 2018	90.96	138.20	41.18	90.12
Mean	**91.33**	**121.90**	**46.16**	

CD (p=0.05) for Observation Period =1.53; S E (m) = 0.54
CD (p=0.05) for Leaf stage =0.80; S E (m) = 0.28
CD (p=0.05) for Observation Period× Leaf stage=2.65; S E (m) = 0.94

The week wise data showed that the mite population was less during rainy season as compared to the summer season (Table 2). Distribution of *T. urticae* was significantly more on grown up leaves (58.44 mites leaf^{-1}) which was statistically higher (CD = 0.26; p = 0.05) than the mite population recorded on tender leaves (34.44 mites leaf^{-1}). Least number of mites was found on the older leaves (15.66 mites leaf^{-1}). Statistical analysis showed a significant effect of observation period (CD =0.66, p = 0.05). Irrespective of leaf age, statistically higher number of mites was recorded during first week of June, 2018 (81.98, mites leaf^{-1}) as compared to the mites number recorded during the rainy season. The ANOVA also revealed a significant interaction between the observation periods and the leaf stage of plants (CD =1.14, p = 0.05).

Table 2: Effect of leaf stage on *T.urticae* population during rainy season

Observation period	Average number of *T. urticae* leaf^{-1}			
	Tender leaf	Grown up leaf	Older leaf	Mean
1st Week June, 2018	81.30	129.06	35.58	81.98
2nd Week June, 2018	72.67	103.66	31.52	69.28
3rd Week June, 2018	65.56	99.09	27.97	64.20
4th Week June, 2018	56.92	89.94	23.90	56.92
1st Week July, 2018	54.38	81.81	20.85	52.35
2nd Week July, 2018	47.27	73.18	18.31	46.25
3rd Week July, 2018	38.63	66.57	16.28	40.49
4th Week July, 2018	31.52	60.48	14.76	35.58
5th Week July, 2018	29.49	56.41	12.73	32.88
1st Week August, 2018	22.89	53.87	11.71	29.49
2nd Week August, 2018	17.81	51.33	10.69	26.61
3rd Week August, 2018	15.77	45.74	9.68	23.73
4th Week August, 2018	14.25	26.44	8.66	16.45
1st Week September, 2018	11.71	16.79	7.65	12.05
2nd Week September, 2018	10.18	14.762	6.63	10.52
3rd Week September, 2018	8.66	12.73	5.10	8.83
4th Week September, 2018	6.63	11.71	4.09	7.48
Mean	**34.44**	**58.44**	**15.66**	

CD (p=0.05) for Observation Period =0.66; S E (m) = 0.23
CD (p=0.05) for Leaf stage =0.26; S E (m) = 0.09
CD (p=0.05) for Observation Period× Leaf stage=1.14; S E (m) = 0.41

The *T. urticae* population recorded during winter (table 3) showed that the grown up leaves harboured significantly greater mite population (77.22 mites leaf^{-1}) as compared to tender (59.56 mites leaf^{-1}) and older ones (23.99 mites leaf^{-1}) (CD = 0.38, p = 0.05). The analysis of data showed a significant effect of the observation period on population buildup of mite (CD = 1.03; p = 0.05) and irrespective of leaf stage, statistically higher number of mites were recorded in the fourth week of November (130.59 mites leaf^{-1}) than the numbers recorded on other observation periods during winter season. After a peak, decline in mite population was observed up to fourth week of February.

Table 3: Effect of leaf stage on *Tetranychus urticae* population during winter season

Observation Period	Average number of *T. urticae* leaf^{-1}			
	Tender leaves	Grown up leaves	Older leaves	Mean
1stWeek October, 2019	10.70	20.35	4.60	11.88^{a}
2nd Week October, 2019	15.78	33.56	13.75	21.03
3rd Week October, 2019	40.16	67.08	20.86	42.70
4th Week October, 2019	66.07	89.95	34.07	63.36
1stWeek November, 2019	77.24	102.14	44.23	74.54^{b}
2nd Week November, 2019	87.91	120.43	57.94	88.76^{c}
3rd Week November, 2019	112.3	142.78	64.04	106.37
4th Week November, 2019	140.24	183.42	68.10	130.59
5th Week November, 2019	130.08	169.19	58.45	119.24
1st Week December, 2019	120.43	142.78	42.19	101.80
2nd Week December, 2019	111.79	127.54	28.99	89.44^{c}
3rd Week December, 2019	101.12	104.68	19.33	75.04^{b}
4th Week December, 2019	75.21	85.37	13.24	57.94
1st Week January, 2019	58.96	67.08	8.16	44.73
2nd Week January, 2019	34.07	44.73	6.12	28.31
3rd Week January, 2019	26.95	37.62	4.09	22.89
4th Week January, 2019	16.28	23.40	2.57	14.08
1st Week February, 2019	9.68	12.73	1.85	8.09
2nd Week February, 2019	11.71	17.3	5.11	11.37^{a}
3rd Week February, 2019	15.78	26.44	7.14	16.45
4th Week February, 2019	21.36	36.60	9.17	22.38
1st Week March, 2019	26.44	43.72	13.75	27.97
Mean	**56.56**	**77.22**	**23.99**	

CD (p=0.05) for Observation Period =1.03; S E (m) = 0.37
CD (p=0.05) for Leaf stage = 0.38; S E (m) = 0.14
CD (p=0.05) for Observation Period× Leaf stage=1.78; S E (m) = 0.64
Values with the same superscript do not differ significantly

In a previous study, Mahato *et al*. (2008) in West Bengal also recorded peak mite population on marigold during the summer (second week of March). Similarly, Ganai *et al* (2018) reported abundant mite population in March. *T. urticae* overwinter as fertilized female adults (Cone *et al*. 1986). Raworth (2007) in Canada revealed that oviposition was most likely to occur during March and the adults develop from winter diapause at the initiation of the summer season. Das (1959) reported that heavy rainfall during rainy season led to reduction in mite population in tea plantations. Reduction in *T. urticae* population at cooler temperature during winter and high humidity during rainy season may be attributed to increased activity of pathogens.

The variations in the sowing time, crop varietiy and climatic conditions also affected the population dynamics of *T. urticae*. Rishi and Rather (1983) found maximum build up of *T. urticae* population during July-August at 30-32°C while, winter forms generally developed at the end of October (15-20°C). In a study on okra, the peak activity of *T.urticae* was recorded in the months of April (Natarajan 1989) and in November on cowpea (Haque *et al*. 2011). Also high density of *T. urticae* population during the summer months (when temperature is high) have been reported in the past on crops such as okra (Gulati 2004; Geroh *et al*. 2010), peach (Riahi *et al*. 2011) and cucumber (Kanika *et al*. 2013) Severely attacked grown up leaves harborured more number of mites (85.86 mites leaf^{-1}) followed by tender (60.78 mites leaf^{-1}) and older leaves (28.60 mites leaf^{-1}) (Fig. 2). The occurrence of *T. urticae* on the top leaves was reported on brinjal (Dutta *et al*. 2012), Okra (Gulati, 2004), (gerberg Shah and Shukla, 2014) and on tomato (Shukla and Pokle, 2015). Abundant mite population on bottom strata of rose by Onkarappa *et al.* (1999), Mondal and Ara (2006) and Fitzgerald *et al*. 2008 has been reported on straw berry. Among the canopy levels, tetranychid preference of middle canopy was shown by Prasanna (2007 on brinjal, Geroh *et al*. (2010) on okra and Kanika *et al*. (2013) on cucumber. This may be due to the fact that when plants become overcrowded with mites and food resources become scarce, the individuals start gathering towards at the plant apex in search of food and for dispersal. Differences may also arise due to different crops studied and the variation of climatic conditions in different areas.

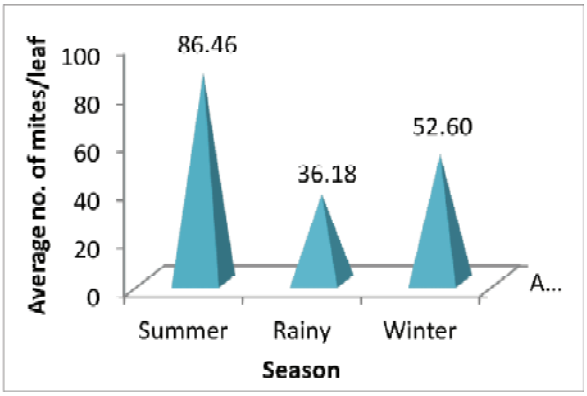

Fig.1: *Tetranychus urticae* population trends in summer, rainy and winter season

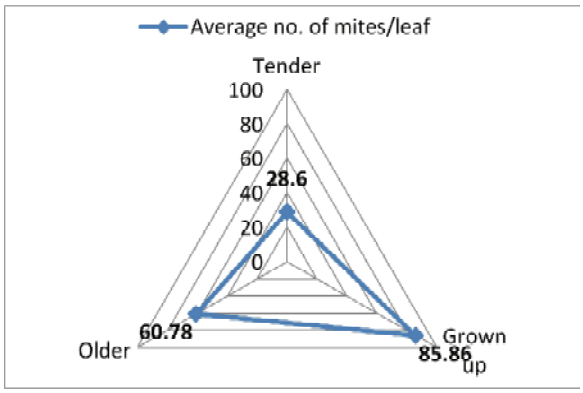

Fig.2: Effect of leaf stage on *T. urticae* population

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Fig. 3: Healthy french marigold plant

Fig.4: Webbing by *T. urticae* on infested french marigold plant

CONCLUSIONS

It was concluded that *T. urticae* activity in the screen house prevailed during all the three seasons *viz*., summer, rainy and winter. However, the severity of damage of french marigold plants was maximum during hot and dry period of April. The grown up leaves harboured maximum population of *T. urticae* followed by tender and older leaves.

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Influence of traps colour and height of its placement in trapping sucking pests and natural enemies in greengram

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ABSTRACT

The study conducted at Regional Agricultural Research station, Lam, Guntur, Andhra Pradesh in greengram for three consecutive years during 2019, 2020 and 2021 using three commercially available double sided yellow, blue, and white sticky traps placed at 0, 1, 2, 3, 4, 5 ft height from the crop canopy in bulk crop showed that blue sticky traps were more attractive to thrips, yellow sticky traps for whiteflies and leafminers. while, the yellow sticky traps were found detrimental to coccinellids. The trapping of insects reduced with the increase in trap height from the crop canopy irrespective of the trap colour. The traps placed upto 2 ft above the crop canopy recorded higher number of sucking pests as well as coccinellids in greengram.

Key words: Sticky traps, trap height, whiteflies, thrips, greengram.

Pulses are the main source of protein and minerals in common people diet besides playing a vital role in socio-economic status of India. India is a major pulse growing country in the world, sharing 35-36 per cent area and 27- 28 per cent production of pulse crops. It is producing 12-14 million tones of pulses from 22-24 million ha of land (Mahalakshmi *et al*., 2016). Among the pulses, greengram is an important pulse crop which is a rich source of carbohydrates (51%), proteins (26%), minerals (4%) and vitamins (3%) (Yadav *et al*., 1994). More than 70 % of the worlds greengram production comes from India (Usha *et al*., 2020), where, it is cultivated annually in an area of 3.83 mha with total production and average productivity of 1.603 mt and 418 kg/ha^{-1}, respectively (Anonymous, 2017). The average productivity of greengram remained static of late in India due to several reasons viz., lack of suitable seed production techniques, cultural practices, inefficient harvest and post-harvest operations, improper storage management practices, etc. Greengram crop suffers from sowing to harvesting by the attack of various insect pests causing the loss upto 73.86 per cent (Pandey *et al*., 1991). Among the different insect pests, sucking pests are the major ones which not only cause the severe yield loss but also because indirect damage by acting as vectors for most dreadful diseases like yellow mosaic virus disease, leaf curl and bud necrosis in greengram. These pests attack the crop from primary stage itself to end of the crop growth, though, several chemical pesticides have been recommended for combating sucking pests. Such reliance on insecticides has created many problems such as excessive residues in the produce, which are the concerns of general consumer health and the environment, pesticide resistance, poisoning, hazards to non-target organisms and increased production costs etc. (Kansagara *et al*., 2018). Therefore, there is a need to test out different non chemical methods to avoid further resistance in these pests. The sticky traps, a cultural control method can be used as a component of integrated pest management. Coloured sticky traps could be a simple and low cost method for determining the relative abundance of insects. It is necessary to determine the colour preference of sucking pests to get maximum catches of those insects. This information will improve monitoring of these pests and enhance integrated pest management programs (Pobozniak *et al*., 2020). Blue, yellow and white coloured sticky traps are commercially available for the control of some insect pests in many countries. However, the use of sticky traps for capturing species of herbivorous insect pests is risky also because they may also capture non-targeted predatory insects and reduce their numbers (Roth *et al*. 2016). Trap height also is important for mass trapping and monitoring insect populations (Byrne *et al*. 1986). Hence, the present study was conducted to know the trapping efficacy of different coloured sticky traps and height of placement of sticky traps in greengram.

MATERIALS AND METHODS

The study was conducted at Regional Agricultural Research station, Lam, Guntur, Andhra Pradesh on greengram for three consecutive seasons i.e during *Rabi*, 2019, 2020 and 2021. For the experiment, the popular greengram variety in Andhra Pradesh, LGG 460 was sown in an area of 4000 sq.m and the crop was kept completely under unprotected conditions for sucking pests, but selective insecticides were sprayed to protect the crop from pod borers

through blanket sprays after flowering initiation. Three commercially available double sided sticky traps such as yellow, blue, and white sticky traps measuring 10 X 24 CM (W X L) were procured locally. The traps were attached to a wooden stake so that their bottom edge was placed in such a way to turn up at 0, 1, 2, 3, 4, 5 ft height from the crop canopy in bulk crop. The experiment was conducted in factorial RBD design with three replications, thus a total of eighteen traps were used for each colour at three different places in the bulk crop. The double sided sticky traps were placed immediately after sowing and maintained upto 45 days after sowing i.e. during vegetative phase of crop growth when the peak activity of sucking pests will be there in the crop. The data was collected at 15 days interval i.e. at 15, 30 and 45 days after sowing and the sticky traps were replaced with fresh traps after data collection and height adjusted according to the crop height. Care was taken to avoid the accidental trapping of insects by the different coloured sticky traps by placing the three different coloured sticky traps side by side for each specified height of placement.

The traps were collected and brought to the laboratory for data regarding at each interval. The data regarding sucking pests such as thrips and whiteflies and leaf miner adults was recorded from three spots of each one square centimeter area from each sticky trap from all the treatments. The data regarding natural enemies such as coccinellids and other insects such as hymenopterans, hemipterans, dipterans and other insects was recorded from the entire trap by counting all the population from both sides of the sticky traps. The data collected during the three seasons was pooled and analysed through two way analysis of variance (ANOVA) after normalizing the data by using proper transformations.

RESULTS AND DISCUSSION

Thrips

The number of thrips trapped varied significantly among the various coloured traps and height of trap placement. Among the various traps, more number of thrips was attracted towards blue coloured sticky traps (11.59 sq.cm^{-1}) when compared to white (6.59 sq.cm^{-1}) and yellow coloured traps (4.18 sq.cm^{-1}) which showed significant differences statistically (Table1 & Fig.1). Hence, blue coloured sticky traps can be considered as more effective in trapping thrips followed by white traps and yellow coloured traps were least effective in trapping the thrips. While the placement of traps also played significant role in trapping of the thrips. The number of thrips was more on the traps place at the height of crop canopy irrespective of trap colour. The mean number of thrips caught by the traps placed at the height of crop canopy (23.17 sq.cm^{-1}) was significantly superior over the remaining traps placed at various heights above the crop canopy. The trapping of thrips was highest at lowest height and vice versa. The mean number of thrips was gradually reduced when the height of trap increased from crop canopy irrespective of the colour of the trap (Table.1). The results are in accordance with several other researchers who reported the efficacy of blue sticky traps against thrips in many crops. Broadsgaard (1993) indicated that thrips were attracted to blue as well as to white colour. The blue traps always caught more adults than the yellow and white traps at an earlier time and in consistently higher numbers when compared to the numbers of thrips found on plants, indicating that blue traps are the most sensitive ones and could function as an early warning of onion thrips activity (Shelton *et al*., 2006). Blue and white have been considered as the most preferred colours for several species of thrips, including *Thrips tabaci* (Liu and Chu 2004; Eric *et al*., 2007). Blue and white have been considered as the preferred or the most preferred colors for several species of thrips in tomato (Ranamukhaarachchi and Wickramarachchi, 2007), in rose crop (Sridhar And Naik, 2015), in onion (Devi and Roy, 2017) and in pea fields (Pobozniak *et al*., 2020). Blue coloured traps were found more effective in trapping thrips compared to yellow, white and green and pink trap (Hossain *et al*., 2020). In contrast, Elango *et al*., (2017) reported more numbers of thrips attracted to yellow coloured sticky traps than other colours such as orange and red in pomegranate.

Table.1: Mean number of thrips on different sticky traps at different heights

Thrips	0 ft	1 ft	2 ft	3 ft	4 ft	5 ft	Mean
Yellow	8.22	5.93	4.66	3.23	2.10	1.05	4.18 (2.28)
White	12.29	9.55	7.51	5.44	3.28	1.49	6.59 (2.76)
Blue	24.93	16.88	12.77	8.54	4.71	1.72	11.59 (3.55)
Mean	15.15	10.79	8.31	5.74	3.33	1.42	
	(4.02)	(3.43)	(3.05)	(2.60)	(2.08)	(1.56)	
	Factors				C.D.	SE(d)	SE(m)
	Factor (A)				0.08	0.04	0.03
	Factor (B)				0.12	0.06	0.04
	Factor(A X B)				0.20	0.10	0.07

*Figures in parenthesis are SQRT (X+1) transformed values

Whiteflies

Yellow coloured sticky traps attracted significantly more number of whiteflies (3.45 sq.cm^{-1}) when compared to white (1.87 sq.cm^{-1}) and blue (1.37 sq.cm^{-1}) coloured sticky traps (table 2 & fig.1). The height of trap placement also showed significant differences in the trapping efficacy, since the mean number of whiteflies was more on the traps placed at the height of crop canopy which significantly superior

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Table 2. Mean number of whiteflies on different sticky traps at different heights

Whitefly	0 ft	1 ft	2 ft	3 ft	4 ft	5 ft	Mean
Yellow	6.77	5.28	4.04	2.90	1.27	0.44	3.45 (2.11)
White	3.66	2.71	2.66	1.32	0.46	0.18	1.83 (1.68)
Blue	3.55	2.28	1.43	0.73	0.21	0.00	1.37 (1.54)
Mean	4.66	3.42	2.71	1.65	0.64	0.21	
	(2.38)	(2.10)	(1.93)	(1.63)	(1.28)	(1.10)	
				Factors	**C.D.**	**SE(d)**	**SE(m)**
				Factor(A)	0.09	0.04	0.03
				Factor(B)	0.12	0.06	0.04
				Factor(A X B)	0.21	0.10	0.07

*Figures in parenthesis are SQRT (X+1) transformed values

Table 3. Mean number of leafminers on different sticky traps at different heights

LM	0 ft	1 ft	2 ft	3 ft	4 ft	5 ft	Mean
Yellow	19.79	15.90	12.11	9.32	5.95	3.01	11.01 (3.47)
White	14.73	11.82	9.84	7.33	4.78	2.73	8.54 (3.09)
Blue	10.07	8.01	6.15	4.17	2.38	1.01	5.30 (2.51)
Mean	14.86	11.91	9.37	6.94	4.37	2.25	
	(3.98)	(3.59)	(3.22)	(2.82)	(2.32)	(1.80)	
				Factors	**C.D.**	**SE(d)**	**SE(m)**
				Factor(A)	0.07	0.03	0.02
				Factor(B)	0.09	0.05	0.03
				Factor(A X B)	0.16	0.08	0.06

*Figures in parenthesis are SQRT (X+1) transformed values

over the rest of the heights. The mean number of whiteflies was more upto 2 ft height above the crop canopy and was very less on the traps placed beyond 3 ft height above the crop canopy (table 2). In corroboration with the results, Bryne *et al*. (1986) reported that populations of whitefly were captured effectively on yellow sticky traps placed at ground level in cotton fields. Premalatha and Rajangam (2011) reported that maximum number of adults whiteflies were attracted towards yellow sticky trap in gerbera. The whitefly catches in the yellow trap was the highest, followed by red, green, blue, white, and black traps (Idris *et al*., 2012). Likewise, Lu *et al*. (2012) reported that yellow sticky traps can be used as an effective method for the control of whiteflies in the greenhouse. Elango *et al*. (2017) mentioned in their study that the yellow colour sticky trap significantly attracted more number of whiteflies in pomegranate. Field trials in brinjal confirmed that yellow sticky trap attracted more number of whiteflies as compared to the green, pink and light green, so yellow color traps may be used in methods of population monitoring of whiteflies (Khuhro *et al* 2020).

Leaf miner

Along with the sucking pests, large number of leaf miner adults were also get attracted and stuck to the sticky traps. The mean number of leaf miner adults trapped on sticky traps was high when compared to thrips and whiteflies irrespective of colour and height of trap placement. The mean number of leaf miner adults was high on yellow coloured sticky trap (11.01 sq.cm^{-1}) which was found significantly superior over the white (8.54 nos/sq.cm) and blue (5.30 sq.cm^{-1}) coloured sticky traps (table 3 & fig.1). The height of trap placement also exhibited significant influence on trapping efficiency, since, the mean number of leaf miners was highest at 0 ft i.e at the height of crop canopy (14.86 nos/sq.cm) which was significantly superior over the remaining heights. The mean trap catch was reduced gradually when the height increased from the crop canopy. The lowest number (2.25 nos/sq.cm) was recorded from the traps placed at 5 ft above the crop canopy (Table.3). Leaf miners, as well as other dipterans have shown higher attraction for yellow and green, with yellow being the most common colour when sticky cards are used for monitoring (Chandler, 1981). Hassan and Mohammed (2004) reported that higest number of leaf miners and thrips were attracted towards fluorescent yellow when compared to pink, green and orange coloured traps in greenhouse conditions. Adult pea leafminer were preferentially attracted to yellow opaque or translucent sticky cards when compared to red, blue, violet, green and white sticky traps in celery (Martin *et al*., 2005). Changizi (2011) reported that there was significant increase in trap catch of leaf miner adults when the yellow sticky trap number was increased compared to blue sticky traps in chickpea field.

Coccinellid beetles

The mean data revealed that the colour and height of placement of trap had a significant role on trapping of natural enemies such as coccinellid beetles. The mean number of ladybird beetles was high on yellow coloured sticky trap (4.30 trap^{-1}) and it was found statistically superior over white (2.37 trap^{-1}) and blue (2.34 trap^{-1}) coloured sticky traps (table. 4 & fig.1). The height of trap placement also had significant influence on trapping of coccinellids. The mean number of coccinellids was highest on the trap placed at the crop canopy level (6.92 trap^{-1}) which was found significantly superior over the other heights. The mean number of coccinellids was reduced gradually when the height of the traps increased from the crop canopy (Table.4). The results are in harmony with Dowell and Cherry (1981) who reported that yellow sticky traps caught significantly more parasitoids and coccinellids and yellow was the most attractive colour for many of coccinellid species in citrus. Similarly, Wallis and Shaw (2008) also reported that hymenopterans and coccinellids were attracted more towards yellow tarps in apple orchards. Hossain *et al*. (2020) reported that maximum catch of coccinellids was observed on yellow sticky traps while it was low on blue sticky traps in chilli crop. The results

Table 4. Mean number of coccinellids on different sticky traps at different heights

	0 ft	1 ft	2 ft	3 ft	4 ft	5 ft	Mean
Yellow	9.40	7.00	4.93	2.90	1.38	0.16	4.30 (2.30)
White	5.84	3.94	2.43	1.46	0.56	0.00	2.37 (1.84)
Blue	5.51	3.72	2.83	1.44	0.54	0.00	2.34 (1.84)
Mean	6.92	4.89	3.40	1.93	0.83	0.11	
	(2.81)	(2.43)	(2.10)	(1.71)	(1.35)	(1.05)	
				Factors	**C.D.**	**SE(d)**	**SE(m)**
				Factor(A)	0.06	0.03	0.02
				Factor(B)	0.08	0.04	0.03
				Factor(A X B)	0.14	0.07	0.05

*Figures in parenthesis are SQRT (X+1) transformed values

showed that yellow sticky traps placed upto 2ft height are detrimental to coccienllids since more number of coccinellids were caught on yellow traps compared to blue and white coloured traps. In corroboration, Pobozniak *et al*. (2020) reported that the yellow traps may be risky because they reduce population densities of the predaceous *A. intermedius* in pea fields, thus leading to an increase in pest numbers.

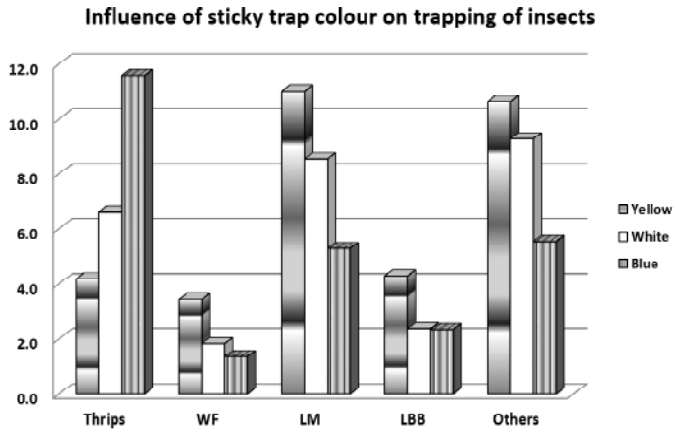
Other insects:

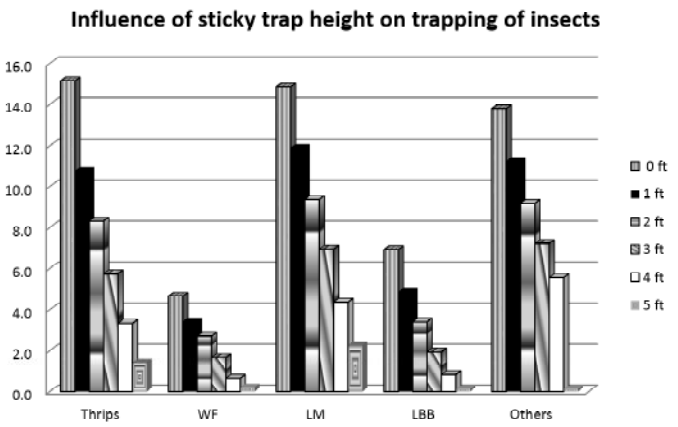
Besides trapping of sucking pests, leafminers and coccinellids, some big dipteran flies, small bugs and some hymenpteran insects such as small wasps and parasitoid wasps were also observed on sticky traps. Among the three colours, yellow and white coloured traps attracted more number of insects when compared to blue coloured sticky traps. The mean number of other insects was significantly less on blue coloured traps (5.55 trap^{-1}) when compared to yellow (10.61 trap^{-1}) and white (9.28 trap^{-1}) coloured sticky traps (table 5 & fig.1). The number of other insects was also high on the trap placed at the level of crop canopy (13.79 trap^{-1}) followed by 1ft (11.25 trap^{-1}) and 2ft (9.18 trap^{-1}) which differ significantly among themselves and superior over the remaining heights (table 5). The results are in concurrence

Table.5: Mean number of other insects on different sticky traps at different heights

Others	0 ft	1 ft	2 ft	3 ft	4 ft	5 ft	Mean
Yellow	16.61	13.40	11.32	8.50	7.99	5.85	10.61 (3.41)
White	15.27	12.85	9.90	8.54	5.61	3.53	9.28 (3.21)
Blue	9.49	7.51	6.32	4.60	3.07	2.33	5.55 (2.56)
Mean	13.79	11.25	9.18	7.21	5.56	3.90	
	(3.85)	(3.50)	(3.19)	(2.87)	(2.56)	(2.21)	
				Factors	**C.D.**	**SE(d)**	**SE(m)**
				Factor (A)	0.11	0.05	0.04
				Factor (B)	0.15	0.07	0.05
				Factor (A X B)	N/A	0.13	0.09

*Figures in parenthesis are SQRT (X+1) transformed values





with Broadsgaard (1993) who reported that generally yellow traps are particularly used for catching coleopteran, hemipteran, hymenopteran and thysanopteran insects. Riley and Schuster (1994) reported that yellow traps are particularly used for catching coleopteran, hemipteran, hymenopteran and thysanopteran insects. The maximum catch of coccinellid predators, honey bee, hoverfly and parasitic wasps was recorded with yellow sticky trap and blue coloured sticky trap also attracted comparatively less number of beneficial insects in onion ecosystem (Devi and Roy, 2017).

Irrespective of sticky trap colour, the trapping of sucking pests and leaf miner adults as well as coccinellids and other insects was more when the traps were placed near to the crop canopy i.e. upto two feet above the crop canopy. As the height of trap increased, the number of insects trapped on sticky traps were reduced irrespective of trap colour and insect type (fig. 2). The results in the present study are in agreement with many of the earlier reports in different crops. Gerling and Horowitz (1984) suggested that more whiteflies on traps at the lowest levels in cotton which might be due to

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better feeding and oviposition sites in the lower part of the cotton canopy. Atakan and Ramazan, (2004) also reported that height of traps negatively affected the trapping of the sucking insect pests, as trap heights increased, the number of insects that were captured on the traps declined in cotton. Martin *et al*., (2005) reported that highest captures of thrips occurred at below 20 cm height when compared to 30,50, 70 and 90 cm from the celery crop canopy. Mao et al. (2018) reported that white or deep sky blue cards placed low heights i.e. near to the ground attracted more number of thrips than the traps placed at middle and higher heights in cowpea.

CONCLUSION

It was concluded that use of blue and yellow sticky traps placed at a height of 2ft above the crop canopy could be used to trap maximum number of thrips, white fly and leaf minor damaging green gram. These can be integrated with other components of IPM program for detection and monitoring of insect population and also to make decisions for initiation of pesticide application.

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Evaluation of coloured sticky traps in attracting rugose spiralling whitefly in coconut

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ABSTRACT

In an investigation carried out at Regional Coconut Research Station, Bhatye, Ratnagiri (M.S.) during 2018-19 and 2019-20 to validate the effect of coloured traps (red, yellow, green, black and white) in attracting rugose spiralling whitefly (RSW), the yellow coloured sticky traps, made with sheets (100 x 50 cm) and smeared with white grease recorded the maximum catch of rugose spiraling whiteflies (18.3) and was significantly superior over black and white colour sticky traps but was on par with green (13.7 nos.) and red coloured sticky traps (7.6 nos.). The black and white coloured sticky traps trapped the minimum of 3.8 and 3.3 rugose spiraling whiteflies at five different points of dimensions of 5 cm^2, respectively.

Keywards: *Coconut, coloured traps, rugose spiraling whitefly, yellow colour*

The Coconut palm (*Cocos nucifera* Linn.) has great socio–economic significance as it is the source of livelihood for more than 20 million people globally, especially small and marginal farmers. Rugose spiralling whitefly, *Aleurodicus rugioperculatus* Martin (Hemiptera : Aleyrodidae) is an invasive pest on coconut along the roads in coastal areas. It attacks a wide range of host plants including palms, woody ornamentals, and fruits. Coconut and banana are among the most preferred host plants. It has been recently been reported from Tamil Nadu (Sundararaj and Selvaraj, 2016) and is also distributed in Karnataka, Kerala, Andhra Pradesh, Goa, Gujarat and Maharashtra. It mainly infests coconut palms and other broad-leaved hosts in its native range. It is distributed in Belize, Guatemala, Mexico(Martin, 2008) and has subsequently spread to 22 other countries in Central and South America, including Florida, USA. This notorious pest on coconut was first reported in Maharashtra during August, 2017. Infestation was observed on coconut seedling that later spread to other crop *viz.*, banana, custard apple, mango, cashew nut, almond, areca palm and bush pepper in Maharashtra. Severe outbreak of RSW was recorded in coconut palms, mango and guava at Changanassery, Kottayam districts of Kerala (Shanas *et al*., 2016). The RSW is highly polyphagous with 118 hosts belonging to 43 plant families including economically important crops in the United States (Francis *et al*., 2016). Its infestations has been observed on guava, citrus, mango, sapota, bhendi, custard apple, jatropha and hibiscus (Selvaraj *et al*., 2016). Among all the host plants, eight were infested by *A. rugioperculatus* in which all the life stages of whitefly were noticed. In other 12 host plants, only the eggs stages were documented in Tamil Nadu (Elango *et al*., 2019). It has become an escalating problem for coconut farmers. Feeding by this pest not only causes stress to its host plant, but due the excessive production of wax and honeydew it creates an enormous nuisance in infested areas. The presence of honeydew results in the growth of fungi called sooty mold, which turns everything in the vicinity covered with honeydew with black mold. Severe infestation has recently been observed in Dapoli and Bhatye village of Ratnagiri district, the major hub of nursery activities supplying planting material throughout the State. Keeping the above facts as also as adverse effect of pesticides on environment and human health in view, the present investigation on effectiveness of different coloured traps in managing spiralling whitefly in coconut was carried out.

MATERIALS AND METHODS

The investigation was carried out at Regional Coconut Research Station, Bhatye, Ratnagiri (M.S.) during 2018-19 and 2019-20. It was conducted in Completely Randomized Block (CRD) design with three replications and five treatments *viz.*, T1-Red colour, T2-Yellow colour sticky trap, T3-Green colour sticky trap, T4-Black colour sticky trap and T5-White colour sticky trap. These coloured plastic sheets having dimension 100 x 50 cm were smeared with white grease over and were hanged in between two palms in RSW infested garden at 1 m from ground level. Observations were recorded by counting numbers of rugose spiralling whitefly populations at five different points of dimensions 5 cm^2 in early morning @ five days interval in a month. The data were subjected to statistical analysis.

RESULTS AND DISCUSSION

The mean data presented in Table 1 revealed maximum catch of rugose spiralling whiteflies (76.8) on yellow coloured sticky traps and was significantly superior over rest of the treatments. Green coloured sticky traps recording 44.7 number of RSW was the second best and significantly superior over black (36.1 nos.) and red colour sticky trap (17.8 nos.). The black coloured traps were found at par with red coloured sticky traps and significantly superior over white (10.7 nos.) during 2018-19. Minimum number of rugose spiralling whitefly was trapped on white coloured sticky traps.

Table 1. Evaluation of different coloured sticky traps for attraction of RSW in Ratnagiri centre during 2018-19

Treatments	No. of RSW trapped at five different points of dimensions 5 cm²				
	December, 2018	January, 2019	February, 2019	March, 2019	Mean
T1-Red colour sticky trap	5.7 (2.5) *	11.3 (3.4)	31.4 (5.6)	22.7 (4.8)	17.8 (4.1)
T2-Yellow colour sticky trap	36.7 (6.1)	77.8 (8.7)	83.8 (9.1)	109.0 (10.4)	76.8 (8.6)
T3-Green colour sticky trap	19.0 (4.4)	61.3 (7.8)	36.6 (6.1)	61.6 (7.9)	44.7 (6.6)
T4-Black colour sticky trap	11.3 (3.4)	43.0 (6.6)	29.5 (5.4)	60.7 (7.8)	36.1 (5.8)
T5-White colour sticky trap	7.3 (2.8)	12.3 (3.5)	11.2 (3.4)	12.1 (3.6)	10.7 (3.3)
SE ±	0.17	0.58	0.38	0.63	0.44
CD at 5%	0.55	1.79	1.17	1.94	1.36

*Figures in parenthesis are square root transformed values

The data shown in Table 2 revealed maximum mean number of rugose spiralling whiteflies attracted on yellow coloured sticky traps (11.7), which was significantly superior over T1 (red colour sticky trap) T4 (black coloured sticky traps) and T5 (white coloured sticky traps). Green traps were the next effective treatment (8.5). The red colour sticky traps with 5.17 catch was significantly superior over T4-black coloured sticky traps (2.5 nos.) and T5- white coloured sticky traps (2.1 nos.). The white coloured sticky traps recorded minimum rugose spiraling whiteflies populations at five different points of dimensions 5 cm², respectively during 2019-20.

The overall mean data depicted in Table 3 revealed maximum rugose spiraling whiteflies (14.2 nos.) on yellow coloured sticky traps which was significantly superior over rest of the treatment. The green coloured traps were the next best (26.6) and significantly superior over T1-red coloured sticky traps (11.4 nos.), T5-white coloured sticky traps (6.4 nos.) and T4-black coloured sticky traps (1.9 nos.). Minimum number of rugose spiraling whitefly populations was observed in T5 white colour sticky traps at five different points of dimensions 5 cm². Overall mean of two years data revealed superiority of yellow coloured sticky traps. The effectiveness of is treatment was also reported by Wang *et al*., 2015. In his study, yellow colour trap was reported most preferred by tea sping white fly, *Aleurocanthus spiniferus.* Idris *et al*. (2012) found that the yellow was the most attractive colour to alate

Table 3. Overall mean populations of rugose spiraling whiteflies at five different points of dimensions 5 cm²during 2018-2020 at Ratnagiri centre

Treatments	Mean		Cumulative average
	2018-19	2019-20	
T1-Red colour sticky trap	17.8 (4.1)*	5.1 (2.1)	11.4 (3.1)
T2-Yellow colour sticky trap	76.8 (8.6)	11.7 (3.1)	44.2 (6.8)
T3-Green colour sticky trap	44.7 (6.6)	8.5 (2.6)	26.6 (4.6)
T4-Black colour sticky trap	36.1 (5.8)	2.5 (1.6)	1.9 (3.7)
T5-White colour sticky trap	10.7 (3.3)	2.1 (1.4)	6.4 (2.3)
SE ±	0.44	0.17	0.30
CD at 5%	1.36	0.54	0.95

(*Figures in parenthesis are square root transformed values)

Table 2. Determination of different coloured sticky traps for attraction of RSW during 2019-20

Treatments	No. of RSW trapped at five different points of dimensions 5 cm²								
	Oct., 19	Nov., 19	Dec., 19	Jan. 20	Feb., 20	March, 20	April, 20	May, 20	Mean
T1-Red colour sticky trap	10.3 (3.2)*	13.0 (3.6)	4.5 (2.2)	2.5 (1.7)	4.9 (2.3)	4.0 (2.1)	1.5 (1.4)	0.5 (1.0)	5.1 (2.1)
T2-Yellow colour sticky trap	27.3 (5.2)	28.0 (5.3)	12.8 (3.6)	5.2 (2.3)	8.8 (3.0)	7.3 (2.8)	3.0 (1.8)	1.2 (1.3)	11.7 (3.1)
T3-Green colour sticky trap	20.6 (4.5)	20.6 (4.4)	10.3 (3.2)	3.2 (1.9)	6.4 (2.6)	4.4 2.2)	2.0 (1.6)	0.7 (1.1)	8.5 (2.6)
T4-Black colour sticky trap	4.3 (2.1)	5.7 (2.5)	3.5 (2.0)	1.7 (1.5)	2.5 (1.7)	1.2 (1.3)	0.7 (1.1)	0.3 (0.9)	2.5 (1.6)
T5-White colour sticky trap	7.3 (2.7)	4.5 (2.2)	0.8 (1.1)	0.7 (1.1)	1.5 (1.4)	1.0 (1.2)	0.7 (1.1)	0.3 (0.9)	2.1 (1.4)
SE ±	0.27	0.35	0.20	0.28	0.07	0.12	0.09	0.05	0.17
CD at 5%	0.83	1.00	0.62	0.84	0.22	0.39	0.29	0.17	0.54

(*Figures in parenthesis are square root transformed values)

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whitefly, regardless of the trap design as it had the highest number of alates caught compared to the other colours. Elangol *et al*., 2017 studied different coloured sticky traps for monitoring the population of sucking pests such as thrips, whiteflies, leaf miner adults and aphids and reported that yellow was most attractive to thrips (177.4 thrips per trap^{-1} per week^{-1}) and white fly (22.1 whiteflies per trap^{-1} per week^{-1}) followed by pale yellow (122.1 thrips per^{-1} trap per^{-1} week^{-1}) & (13.8 whiteflies per trap^{-1} per week^{-1}) and green (110.9 thrips per^{-1} trap per^{-1} week^{-1}) & (13.1 whiteflies per trap^{-1} per week^{-1}). Generated results of field trials confirmed that yellow sticky trap attracted more number of whiteflies as compared to the green, pink and light green, so yellow color traps may be used in monitoring insect population (Khuhro *et al*., 2020). These results are in agreement with Premalatha and Rajangam (2011) who reported maximum number of whitefly, *Trialeurodes vaporariorum* (Westwood) attracted towards yellow sticky trap in gerbera. Likewise, Lu *et al*., (2012) advocated use of yellow sticky traps as an effective method for the control of whiteflies, *Bemisia tabaci* in the greenhouse. Yellow sticky traps are a commonly used method for population monitoring of many pests. In recent decades, studies of these traps mainly focused on how to use them to monitor populations of pest species such as whiteflies, leafminer and aphids (Shen and Ren, 2003; Zhou *et al*., 2003; Qiu *et al*., 2006; Gu *et al*., 2008).

CONCLUSION

The yellow coloured sticky trap can effectively be used as an important component of IPM in the management of rugose spiraling whiteflies in coconut.

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Identification of fungal seed-borne pathogens in farm saved seeds of sorghum (*Sorghum bicolor*) in Hamelmalo sub-zoba of Zoba Anseba, Eritrea

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ABSTRACT

The study carried out to assess seed mycoflora on sorghum collected from five village's of Sub-Zoba Hamelmalo, viz; Wazentit, Hamelmalo, Hitsats, Basheri and Kurba-bered in Eritrea revealed preponderance of eight fungal genera in the region. All the eight fungal pathogens viz., *Alternaria, Aspergillus, Fusarium, Penicillium, Helminthosporium, Mucor, Rhizopus* and *Curvularia* occurred in unwashed seeds whereas, only six of the eight (*Alternaria, Aspergillus, Fusarium, Penicillium, Helminthosporium* and *Rhizopus*) occurred in the tap water washed and cattle urine treated samples. Over all, the pathogen frequency was higher in unwashed (35.76%) and lower (14.66%) in cattle urine treated seeds.

Key words: seed borne fungi, sorghum, farmer saved seeds

Seed is the most important single input for arable cultivation determining the potential of production and productivity of all other inputs (Friis-Hansen, 1995). About 90 per cent of the world food crops including sorghum (*Sorghum bicolor* (L) Moench) are propagated by seed (Maude, 1996). However, these act as a passive carriers of pathogens under suitable environmental conditions. Fungi, bacteria, viruses and nematodes can be carried with, on or in the seeds resulting in tremendous yield losses (Neergaard, 1979).

Sorghum (Mishella) and pearl millet in Eritrea are the most important and widely grown cereal crops, especially in lowlands. Sorghum ranks first in the contribution towards national economy and diet and on an average 45 per cent of the bulk total food production for the nation comes from it. Sorghum is affected by a range of fungal seed borne diseases including ergot (*Claviceps africana*), seed rot (*Fusariummoniliforme*), zonate leaf spot (*Gloeocercospora sorghi*), downy mildew (*Sclerospora sorghi*), loose smut (*Sphacelotheca cruenta*), covered smut (*Sphacelotheca sorghi*), leaf spots (*Phomasorghina*), *Bipolaris bicolor,* anthracnose (*Colletotrichum graminicola*) and grey leaf spot, (*Cercospora* sp.) (Almekinders and Louwaars, 1999; Kaula and Chisi, 2002 and Neergaard, 1979). Seed-borne mycoflora of sorghum reported from different parts of the world include *Alternaria alternata, Aspergillus flavus, A. fumigatus, A. niger, Cladosporium* spp., *Fusarium moniliforme, F. oxysporum, F. pallidoroseum, Drechslera tetramera, Nigrospora* spp., *Phoma* spp., and *Rhizopus* spp.

Most of small holder farmers that venture into sorghum production use farm saved seed, which is of poor quality. This is because the certified seed is expensive and farmers can hardly afford purchasing it. Sometimes, the economic gains from using a higher seed quality do not justify the purchase of seed (Almekinders and Louwaars, 1999). In most cases, it is likely that seed borne pathogens accumulate in farm saved seeds with time. Thus, there is a need for studying the prevalence of fungi in sorghum seeds collected from different sorghum growing areas of the sub-zoba Hamelmalo and development of suitable control measures for pathogenic fungi prevalent in sorghum seeds.

MATERIALS AND METHODS

The experiment was conducted at the research laboratory of the Department of Plant Protection, Hamelmalo Agricultural College, sub-zone of Hamelmalo, Eritrea which has an average temperature of 36°C annual rainfall of 400mm and is situated at an altitude of 1328 meter above sea level. The sorghum seed were collected from farmers during August 15 up to September 16, 2019. Five samples of sorghum seeds collected from five villages viz; Basheri, Hamelmalo, Hitsat, Kurba-bered and Wazentit; of Sub-zoba Hamelmalo were used for the isolation and detection of seed borne fungi. Farmers were selected randomly for sampling. From each farmer, samples of 250 gram seeds were taken and checked in the laboratory for seed germination, identification of seed borne fungal pathogen and percent frequency. The standard blotter method as

described by the International Seed Testing Association (ISTA 1976), was used for the isolation of the seed-borne fungi associated with the sorghum seed samples. The seed samples in their various forms (washed, unwashed, and treated with cattle urine @ 5 ml kg^{-1} seed) were inoculated on three moistened 9.0 cm filter papers in 9.0 cm Oswald Petri-dishes. Twelve seeds were arranged at the periphery of the plate, nine at the middle, and four at the centre of sorghum. A total of five seed samples, with three replications, were used, and kept in dark place to check germination percentage and fungal identification and pathogen frequency.

Sampling for germination was done at 72 hrs (3 days) after incubation, while identification of fungi was done at 7 days. The petri dishes were brought to the examination area in the laboratory, where each seed was examined under a microscope. Growth habits of the various fungi growing in the petri plates were observed carefully. Slide preparations of the various fruiting structures of the fungi were made and observed under a microscope for identification. The various types of fungi were identified using identification keys and cross-checked for each seed plated to identify the type of fungus growing on each seed. After 7 days of incubation, the fungal species growing on seeds surfaces were identified and their percentage frequency (PF) was calculated by applying the following formula:

PF= [(No. of seeds on which fungus appear/total no. of seeds) X100]

Analysis of variance method was applied for drawing conclusion from the data. The calculated value was compared with the tabulated value at 5 per cent level of probability (Fisher and Yates, 1968) for the appropriate degrees of freedom (d.f.).

RESULTS AND DISCUSSION

Seed germination

The results produced in table 1 indicate that there was no significant difference among the treatments. However, comparatively higher germination percentage (36%) was recorded in Hitsats sample followed by Kurba-bered samples (32%). Lowest germination percentage (24%) was obtained in

S-1(Basheri) followed by S-2 (Wazentat) and Hamelmalo samples. The germination percentage was tasted by washing the seed samples using tap water on the same technique taken under the blotter taste as those of unwashed. However, the result obtained in this trial was significantly different as compared to the unwashed. The highest germination percentage (45.33%) was recorded in S-5 (kurba-bered) followed by S-3 (Hamelmalo), S-4 (Hitsat) and S-2 (Wazentat) having 44.0, 40.0, and 36.0 percent, respectively. No more readable difference was recorded. The lowest germination (21.333%) was obtained in S-1(Basheri).

Table 1: Seed germination percentage of farmers saved sorghum

Samples	Seeds		
	Un-washed	Washed with tap water	Treated with urine
S_1 (Basheri)	24.000	21.333	32.000
S_2(Wazentit)	28.000	36.000	40.000
S_3(Hamelmalo)	28.000	44.000	40.000
S_4(Hitsat)	36.000	40.000	49.333
S_5(Kurba-Bered)	32.000	45.333	46.666
Cv	25.995	12.824	NS
LSD	NS	18.247	20.205
Average	29.60	37.32	41.54

Table 2: Pathogen frequency of farmers saved sorghum seeds

Samples	Seeds		
	Un-washed	Washed with tap water	Treated with urine
S_1 (Basheri)	38.666	24.000	16.000
S_2(Wazentit)	29.333	18.666	8.000
S_3(Hamelmalo)	29.333	16.000	18.666
S_4(Hitsat)	30.666	20.000	17.333
S_5(Kurba-Bered)	44.000	24.000	13.333
LSD	NS	NS	NS
CV	21.381	17.604	38.885
Average	35.76	20.52	14.66

The germination percentage was also tested by washing the seed samples using urine @ 5ml kg^{-1}. The result showed no valuable difference as the table indicate highest germination percentage (49.333%) in S-4 (Hitsat) followed by S-5 (Kurba –bered). Having(46.666%). S -2(wazentit) and S -3(Hamelmalo) have the same germination percentage (40%).The lowest germination percentage (32.00%) was recorded in S-1(Basheri).

Pathogen frequency

The result (Table 2) showed that the pathogen frequency of sorghum seeds in the five treatments have no significant difference. It was found to be the lowest in S-2 and S-3 recorded as (29.33%) each followed by S-4which have (30.666%). The obtained result of pathogen frequency in S-5 was found to be the highest (44.00) followed by S-1 (38.66). The percentage frequency of occurrence of the pathogen in sorghum seeds washed with water (Table 2) showed almost no difference. It was higher in S-1(basheri) and S-5(kurba-bered) each having (24%) followed byS-4

(Hitsat) which have (20%). The lowest pathogen frequency was obtained to be(18.666%) in S -2(wazentet) following S-3 (Hamelmalo) which have (16.00).

The percentage frequency of the fungal pathogen in sorghum seeds washed with urine showed no significant difference. It was higher-in-S-3 (18.666%) followed by S-4(hitsat), S-1(Basheri) and S-5 (kurba-bered) having 17.33, 16.00 and 13.33 per cent, respectively. The lowest percentage (8.0%) was obtained in S-2 (Kurba-bered). The result (Table 1) and (Table 2) shows the average germination percentage and average percent pathogen frequency of farm saved sorghum seeds collected from five villages of Hamelmalo sub-zone, respectively. The result reveals that the germination percentage was higher in S-3(41.54) and the pathogen frequency was highest in S-1 (unwashed), where as the reverse was found in both the cases. The result obtained in the two treatment means is medium in both the aspects.

The results showed that seeds washed with cattle urine was more effective as it increased germination percentage by reducing the pathogen frequency of farm saved sorghum seeds grown under laboratory conditions. Although the effectiveness is not as much as with urine treatment, the result obtained in S-2(washed with water) also showed readable results.

Sorghum seed mycoflora

The table 4 and 5 indicated that a total of eight fungal genera viz; *Alternaria, Aspergillus, Fusarium, Penicillium, Helminthosporium, Mucor, Rhizopus* and *Curvularia* were found in sorghum seeds of Hamelmalo sub zone. The three treatments showed no difference in the kind of fungal pathogens. However, population of the pathogens was high in the unwashed samples. The lowest fungal population was recorded in the cattle urine treated seeds. The results showed that of all the samples tested were found associated with at least one known pathogen i.e. *Aspergillus*. These results are in agreement with those of Kamal and Mughal(1968) and Khan et al.(1974), who reported the presence of *Alternaria, Helminthosporium, Fusarium, Rhizopus, Aspergillus* and *Penicillium* species in sorghum seeds. The result also collaborate with those of Khan and Bhuta (1994), who reported the occurrence of

Table 3. Identification of seed-borne fungal pathogens detected in various unwashed seed samples of sorghum collected from sub-Zoba- Hamelmalo

Samples	*Alternaria*	Aspergillus	Penicillium	Rhizopus	Fusarium	Curvularia	Helminthosporium	Mucor
S_1(Basheri)	+	+	-	-	+	+	+	-
S_2(Wazentit)	-	+	-	+	-	-	-	-
S_3(Hamelmalo)	-	+	-	-	+	-	-	-
S_4 (Hitsat)	-	+	+	+	-	-	-	-
S_5 (Kurba- Bered)	-	+	-	-	-	-	-	-

Table 4. Identification of seed-borne fungal pathogens detected in various seed samples washed with tap water

Samples	Alternaria	Aspergillus	Penicillium	Rhizopus	Fusarium	Curvularia	Helminthosporium	Mucor
S_1 (Basheri)	+	+	-	-	+	+	+	-
S_2 (Wazentit)	-	+	-	+	-	-	-	-
S_3 (Hamelmalo)	-	+	-	-	+	-	-	-
S_4 (Hitsat)	-	+	+	+	-	-	-	-
S_5 (Kurba-Bered)	-	+	-	-	-	-	-	-

Table 5. Identification of seed-borne fungal pathogens detected in various seed samples treated with cattle urine @ 5ml/Kg

Samples	Alternaria	Aspergillus	Penicillium	Rhizopus	Fusarium	Curvularia	Helminthosporium	Mucor
S_1 (Basheri)	+	+	-	-	-	+	+	-
S_2 (Wazentit)	-	+	+	+	-	-	-	-
S_3 (Hamelmalo)	-	+	-	-	-	-	-	-
S_4 (Hitsat)	-	+	-	-	+	-	-	-
S_5 (Kurba-Bered)	-	+	-	+	-	-	-	-

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Helminthosporium and *Fusarium moniliforme* as major pathogens of sorghum seed. Other reports by Singh (1983) also showed that *Aspergillus, Helminthosporium* and *Fusarium* spp. were common in associates of stored sorghum seeds. The common occurrence of other pathogens like *Alternaria, Fusarium, Aspergillus,* and *Penicilium* has been widely reported (Martin et al.,1984).The implications of this wide spread sees infestation is highlighted in the report of Dharmvir et al (1968),who determined that sorghum seeds colonized during storage were responsible for reducing plant population by 42 per cent in the field. The consequence of such infestation is not only limited to yield losses but also accounts for the buildup of mycotoxin in infected grains. The finding of this study are therefore important as they highlight the need for effective measures aimed at reducing seed –borne infection of sorghum seeds in Eritrea.

CONCLUSION

The study concludes association of eight seed borne fungal genra with the un-washed/untreated sorghum seeds used by the farmers for sowing in five villages (Basheri, Wazintet, Hamelmalo, Histate and Kurba- Bered) of sub-zone of Hamelmalo, Zoba Anseba, Eritrea. *Alternaria, Aspergillus, Fussarium, Rhizopus, Curvularia, Penicillium, Helminthosporium* and *Mucor* sp. were the main fungi occurring frequently in sorghum seeds. Thus, there is need to increase public awareness on aspects related to seed health and to develop suitable management practices for improving the quality of seeds. There is also need to campaign for public awareness for seed health and to develop eco-friendly seed borne pest management tactics for improving the seed quality.

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Integrated management of *Alternaria* blight disease of Pigeon pea

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ABSTRACT

Blight disease of pigeon pea caused by *Alternaria alternata* is occurring in an alarming proportion in almost all pigeonpea growing areas of the world. Its management through integrated use of bio-agent and fungicides were evaluated under field conditions. All the fungicides tested *in vitro* were found effective in inhibiting the pathogen over control, except the carbendazim and thiophanate methyl, which showed the per cent inhibition of 44.79 and 29.15 per cent, respectively. Among the 12 treatments tested under field condition, the application of two sprays of difenconazole (0.1%) interspersed with *Pseudomonas fluorescens* (0.5%) recorded minimum per cent disease index of 25.75 with maximum yield (13.94 q ha^{-1}), which was statistically on par with the application of two sprays of propiconazole (0.1%) interspersed with *P. fluorescens* (0.5%).

Key words: *Alternaria alternata*, fungicides, pigeon pea, *Pseudomonas fluorescens*

Pigeon pea [*Cajanus cajan* (L.) Millsp.] is an important pulse crop commonly known as redgram or arhar and is grown in the tropics and sub-tropics of the world. It provides high quality protein, aminoacids and acts as fuel wood and animal feed. In addition, this crop is resilient, widely adopted and drought tolerant and restores soil fertility by fixing atmospheric nitrogen and solubilizes fixed phosphorus (Ae *et al*. 1990). The global pigeon pea production scenario accounts an area 6.99 million ha with 5.96 million tonnes of production with the productivity of 852 kg ha^{-1} (Anon., 2018). In India, the total area coverage, production and productivity accounts 4.78 million ha, 3.59 million tonnes and 751 kg ha^{-1}, respectively. The state-wise trend showed that Maharashtra ranks first both in area and production (29.19 % and 29.68 %) followed by Karnataka (19.23 % and 15.96 %). The maximum productivity was recorded by Bihar (1739 kg ha^{-1}) followed by Haryana (1111 kg ha^{-1}) and Gujarat (1105 kg ha^{-1}). Whereas, the minimum production was observed in Andhra Pradesh (521 kg ha^{-1}) followed by Chhattisgarh (623 kg ha^{-1}) and Karnataka (648 kg ha^{-1}) (Anon., 2019).

Several diseases have been reported to infect pigeon pea crop at different stages during its growth period in different parts of the country. The most important diseases include wilt, dry root rot, *Phytophthora* blight, sterility mosaic disease and leaf spots and blights. Among these, leaf blight incited by *Alternaria alternata* is occurring in an alarming proportion and causing menace to the tur production (Nene *et al.*, 1989). Generally, the chemical management is the most common practice and practical method to manage blight of pigeon pea. However, fungicide tolerance by the pathogen often arises quickly, if a single compound is relied upon too heavily and also residual effect on the consumer. Keeping this in view, the management of leaf blight of pigeon pea has become an issue in present condition. Therefore, a laboratory experiment to test the efficacy of fungicides under *in vitro* and field experiment to test the efficacy of fungicides interspersed with the bacterial bioagent *P. fluorescens* for the management of blight of pigeon pea was carried out.

MATERIALS AND METHODS

In vitro evaluation of fungicides was carried out at College of Agriculture, University of Agricultural Sciences, Raichur, Karnataka. The efficacy of fungicides *viz*., Chlorothalonil, iprodione, carbendazim, hexaconazole, thiophanate methyl, propiconazole, difenconazole, mancozeb and carbendazim+mancozeb (Combi fungicide) were evaluated at 0.1, 0.15 and 0.2 per cent concentration by poison food technique (Shravelle, 1961). Required quantity of individual fungicide was added separately into molten and cooled potato dextrose agar to get the desired concentration. Later, 20 ml of the poisoned medium was poured into sterile Petri plates. Mycelial disc of 5 mm size from actively growing culture of the fungus was cut using a sterile cork borer and one disc was placed at the centre of each agar plate. Control was maintained without adding any fungicide to the medium. Each treatment was replicated thrice and incubated at room temperature (28±2 °C) for nine days and the colony diameter was measured. The efficacy of fungicides was expressed as per cent inhibition of mycelial

growth over control. The per cent inhibition was calculated by using the formula suggested by Vincent (1947).

$$I = \frac{C - T}{C} \times 100 \text{ where,}$$

I=Per cent inhibition

C= Colony diameter of fungus in control

T= Colony diameter of fungus in treatment

Field trials were carried out during 2017 and 2018 with pigeon pea variety TS-3R (susceptible to leaf spot) for the management of this disease. The experiment comprised of 12 treatments the combinations of systemic fungicides, contact fungicides, combi-fungicides which were interspersed with *P. fluorescens* with three replications each in a randomised block design with each plot measuring 4m X 4m.

The first spray was initiated immediately after the onset of disease and repeated at fortnightly interval. All agronomical operations recommended for pigeon pea cultivation were followed from sowing to till harvest. Observations were made on the severity of *Alternaria* blight of pigeon pea on leaves at weekly intervals by using disease index standard scale 0-5 (0= no disease, 0.1= up to 5 per cent of leaf area with disease symptom, 2= 5.1-10 per cent disease; 3= 10.1-25 per cent; 4= 25.1-50 per cent disease and 5= >50 per cent leaf area with disease symptom) (Balai and Singh *et al.*, 2013) and PDI was worked out as per the standard protocol (Wheeler, 1969). The experimental data was statistically analysed by Anova after the transformation of values (Snedecor and Cochran, 1957).

Table 1. Efficacy of fungicides in inhibiting the mycelial growth of *A. alternata*

Sl. No.	Chemicals	Per cent inhibition			Mean
		0.1	0.15	0.2	
1	Chlorothalonil	100 (90.00)	100 (90.00)	100 (90.00)	100 (90.00)
2	Iprodione	100 (90.00)	100 (90.00)	100 (90.00)	100 (90.00)
3	Carbendazim	15.66 (23.31)	27.71 (31.76)	44.58 (41.88)	44.79 (42.01)
4	Hexaconazole	100 (90.00)	100 (90.00)	100 (90.00)	100 (90.00)
5	SAAF	100 (90.00)	100 (90.00)	100 (90.00)	100 (90.00)
6	Thiophanate methyl	30.72 (33.63)	37.35 (37.66)	46.99 (43.27)	27.77 (32.64)
7	Propiconazole	100 (90.00)	100 (90.00)	100 (90.00)	100 (90.00)
8	Mancozeb	100 (90.00)	100 (90.00)	100 (90.00)	100 (90.00)
9	Difenconazole	100 (90.00)	100 (90.00)	100 (90.00)	100 (90.00)
	Mean	82.93 (77.69)	85.01 (78.94)	87.95 (80.92)	
		Fungicides	**Concentrations**	**Interaction (FXC)**	
	S. Em. ±	0.27	0.15	0.47	
	C.D. at 1 %	1.05	0.57	1.81	

RESULTS AND DISCUSSION

Results on the efficacy of different fungicides on mycelia growth of *A. alternata* are presented in table 1 (Plate 1). It was observed that all the tested fungicides showed their efficacy to inhibit the growth of the pathogen over control except carbendazim and thiophanate methyl. Among the tested fungicides, chlorothalonil, iprodione, hexaconazole, mancozeb, propiconazole, carbendazim+mancozeb and difenconazole were effective with an average of 100 per cent inhibition of mycelium and were significantly superior over carbendazim (44.79 %) and thiophanate methyl (29.15%), respectively. Among the three concentrations of fungicides tested, the maximum mycelial inhibition was recorded at 0.2 per cent, which was significantly superior over other concentrations tested with 87.95 per cent inhibition. Interaction effects of fungicides and concentrations on per cent inhibiton were found to be significant with highest per cent inhibition recorded by chlorothalonil, iprodione, hexaconazole, carbendazim+mancozeb, propiconazole, mancozeb and difenconazole of 100 per cent at all the concentrations tested. Whereas, the least per cent inhibition was recorded by carbendazim at 0.1 per cent concentration (15.66 %).

The pooled mean data of the field experiments recorded during two seasons (Table 2) clearly revealed that all the treatments were significantly superior over control in managing *Alternaria* leaf blight. However minimum PDI (25.75 %) was recorded with two sprays of difenconzole (0.1%) interspersed with *P. fluorescens* (0.5%) at fortnightly interval with the maximum grain yield of 13.49 q/ha. This treatment was on par with the application of two sprays of propiconazole interspersed with *P. fluorescens* and found second best in the order of efficacy (25.94 % PDI &13.34 q ha^{-1}) followed by application of two sprays of hexaconazole at 0.1 per cent interspersed with *P. fluorescens* (0.5 %) with the PDI of 26.12 per cent and recorded the grain yield of 13.17 q ha^{-1}. Among the different fungicide treatments, the least per cent inhibition (38.73 %) and grain yield (9.69 q ha^{-1}) was recorded in the treatment of two sprays of carbendazim (0.1%). The untreated control plot recorded significantly highest PDI of 43.67 with the lowest yield of 9.46 q ha^{-1}.

The cost benefit analysis revealed that the application of two sprays of hexaconaxole interspersed with *P. fluorescens* has recorded the highest benefit to the cost incurred accounting to 2.58 (Plate 2). This clearly revealed that the application of hexaconazole played an important role in reducing the disease severity with an enhanced yield and returns.

Table 2. Effect of various treatments on per cent disease index of *Alternaria* leaf blight and grain yield of pigeon pea

Tr. No.	Treatment details	Per cent disease index			Grain yield (t ha^{-1})			BC ratio
		2017	2018	Pooled mean	2017	2018	Pooled mean	
T_1	Spray of chlorothalonil 75WP (0.2%) - *P. fluorescens* (0.5%) - chlorothalonil 75WP (0.2%)	30.27 (33.38)	28.13 (32.03)	29.20 (32.71)	11.13	10.81	10.97	2.09
T_2	Spray of Iprodione 50 WP (0.2%) - *P. fluorescens* (0.5%) -iprodione 50 WP (0.2%)	28.20 (32.07)	26.73 (31.12)	27.47 (31.60)	11.17	11.85	11.51	2.11
T_3	Spray of carbendazim 50 WP (0.1%) - *P. fluorescens* (0.5%) - carbendazim 50 WP (0.1%)	33.33 (35.26)	30.70 (33.65)	32.02 (34.46)	9.93	10.10	10.02	1.95
T_4	Spray of hexaconazole 5% EC (0.1%) - *P. fluorescens* (0.5%) - hexaconazole 5% EC (0.1%)	26.93 (31.26)	25.30 (30.19)	26.12 (30.73)	12.40	13.94	13.17	2.58
T_5	Spray of iprodione 50 WP (0.2%) - *P. fluorescens* (0.5%) - carbendazim 50 WP (0.1%)	30.53 (33.54)	28.77 (32.43)	29.65 (32.99)	11.13	10.57	10.85	2.05
T_6	Spray of SAAF (carbendazim 12% + mancozeb 63%) (0.2%) - *P. fluorescens* (0.5%) - SAAF (carbendazim 12% + mancozeb 63%) (0.2%)	29.07 (32.61)	27.83 (31.83)	28.45 (32.23)	11.47	10.85	11.16	2.14
T_7	Spray of thiophanate methyl 70WP (0.1%) - *P. fluorescens* (0.5%) - thiophanate methyl 70WP (0.1%)	32.13 (34.53)	29.97 (33.19)	31.05 (33.86)	10.13	10.39	10.26	1.99
T_8	Spray of propinconazole 25 EC (0.1%) - *P. fluorescens* (0.5%) - propinconazole (0.1%)	26.40 (30.92)	25.53 (30.34)	25.97 (30.63)	13.63	13.05	13.34	2.53
T_9	Spray of difenconazole 25 EC (0.1%) - *P. fluorescens* (0.5%) - difenconazole (0.1%)	26.53 (31.00)	24.97 (29.98)	25.75 (30.49)	12.83	14.15	13.49	2.50
T_{10}	Two sprays of mancozeb 75WP (0.2%)	30.40 (33.46)	27.63 (31.71)	29.02 (32.59)	11.47	11.68	11.58	2.28
T_{11}	Two sprays of carbendazim (0.1%)	42.40 (40.63)	35.07 (36.31)	38.73 (38.49)	9.53	9.85	9.69	1.91
T_{12}	Control	46.40 (42.94)	40.93 (39.77)	43.67 (41.36)	9.30	9.63	9.46	1.97
	S. Em. (±)	0.36	0.43	0.24	0.72	0.77	0.49	
	CD at 5%	1.06	1.25	0.71	2.10	2.25	1.43	

The treatment with *P. fluorescens* resulted in encouraging result for the management of *Alternaria* blight of pigeon pea. The results are in accordance with Mishra and Arora (2010). They reported that the seed and seedling treatment with *P. fluorescens* significantly managed black rot of crucifer. Seed treatment with *P. fluorescens* (5g kg^{-1}) followed by two sprays of difenconazole (0.1%) interspersed with *P. fluorescens* (0.5 %) resulted in effective management. *P. fluorescens* acted as systemic resistance inducing agent, which increased the vigour of seedling. The additional spray of *P. fluorescens* in between the fungicide has resulted in reduction of fungicide tolerance and decrease in disease incidence. The findings are in accordance with Savitha *et al* (2012) where they discussed about application of *P. fluorescens* in inducing resistance in sesame crop against *A. sesami*. Difenconazole was reported to be effective in reducing the mycelial growth of *A. porri in vitro* (Chethana *et al*., 2011). In the present experiment, though difenconazole was found best in disease managing, the analysis of cost benefit ratio revealed that the efficacy of hexaconazole in reducing the disease was almost at par with good yield. Staub (1991) discussed about development of resistance by anthracnose disease of chilli as against fungicide, if a single compound is relied upon too heavily. So two sprays of hexaconazole interspersed with *P. fluorescens* were found better in managing the *Alternaria* leaf blight of pigeon pea.

CONCLUSION

The study concludes that the *Alternaria* blight of pigeon pea could successfully be managed by two sprays of hexaconazole (0.1%) interspersed with spray of *P. fluorescens* (0.5%) at fortnightly interval starting from the onset of the disease.

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Management of tomato wilt caused by *Fusarium oxysporum* f. sp. *lycopersici* through organic amendments

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ABSTRACT

The present study explores the potential of different organic amendments i.e. vermicompost, spent compost mushroom, calotropis leaf extract, panchgavya and *Trichoderma* against tomato fusarium wilt caused by *Fusarium oxysporum* f. sp. *lycopersici* in pots under greenhouse conditions. The population of *F oxysporum* f. sp. *lycopersici* was significantly reduced after application of organic amendments. Drenching of soil with *Trichoderma asperellum* @10 g^{-1} + Chitosan @ 0.1% (T3) recorded lowest percent disease index (34.40 %) and was at par with chemical control with carbendazim @ 0.1%. Treatment (T1) soil application of FYM + *Trichoderma asperellum* @ 2% w/w and (T2) vermicompost @ 2 % w/w were also effective in minimizing the disease and PDI recorded at 43.30 and 46.63, respectively. Panchgavya @ 30 per cent was also at par with vermicompost recording 49.96 per cent PDI. Maximum PDI was recorded in control (73.30%).

Key words: Tomato wilt, *Fusarium oxysporum* f. sp. *lycopersici*, vermicompost, panchgavya, *Trichoderma asperellum,* Percent Disease Index

Tomato (*Solanum lycopersicon* Mill.) is one of the most popular vegetable crop in India. It is popular due to its very high content of vitamin A and C and also because of presence of good nutrients. Tomato has become most profitable major crop for the farmers of Bihar which they grow throughout year in poly house and field. The area under tomato is 46000 ha with total production of 7.36 lakh tones, with a productivity 16 tonnes ha^{-1}. (Horticultural Statistics at a Glance, 2019). The productivity of this crop is not upto the mark in Bihar and the factors attributed for this is the use of local varieties and the loss caused due to its infestation by large number of insect pests and diseases. Tomato is affected by many diseases and disorders like fusarium wilt, verticillium wilt, damping off, late blight, early blight, bacterial wilt and tomato mosaic virus etc.

Fusarium wilt of tomato is one of the most serious, and devastating diseases of in all the tomato growing regions of India. The development of fungicide resistance and failure of host resistance is also an emerging issue and it is a major concern in the case of soil borne pathogens such as in *F oxysporum* f. sp. *Iycopersici* (McDonald and Linde, 2002). Amendment of soil with organic material such as oil cakes, which is easily decomposable, has been reported in the management of many *Fusarium* spp. (Toussoun et *al.*, 1970). All types of cultural and agronomic practices including the use of crop rotation, use of organic compost and different types of tillage systems controls the soil-borne pathogens like *Fusarium* (Abawi and Widmer, 2000). On the contrary, the use of chemicals have poor effect on the other groups of soil mycoflora as well, especially to the beneficial soil fungi. Generally, the soil-borne diseases are very much severe and often act as the most curbing factor in the conventional system of crop production in agriculture (Cook and Baker, 1983). Considering these points and more demand of organic produce, the present study to find out the most effective organic amendments for the management of tomato wilt caused by *Fusarium oxysporum* f. sp. *lycopersici* was conducted.

MATERIALS AND METHODS

The present investigation was carried out in the Department of Plant Pathology. PG College of Agriculture, Dr. Rajendra Prasad Central Agricultural University, Pusa, Samastipur, Bihar.

The tomato plants showing typical wilt symptoms were collected from the farmer's field in Harpur, Samastipur in polybags and brought to the laboratory for the isolation of pathogen. Isolation of the pathogen was done using the direct plating method of Okhuoya *et al.* (2012). Infected parts i.e. root, stem and leaves were washed thoroughly in the running water, cut into small pieces by the help of a sterilized scalpel, cut bits were surface-sterilized in 1 per cent solution of sodium hypochlorite followed by rinsing thrice in sterile distilled water, dried on a sterile filter paper and then finally plated on the petri-dishes containing Potato Dextrose Agar (PDA) media. The inoculated plates were properly labelled

and incubated at 28ÚC ± 2ÚC for 3 days. Sub-culturing was done by taking little amount of mycelial mass from the culture and inoculating them into the new petri-dishes containing sterilized PDA with the tip of a sterilized inoculating needle and was incubated at 28ÚC ± 2ÚC in the BOD incubator.

The isolated fungus was identified on the basis of visual observations of colony and morphological characters. The culture was stained with the lactophenol blue solution and observed under the microscope for macroconidia, microconidia and chlamydospores.

The efficacy of different organic amendments against *Fusarium oxysporum* f. sp. *lycopersici* was studied in plastic pots (2 kg capacity) filled with well pulverized and sterilized soil under greenhouse conditions at Department of Plant Pathology, Dr. RPCAU, Pusa, Samastipur. Each pot was inoculated with the culture of *Fusarium oxysporum* f. sp. *lycopersici* @ 10g pot^{-1} before 10 days of transplanting. All the treatments were also incorporated in the pots simultaneously after 2 days of inoculation. The 21 days old tomato seedlings (Variety MJ- 3) were transplanted in each pot (5 in no.) after one week of pathogen inoculation while without any treatment served as check. Watering of pots was done as and when required to maintain the optimum level of moisture. The greenhouse chamber temperature was maintained at 25ÚC. The experiment was laid out in a Completely Randomized Design (CRD) with three replications of each treatment. Appearance of typical disease symptoms were monitored and recorded after 45, 60 and 75 days after transplanting.

The eight organic amendment treatments evaluated were, soil application with FYM + *Trichodera asperellum* @ 2% w/w (T_1), soil application with vermicompost @ 2% w/w (T2), soil drenching with *T. asperellum* @ 10 g/l + chitosan @ 0.1% (T3), soil application with spent mushroom compost @ 2% w/w (T4), soil drenching with calotropis leaf extract @ 30% (T5), soil drenching with panchgavya @ 30% (T6), soil drenching with carbendazim @ 0.1% (T7) and Control – inoculated (T8)

The disease severity on inoculated plants was calculated using a scale of 1-6 as described by Marley and Hillocks (1996) as follows:

1= no infection on branches

2= wilting and chlorosis of the first branches

3= wilting and chlorosis of second and third branches

4= wilting and chlorosis above third branches

5= wilting, chlorosis, extensive drying and desiccation

6 = complete wilting and drying, eventually plants die

The Per cent Disease Index % was calculated by the following formula -

$$\text{Per cent Disease Index (\%)} = \frac{\text{Sum of all the disease ratings}}{\text{Total no. of diseased branches observed x Maximum rating}} x100$$

RESULTS AND DISCUSSION

The data presented in table 1 revealed that all the treatments significantly reduced the disease severity as compared to inoculated control. Lowest per cent disease index (34.40 %) was observed with soil drenching of *T. asperellum* @ 10 g/l + chitosan @ 0.1 per cent (T3) followed by 43.3 per cent observed with soil application of FYM + *T. asperellum* @ 2 per cent w/w (T1). Treatment (T1) soil application of FYM + *T. asperellum* @ 2 per cent w/w and treatment (T2) vermicompost @ 2 % w/w were also effective in minimizing the disease and PDI recorded 43.30 and 46.63, respectively. Panchgavya @ 30 per cent was also at par with vermicompost and PDI recorded at 49.96 per cent. However, chemical control with carbendazim @ 0.1 per cent was at par with treatment (T3) and PDI recorded 39.96 per cent. Maximum per cent disease index (73.30 %) was recorded in inoculated control (Plate 1, Fig1). The effectiveness of *Trichoderma*, FYM, vermicompost, plant extracts against different plant diseases have been reported by several worker. The majority of studies on compost suppreressiveness demonstrate a relationship between disease suppression and microbial activity. Suppression of fusarium wilt of tomato using compost could have been caused by compounds such as cyanide, as well as to some of compost's indigenous bacterial strains that act as antagonists. After sterilization, compost lost the ability to suppress *fusarium*. The inability of sterilized compost to

Table 1 : Efficacy of different organic amendments against *Fusarium oxysporum* f. sp. *lycopersici in vivo* (greenhouse)

Treatment	Treatment composition	PDI (%)
T1	SA with FYM + *Trichoderma asperellum* @ 2% w/w	43.30
T2	SA with Vermicompost @ 2% w/w	46.63
T3	SD with *T. asperellum* @ 10 g/l + Chitosan @ 0.1%	34.40
T4	SA with spent mushroom compost @ 2% w/w	61.06
T5	SD with calotropis leaf extract @ 30%	57.73
T6	SD with panchgavya @ 30%	49.96
T7	SD with carbendazim @0.1%	39.96
T8	Control (inoculated)	73.30
C.D. (P<0.5)		5.816
S.E(m) ±		1.923
C.V.		6.558

*mean of three replications

*PDI – Per cent disease index *SA- Soil application, SD –Soil drenching

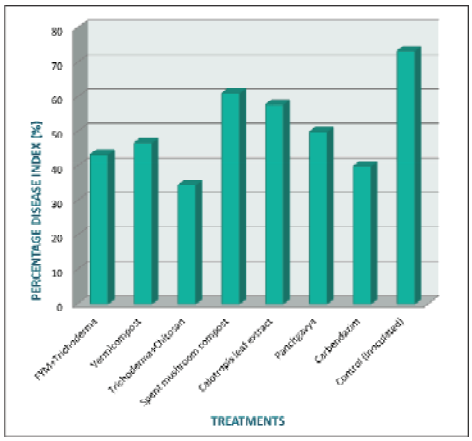

Fig. 1: Efficacy of different organic amendments for management of tomato wilt *in vivo* (greenhouse)

Plate 1: T3- Soil Drenching with *Trichoderma asperellum* @ 10 g/l + Chitosan @ 0.1% T8- Control (Inoculated)

suppress fusarium wilt indicates the importance of compost microflora after biological control. Hoitink *et al.* (2000) showed that compost microflora had no significant effect on the fungus *fusarium oxysporum,* which could account for the less-efficient suppression of the pathogen with the nonsterilized compost. Our results were consistent with those obtained by (Bugg, 1990) who showed that when the compost was sterilized, it did not control disease, and that compost microflora induced host resistance to the pathogen. Several workers have also reported the effectiveness of biological agents like *Trichoderma* singly or in combination with organic chemicals and plant extract (Parvu, 1998; Okigbo *et al*., 2009). Maurya *et al*. (2020) observed good result against tomato wilt when tomato seedling treated with *T asperellum* @ 5g lit^{-1} + chitosan @ 0.1 per cent followed by its foliar spray and PDI recorded 44.66. Akrami and Yousefi (2015) also reported that the use of *T. asperellum, T. virens* and *T. harzianum* controlled the pathogen effectively either separately or in combination when applied as a suitable treatment for management of tomato wilt. Rathore *et al*. (2019) studied the effectiveness of panchagavya in inhibiting the growth of *F solani* and *S rolfsii* in vitro and inhibited growth by 83.3 and 100 per cent, respectively at 10 per cent concentration.

CONCLUSION

The findings indicate that *Trichoderma*, FYM, plant extracts and panchagavya could effectively be exploited in minimizing tomato wilt disease. Among different organic treatments, soil drenching with *T. asperellum* @10g/l + chitosan @ 0.1 per cent recording minimum per cent disease index can be recommended for eco-friendly management of tomato wilt.

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Management of *Alternaria* blight disease in Cumin through plant extracts

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ABSTRACT

Studies on the relative efficacy of plant extracts carried out *in vitro* and *in vivo* conditions indicated that the garlic clove and neem seed karnel exract could provide maximum inhibition of mycelial growth and spore germination of the fungus *in vitro.* Garlic clove (10%) and neem seed karnel (10%), provided effective control of blight disease and significant increase in the cumin seed yield under field conditions.

Keywords: *Alternaria* blight, plant extracts, cumin, NSKE, Garlic

India is one of the largest producers, consumers and exporters of seed spices (Peter *et al.,* 2000). Among the seed spices, cumin (*Cuminum cyminum* L.), locally known as "*Zeera*" in hindi, is one of the important crops belonging to order umbellales and family *umbelliferae* and is believed to have originated from Egypt (Edison and Kallupurackal, 1989). Cumin is popularly used for flavouring food and as herbal medicine and culinary for flavouring vegetables, pickles and soups, etc. Its seeds contain 17.7pre cent protein, 23.8 per cent fat, 35.5 pre cent carbohydrates and 7.7 per cent minerals. It also contains 6.2 per cent moisture, 0.09 per cent calcium, 0.45 per cent phosphorus, 0.048 per cent iron, 1.6 per cent sodium, 2.1 per cent potassium and also vitamin B_1, B_2, niacin, vitamin-A, vitamin-C etc. (Shankaracharya and Natrajan, 1971 and Chadha, 2006). Cumin seeds are aromatic and nutty flavored. Volatile oil from cumin seeds is used in perfumery, liquor, flavoring and cardinals and has stimulatory carminative, stomatic, antidiarrhoeal and dyspepsial medicinal properties (Patel, 1993).

Alternaria blight caused by *Alternaria burnsii* is one of the most dreaded diseases and a major production constraint for the successful cultivation of cumin crop. The blight disease of cumin was first reported from Bombay province and the causal agent was identified as *Alternaria* spp. (Uppal, 1933) but later on the fungus was named as *Alternaria burnsii* (Uppal *et al.,* 1938). The disease is now widespread in all the cumin growing states of India as well as in Pakistan (Shakir *et al.,* 1995). The pathogen being internally and externally seed-borne (Swarup and Mathur, 1972) leads to serious yield losses under favourable weather conditions (Patel and Desai, 1971). Seed losses to the extent of 83 per cent due to blight has been reported. The persistant cold and cloudy weather is congenial for the blight development (Gemawat and Prasad, 1969, Bhatnagar *et al.,* 1995).

MATERIALS AND METHODS

Many phyto-extracts are currently under use as fungitoxicants for the management of various plant diseases. In the percent experiment six different phyto-extracts have been evaluated to see their antimycotic behaviour on *Alternaria burnsii* both *in vitro* and *vivo* conditions. The details of the test plant extracts are given below:

Plant extract	Botanical name	Dose *in vitro* (%)	Dose *in vivo* (%)
Neem Seed Karnel	*Azadirachta indica.*	5, 10, 15	10
Ginger rhizomes	*Zingiber officinale*	5, 10, 15	10
Garlic clove	*Allium sativum*	5, 10, 15	10
Kaner leaf	*Nerium odorum* L.	5, 10, 15	10
Dhatura leaf	*Datura stramonium*	5, 10, 15	10
Aak leaf	*Calotropis gigantia* L.	5, 10, 15	10
Control	-	-	-

The respective plant parts were thoroughly washed with sterile distilled water and air dried. One hundred gram of plant parts was homogenized in 100 ml of distilled water for ten to fifteen minutes. Later, the respective macerates were filtered through double layered cheese cloth and centrifuged at 10,000 rpm for ten minutes. The supernatant was decanted and filtered through bacteria proof filter using vacum filtration unit. The filtered extracts were stored in refrigerator for further use in laboratory studies.

The inhibitory effect of plant extracts were tested on potato dextrose agar (PDA) medium. The plant extracts prepared in distilled water were tested at three concentrations i.e. 5, 10 and 15 per cent. Suitable quantity of plant extract prepared in distilled water was added to sterile and molten potato dextrose agar medium so that the final concentrations of the plant extracts in amended medium were 5, 10 and 15 per cent. The mycelial disc of five mm diameter taken from periphery of seven days old actively growing

culture of *A. burnsii* was transferred at the centre of agar surfaces in Petri dishes. The inoculated Petri dishes were kept in BOD incubator at 25 ± 1^{0}C for seven days. Three replications were kept for each treatment. The mycelial growth was recorded after seven days i.e. when the full growth of pathogen was recorded in control Petri dishes. The potato dextrose agar without plant extract served as control. The inhibition of mycelial growth of *A. burnsii* was calculated as follows: (Vincent,1947)

$$\text{Per cent mycelial growth inhibition} = \frac{C-T}{C} \times 100$$

Where,

C= Mycelial growth observed in control

T= Mycelial growth observed in treatment

Efficacy of plant extracts against cumin blight in fields was studied under artificial inoculated conditions during two *rabi* crop seasons of 2014-15 and 2015-16. Cumin variety RZ-19 was used in the trial. The experiment was conducted in Randomized Block Design (RBD) with three replications in 3 x 3 m^2 plots sized. The crop was sown on 20th November in both the years. The culture of *A. burnsii* was raised on potato dextrose agar. The spore suspension was prepared in sterilized distilled water and the concentration was adjusted to 10-15 spores/microscopic field observed at 10 x magnification. The spore suspension was sprayed 60 days after sowing. The plant extract were sprayed after three days of fungal inoculation. Second spray was given at seven days interval. Percent loss in grain yield was calculated over inoculated control using the following formula:

$$\text{Percent estimated loss in yield} = \frac{\text{Yield of plants protected with plant extracts} - \text{yield of unprotected plants}}{\text{Yield of plants protected with plant extracts}} \times 100$$

RESULTS AND DISCUSSION

In vitro effect of plant extract against *Alternaria burnsii* under *in vitro* conditions

Effect of plant extracts was tested at 5, 10, and 15 per cent concentration against inhibition of mycelial growth of *Alternaria burnsii* by poison food technique. The growth of the fungus decreased with the increase in concentrations of respective plant extracts. The garlic clove was significantly superior in inhibiting the mycelial growth (82.60%), followed by neem seed karnel extract (77.08%), ginger rhizomes (68.88%), *dhatura* leaf extract (59.83%) and aak leaf extract (52.46%) kaner leaf extract was found least effective (48.34%) against inhibition of mycelial growth of the fungus. As the concentration of plant extracts increased, the inhibition of mycelial growth decrease (Table 1).

Table 1 Effect of plant extracts on mycelial growth inhibition of *Alternaria burnsii* after 7 days of incubation at 25±1^{0}C

Plant extracts	Per cent growth inhibition* at different concentration (%)			Mean
	5	10	15	
Neem Seed Karnel	74.24	76.80	80.20	77.08
	(59.50)	(61.21)	(63.58)	(61.40)
Ginger rhizomes	64.20	68.15	74.30	68.88
	(53.25)	(55.64)	(59.54)	(56.09)
Garlic clove	78.20	82.30	87.30	82.60
	(62.17)	(65.12)	(69.12)	(65.35)
Kaner leaf	43.37	48.20	53.44	48.34
	(41.19)	(43.97)	(46.97)	(44.05)
Dhatura leaf	56.10	61.40	62.00	59.83
	(48.50)	(51.59)	(51.94)	(50.67)
Aak leaf	48.54	53.62	55.21	52.46
	(44.16)	(47.08)	(47.99)	(46.41)
Control	0.00	0.00	0.00	0.00
	(0.00)	(0.00)	(0.00)	(0.00)
	SEm±	CD (p=0.05)		
Plant extract (P)	1.44	4.12		
Concentration (C)	0.94	2.70		
P x C	2.50	7.13		

*Average of three replications Figures in parentheses are angular transformed values

Effect of plant extract against *Alternaria burnsii* and seed yield of cumin under field condition

Effect of different plant extracts at 10 per cent concentration was tested against blight of cumin under field conditions. Results (Table 2) of pooled analysis showed minimum disease intensity in garlic clove (40.43%) followed by neem seed karnel extract (43.31%), ginger rhizomes (52.79%), *dhatura* leaf extract (55.53%) and aak leaf extract (58.30%) with decrease in per cent disease intensity to 37.63, 33.18,18.56, 14.33, 10.06 and 07.03, respectively over control. Maximum per cent disease intensity was recorded in kaner leaf extract (60.26%).

Analysis of two years pooled data on seed yield indicated that the garlic clove extract recorded highest seed yield (3.79 q ha^{-1}) followed by neem seed karnel extract (3.59 q ha^{-1}), ginger rhizomes extract (3.21 q ha^{-1}), *dhatura* leaf extract (3.00 q ha^{-1}) and aak leaf extract (2.95 q ha^{-1}) showing 93.37, 83.16, 63.78, 53.06, 50.51 and 47.45 per cent increase in seed yield, respectively over the control. Kaner leaf extract recorded lowest yield of 2.89 q ha^{-1} (Table 2).

Table 2 Effect of plant extracts on blight disease and seed yield in cumin

Plant extract	Concentration (%)	Per cent disease intensity*			Decrease in PDI over control (%)	Yield (q ha^{-1})*			Increase in yield over control (%)
		2014-15	2015-16	Pooled		2014-15	2015-16	Pooled	
Neem seed karnel	10	41.30 (39.99)	45.31 (42.31)	43.31 (41.15)	33.18	3.78	3.40	3.59	83.16
Ginger rhizomes	10	51.13 (45.65)	54.44 (47.55)	52.79 (46.60)	18.56	3.41	3.00	3.21	63.78
Garlic clove	10	38.45 (38.32)	42.40 (40.63)	40.43 (39.48)	37.63	3.95	3.62	3.79	93.37
Kaner leaf	10	57.12 (49.09)	63.40 (52.77)	60.26 (50.92)	7.03	3.03	2.74	2.89	47.45
Dhatura leaf	10	53.65 (47.09)	57.40 (49.26)	55.53 (48.17)	14.33	3.09	2.91	3.00	53.06
Aak leaf	10	55.50 (48.16)	61.10 (51.41)	58.30 (49.78)	10.06	3.01	2.89	2.95	50.51
Control	-	61.44 (51.61)	68.20 (55.67)	64.82 (53.62)	-	2.07	1.84	1.96	-
SEm±		1.24	1.43	1.24		0.12	0.12	0.13	
CD (p=0.05)		3.68	4.24	3.68		0.34	0.36	0.38	
CV		6.46	7.04	6.28		7.24	8.21	8.45	

*Average of three replications Figures in parentheses are angular transformed values

Garlic clove extract was found most effective in inhibiting fungal mycelial growth (82.60%) followed by neem seed karnel (77.08%) *in vitro* and 37.63 per cent disease control and 3.79 q ha^{-1} seed yield under field conditions. Neem seed karnel extract providing 33.18 per cent disease control and 3.59 q ha^{-1} seed yield under field condition was found the next best. Plant extracts are the rich sources of naturally occurring active compounds. Garlic and neem seed kernel extracts might be containing variety of naturally occurring volatile and /or other compounds. Such compounds might be positively active as physical barriers or directly interfere the growth and development in different ways. Further research may provide promising clue against plant pathogens. These results are in confirmation with the findings of Shekhawat and Prasad (1971) who reported that the leaf extracts of *Veronica cinerae* and beet inhibited the growth of *Alternaria burnsii* under laboratory conditions. Sharma (1995) reported the efficacy of *neem* extracts (seed, leaf and bark) and *tulsi* (seed and leaf) *in vitro* against spore germination of *Alternaria burnsii* at various incubation at two concentrations (50% and 100%). All plant extracts were effective but neem bark extract significantly reduced the spore germination of *Alternaria burnsii*. The inhibition of spore germination increased with increase in concentration as well as increase in time interval.

Karade and Sawant (1999) tested extracts of 12 medicinal plants against *Alternaria alternata* and reported 100 per cent inhibition of spore germination in presence of *Allium sativum* extract. Singh and Majumdar (2001) observed that mycelial growth of *Alternaria alternata* responsible for fruit rot of pomegranate was significantly inhibited by extracts of *Azadirachta indica, Datura stramonium, Ocimum sanctum, Curuma longa, Zingiber officinale, Allium cepa* and *Allium sativum* at 15 per cent concentration prepared in distilled water, ethanol or acetone. Patini *et al.* (2005) studied the effect of methanol extract of six medicinal plants *viz., Azadirachta indica, Parthenium histerophorus, Calotropis procera, Datura alba, Eucalyptus globulus* and *Polyalthia longifolia* against Alternaria blight of mustard caused by A. *brassiceae.* In another study Khurana *et al.* (2005) reported the sensitivity of 14 isolates of A. *brassiceae* towards aqueous extracts of *Azadirachta indica, Mentha piperita, Lousonic inermia, Eucalyptus tereticornis, Bougainvillia spectabilis* and *Allium sativum*. Garlic and neem proved to be most effective in inhibiting the spore germination of A. *brassiceae.* Kumari *et al*. (2006) reported the efficacy of five plant extract against Alternaria blight of periwinkle *(Catharanthus roseus)* aqueous extract of garlic clove most effectively inhibited the conidial germination of the pathogen and also reduced the disease severity.

CONCLUSION

The garlic clove extract was found most effective followed by neem seed karnel extracts in both *in vitro* and *in vivo* conditions against blight disease of cumin. The treatment recording lowest disease intensity (40.43%), maximum (37.63) per cent disease control, seed yield (3.79 q ha^{-1}) and highest per cent increase in seed yield (93.61) over the control can therefore be successfully used in managing *Alternaria* blight disease in cumin.

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Bio-efficacy of essential oils in eco-friendly management of *Tilletia barclayana* causing kernel smut in rice

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ABSTRACT

Studies undertaken to assess the antifungal efficacy of some essential oils and aroma chemicals @ 1000 ppm against *Tilletia barclayana* (Bref.) Sacc. & P. Syd., causing kernel smut of rice, revealed that palmarosa oil alone is significantly superior showing inhibition of 77.78 per cent in teliospore germination followed by cedar wood oil (72.22 per cent). Among aroma chemicals, the phenyl ethyl propionate showed potent antifungal efficacy with maximum inhibition of 83.70 per cent in teliospores germination. The findings positively provide an impetus for the development and commercialization of plant-based pesticide with higher potency for management of kernel smut of rice.

Key words: Rice, kernel smut, essential oil, aroma chemical, bio-efficacy

Rice is one of the most important cereals of the world and is a staple food for nearly half of the world's seven billion people. It is globally produced with annual production of about 596.74 million tonnes and average productivity of 4.4 t ha^{-1}, whereas, in India, its production is 120.62 million tonnes with relatively low productivity of 3.3 t ha^{-1} (FAO 2020). The rice crop under natural conditions is vulnerable to more than 40 parasitic diseases that are responsible for taking away a big chunk of production. Among the seed-borne fungal diseases, the black smut/ covered smut/ grain smut/ kernel bunt/ paddy bunt/ kernel smut caused by *Tilletia barclayan* (Bref.) Sacc. & P. Syd. (syn.: *Neovossia horrida* (Takah) Padwick & A. Khan) is considered economically important, causing stunting of seedlings, reduction in tillers and quite serious losses in yield and may pose serious threat to most rice producing countries including Bangladesh, Brazil, Burma, China, India, Indonesia, Italy, Japan, Korea, Malaysia, Mexico, Nepal, Pakistan, Philippines, Senegal, Thailand and USA as yield losses as high as 15.00 per cent (CAB International 2007). Kernel smut or bunt caused by *T. barclayana* appearing now in higher incidence becomes a major problem in seed certification because of quarantine concern in several countries (Pannu et al. 2002).

The perusal of literature reveals that most of the fungicides and management options used are ineffective against this destructive pathogen (Sharma and Sharma 2004). Therefore, an attempt was made to assess the antifungal efficacy of some essential oils, aroma chemicals as well as physical method such as hot water treatment (HWT) alone and in combination to minimize seed-borne inoculum of *T. barclayana*.

MATERIALS AND METHODS

To detect *T. barclayana* in paddy (kernel smut), the seed samples were first examined visually and then under stereo-binocular microscope for the presence of infected seeds. Later, seeds were subjected to NaOH test (Mathur and Kongsdal 2003). The infected seeds showed shiny jet-black discolouration. To confirm the identity of the pathogen, smutted kernels were ruptured in a drop of water to obtain smut spores. Mounts were prepared for compound microscopy. Three sets of infected seed samples were taken for further investigation.

Spore germination using poisoned food technique (Schmitz 1930) and hanging drop method (John 1969) was used to study the effect of essential oils, aroma chemicals and HWT on inhibition of teliospore germination of *T. barclayana*. To assess antifungal efficacy, nine essential oils, i.e. T_1= cedar wood oil; T_2= citronella oil; T_3= cotton seed oil; T_4= geranium oil; T_5= karanj oil; T_6= lemon grass oil; T_7= neem oil; T_8= palmarosa oil; T_9= patchouli oil and six aroma chemicals, i.e. T_{10}= eugenol; T_{11}= methyl anthranilate; T_{12}= terpenyl; T_{13}= anethol; T_{14}= Winter green; T_{15}= phenyl ethyl propionate were evaluated at concentration of 1000 ppm separately and in combination with HWT. In addition, a fungicide, vitavax @ 500 and 1000 ppm as well as sterilized distilled water served checks to compare efficacy. To determine the effect of temperature, two sets were prepared by giving HWT at 52±1°C and 57±1°C for exposure period of 30 minutes followed by drying of seed samples in dryer at 42±1°C for 48 hr and third set was without HWT. Smutted kernels from all the three sets were picked up and teliospore suspensions were made using sterilized distilled water maintaining concentration. Desired quantity of essential oils, aroma chemicals and fungicide were added separately.

In poisoned food technique, suspension of teliospores (8-10 μl^{-1}) was spread uniformly @ 0.5 ml/petri plate containing water agar (2.0%) amended with different treatments separately and on unamended water agar which served as check. Petri plates were incubated at 27±1°C for 96 hours in triplicate manner. Similarly, in hanging drop method, teliospore suspensions (8-10 μl^{-1}) of each treatment was placed separately @10 µl on cavity glass slides. These slides were later placed in petriplate having moist blotting sheets to maintain humidity and incubated at 27±1°C for 96 hours in triplicate manner. Teliospore germination was recorded. The percent inhibition was calculated by the following formula described by Vincent (1947).

$$\text{Per cent inhibition} = \frac{\text{Germination in check - Germination in treatment}}{\text{Germination in check}} \times 100$$

The data pertaining to inhibition of teliospores germination were analyzed statistically using OPSTAT after suitable transformation in all the experiments using completely randomized design for efficacy of treatments (reference). Further, correlation analysis was done to assess the effectiveness of various treatments, through scattered plotting using Excel 2016 and heatmap through principal component Aanalysis (PCA) using ClustVis software (Metsalu and Vilo 2015).

RESULTS AND DISCUSSION

Seed health testing of ~1800 rice seed samples collected from 16 states representing different agroclimatic zones of India revealed that crop under natural conditions was often affected by *T. barclayana* showing variable incidence in different states. Further, observations revealed that the smutted seeds were detected in the samples from Odisha, Haryana, Punjab and Andhra Pradesh only. Moreover, the maximum infection frequency (incidence) was observed from Odisha followed by Punjab and Haryana. Whereas, seed infection ranging from 0.2 to 0.7 per cent in Odisha, 0.3 to 0.8 per cent in Haryana, 0.7 to 1.9 per cent in Andhra Pradesh and 0.7 to 3.5 per cent in Punjab was observed across the states (Table 1).

Table 1: Detection of Kernel smut in rice germplasm from different states

Source	Accession	Infection (%)
Jorhat, Assam	IC275935	0.0 - 1.0
Cuttack, Odisha	IC513877	0.2 – 0.7
Karnal, Haryana	IET24537	0.3 - 0.8
Ludhiana, Punjab	PR 106	0.7 – 3.5
West Godavari, Andhra Pradesh	MTU-1155	0.7 - 1.9

In order to assess antifungal efficacy of essential oils and aroma chemicals @ 1000 ppm against *T. barclayana* as well as inhibitory effect of essential oil alone and in combination with HWT, the teliospore germination was observed and the data on inhibition are presented in Table 2 and Figure 1-2. Among treatments, phenyl ethyl propionate showed maximum inhibition (83.70%) in teliospore germination followed by palmarosa (77.78%) and cedar wood oil (72.2%) at room temperature.

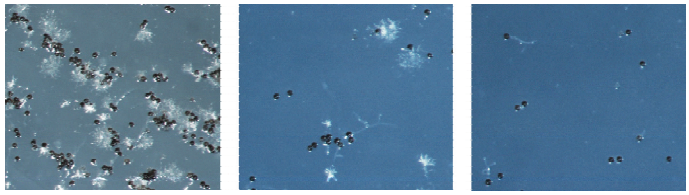

Figure 1. Effect of hot water treatment on teliospore germination on water agar at; a) 57±1°C, b) 52±1°C, and c) without HWT (control).

Table 2. Evaluation of essential oils and aroma chemicals in combination with hot water treatment on teliospore germination of *T. barclayana*

Treatment	Spore germination at different temperature					
	WHWT		HWT at 52±1°C		HWT at 57±1°C	
	(G)	(I)	(G)	(I)	(G)	(I)
Essential oil						
T_1= cedar wood	25.00*	72.22	22.50	71.88	20.00	69.23
T_2= citronella	90.00	0.00	77.50	03.13	55.00	15.38
T_3= cotton seed oil	81.94	8.95	62.00	22.50	47.50	26.92
T_4= geranium oil	30.00	66.67	32.50	59.38	30.00	53.85
T_5= karanj oil	31.79	64.68	31.79	60.26	31.79	51.09
T_6= lemon grass oil	32.50	63.89	32.50	59.38	25.00	61.54
T_7= neem oil	45.00	50.00	35.00	56.25	25.00	61.54
T_8= palmarosa oil	20.00	77.78	32.50	59.38	20.00	69.23
T_9= patchouli oil	75.00	16.67	70.00	12.50	55.50	14.62
Aroma chemical						
T_{10}= eugenol	85.00	5.56	67.50	15.63	57.00	12.31
T_{11}=methyl anthranilate	81.67	9.26	80.00	00.00	56.67	12.82
T_{12}= terpenyl	75.00	16.67	57.00	28.75	52.50	19.23
T_{13}= anethol	68.33	24.07	60.00	25.00	52.50	19.23
T_{14}= winter green	42.50	52.78	32.50	59.38	22.50	65.38
T_{15}= phenyl ethyl propionate	14.67	83.70	17.17	78.54	14.67	77.43
Fungicide						
T_{16}= Vitavax (500 ppm) control	68.33	24.07	58.33	27.09	57.50	11.54
T_{17}= Vitavax (1000 ppm) control	47.50	47.22	47.50	40.63	35.00	46.15
T_{18}= Untreated control	90.00	0.00	80.00	00.00	65.00	00.00

CD (p=0.05) for main treatments = 0.70
CD (p=0.05) for sub-treatments = 0.28
CD (p=0.05) for interaction = 0.16
*Values are arcsine transformed before analysis

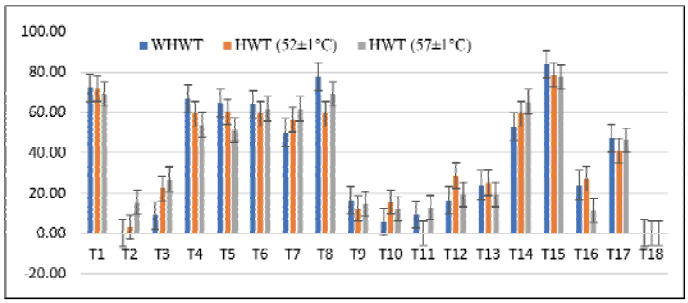

Figure 2. Effect of essential oils, aroma chemicals and vitavax on inhibition of teliospores germination of *Tilletia barclayana*.

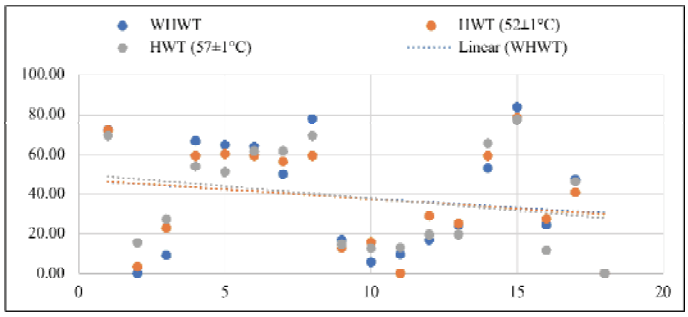

Figure 3. Scattered plotting toward effectiveness of essential oils, aroma chemicals and vitavax in combination with HWT at 52±1 and 57±1°C against teliospores germination of *Tilletia barclayana*.

Hot water treatment left no significant impact in inhibition of spore germination at 52±1 and 57±1°C except increase in inhibition by cedar wood oil with HWT 52°C °C (75%), neem oil and lemon grass oil with HWT 52±1°C (68.75%). The vitavax fungicide (1000 ppm) inhibited spore germination less than 50% both at room temperature and in combination with HWT (Table 2). This is also evident from scattered plotting (Fig. 3) which revealed negative correlation between variables, i.e. temperature and essential oils treatments. Similarly, PCA matrix also described patterns of differentiation with very poor correction between these two variables (Fig. 4).

The study revealed that kernel smut (*T. barclayana*) is now appearing with higher incidence and posing serious problem in maintaining quality seed production through seed certification programme as well as quarantine clearance during international exchange of rice germplasm. Antifungal efficacy testing of various essential oils revealed that hot water treatment has no significant effect on inhibition of teliospore germination. Similarity, Chahal et al. (1993) also reported no inhibition in teliospore germination. Instead, he reported increase in spore germination in bunted grains when subjected to HWT at 60°C for at 30 min. whereas, Stroube (1954) has marked the increase in germination up to

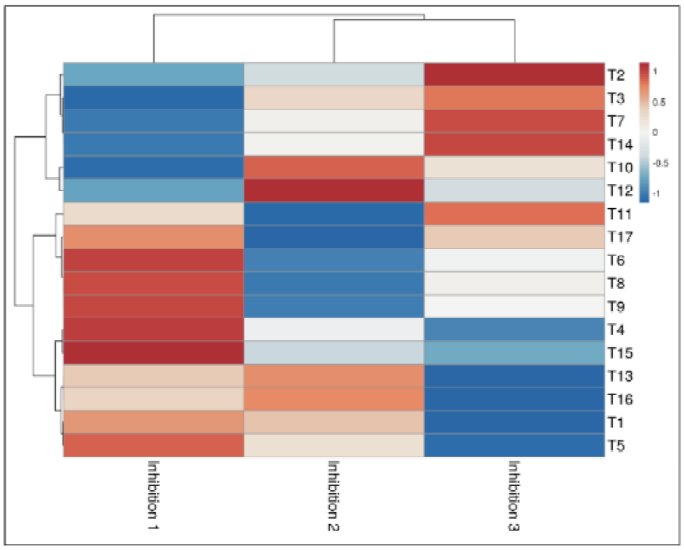

Figure 4. Correlation heatmap generated through Principal Component Analysis (CA); Inhibition 1= inhibition without hot water treatment, Inhibition 2= inhibition with hot water treatment (52±1°C) and, Inhibition 3= inhibition with hot water treatment (57±1°C).

12 per cent by treatment at 70°C for 30 min. Sharma and Sharma (2004) also reported that vitavax is not very effective to control kernel smut of rice. Among essential oils, palmarosa oil @1000 ppm could inhibit teliospore germination to the tune of 77.78 per cent followed by cedar wood oil (72.2 per cent) without hot water treatment. Whereas, among constituents of essential oils, phenyl ethyl propionate showed maximum inhibition of 83.70% in teliospore germination. Dev et al. (2004) study reported *in-vitro* inhibition of 16 aroma chemicals against five seed-borne fungal pathogens viz., *Bipolaris sorokiniana, Phomopsis sojae, Fusarium solani, Colletotrichum graminicola* and *Macrophomina phaseolina* and 2-phenethyl propionate along with eugenol, methyl eugenol, citral, 2- phenethyl alcohol and 2-phenethyl acetate were found to be highly effective against all these fungi. Karanj, geranium, lemon grass and winter green oil also caused more than 50% inhibition in teliospore germination. Hammer et al. (1999) also reported significant inhibition by lemon grass, cedar wood oil, citronella, eucalyptus, winter green oil against 10 fungal pathogens. The application of these essential oils/ aroma chemicals could be an effective way to manage the seed-borne infection of kernel smut of paddy which is an eco-friendly substitute instead of chemical fungicides for managing this disease. There was significant inhibition in teliospore germination with application of different essential oils/ aroma chemicals.

CONCLUSION

Essential oils are famous naturally occurring phytochemicals throughout the world for various applications. Now, demand for safe and effective natural products is increasing. Hence, findings on antifungal efficacy of essential oils provide an impetus for the development and commercialization of plant-based pesticide with higher potency for management of kernel smut and thereby benefiting various stakeholders such as seed certification and plant quarantine agencies, researchers and farmers.

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Evaluation of bioagents against *Macrophomina phaseolina* and its effect on root weight of cowpea (*Vigna unguiculata* (l.) Walp)

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ABSTRACT

The efficacy of bio-agents on dry root weight of cowpea (*Vigna unguiculata* (L.) Walp) studied consecutively for three year at bundelkhand region showed maximum increase in the dry weight of root due to *Trichoderma harzianum* followed by *Trichoderma viride* at various doses. Both the biological agents acted more effectively at 10 to15 g dose. The study also revealed that *T. pseudokoningii* and *T. koningii* did not visualize any significant difference in their efficacies when compared with each other. *Aspergillus flavus* and *Aspergillus niger* were least effective in comparison with other bioagents

Key words: Bioagents, *Macrophomina phaseolina*, Root rot, Cowpea,

The cowpea (*Vigna unguiculata* (L) walp), belonging to family Fabaceae, is one of the most ancient and important fodder legume crop known to man. Cowpea is known to be the most important heat-loving, drought-tolerant crop with high protein content and lower soil fertility requirements than many other crops (Coetzee, 1995). It is commonly grown in Northern and Central India besides in some parts of Rajasthan, Gujarat, Maharashtra, Karnataka and Tamilnadu. It is now rapidly spreading to the rest of the country due to evolution of better varieties suited to different agro-climatic zones/region. In addition, it is shade tolerant, and therefore, compatible as an intercrop with maize, millet, sorghum, sugarcane and cotton (Blade *et al*., 1992; Singh *et al*., 1997). It is not only one of the cheapest source of nitrogen and palatable nutrition for livestock but also valuable in restoring and maintaining its structure and soil fertility. It being rich in protein contents forms an excellent mixture with maize, sorghum and bajra and teosinite for increasing the flow of milk. It can be fed green, good quality hay or as silage after mixing with cereals. Its other uses, it can be used as green manure, cover crop in plantation, vegetable or as a pulse.

Cowpea is infected by a number of pests and plant pathogens. Among these, the fungi, *Macrophomina phaseolina* (Tassi) Goid is the most widespread and destructive causing dry root rot in cowpea. The pathogen infect the crop alone or as a complex resulting in root rots before and after emergence of seedlings and wilting of plants (Sumner, 1985; Gokulapalan *et al,* 2006). The fungus is mainly a soil borne pathogen that spread from plant to plant through irrigation water, food and implements during cultural operations. The pathogen affect the cowpea resulting in low yield (Sumner, 1985; Singh and Gurha, 1996; Bhatnagar and Bansal, 2003; Gokulapalan *et al,* 2006). *M. phaseolina* (Tassi). It has a wide host range and can infect the root, lower stem and is responsible for causing losses on more than 500 plant species (Chidambaram and Mathur, 1975; Dhingra and Sinclair, 1977; Dhingra and Sinclair, 1978). The losses in green fodder and seed yield were estimated to be 28.8 and 39.7 per cent (Ram and Gupta, 1988). Management of soil borne fungal pathogens is difficult due to its long term survival and wide host range. Theses pathogens not only persist in the soil as saprophytes along with other soil organisms but also is transmitted from seed. Considering the hazards of pesticides on public health and the environment there is need to find alternative to synthetic fungicides conventionally used in its management. So there has been growing awareness towards biological control of plant pathogens, which could bring about reasonable reduction in the crop damage, ensure sustainability of production, cost effectiveness and provide healthy eco-system (Cohen Kupiec *et al.*, 1998). Use of native microorganism has gained much interest among the growers and researchers. It has been well documented that *Trichoderma* spp. boost the crop growth and suppress the diseases in various economically important crops. It protect the plant from plant pathogens by responses that are similar to systemic acquired resistance (SAR). It mobilizes the locked-in phosphorus and increases the uptake of Fe, Mn, Cu and Zn in cucumber crop (Yedidia *et al.,* 2001). Therefore, the present study on evaluation of the efficacy of different doses of native bio-control agents against the root rot pathogens and its effect on root weight of cowpea was aimed at.

MATERIAL AND METHODS

The experiments was carried out under pot conditions for three consecutive crop seasons at the Indian Grassland and Fodder Research Institute, Jhansi using root rot susceptible cowpea variety IFC-901. Isolation of *M. phaseolina* was done in culture media from the collected field diseased plant part. The antagonistic biological agents were isolated from rhizosphere of cowpea fields in bundelkhand Jhansi region and further isolation was carried out by serial dilution. *Trichoderma* species *viz*., *T. harzianum, T. koningii, T. pesudokonigii* and *T. viride* were isolated on *Trichoderma* specific media (TSM) from the root zone of cowpea grown at the Indian Grassland and Fodder Research Institute, Jhansi. The fungal antagonists were further used as seed treatment against *M. phaseolina* under pot conditions consecutively for three year to determine the plant growth promoting activity in terms of dry weight of these isolates by seed treatment. Dry powder of the antagonist was prepared by growing the antagonists on soaked and sterilized sorghum grains for 20 days.

Different bioagents were tested under pot condition. Fifteen cm diameter earthen pots were filled with 1 kg autoclaved soil manure mixture of 3:1. These pots were pre inoculated with the test pathogens separately. Different doses of inoculum *viz*. 2 g, 5 g, 10 g and 15 g were prepared and treated with cowpea seed. Treated cowpea seeds were sown. The pots were arranged in complete randomized block design and irrigated daily. There were 3 replicates of each treatment including the treated control as well as untreated control. Observations in respect of dry weight of root were recorded. The plants after 50 per cent flowering were uprooted and dry root weight was recorded. The data were analyzed by the standard statistical methods.

RESULT AND DISCUSSION

The effect of bioagents on the dry root weight of cowpea is depicted in table 1. The pooled data of three year experimentation (table 1) revealed that all the treatments were significantly superior in increasing the dry root weights of the plant as compared to untreated control. Among all treatments, the most effective bioagent found against *M. phaseolina* was *T. harzianum*. It was significantly superior over others offering maximum dry root weights(150.0%) followed by *T viride* (141.7%) over untreated control. The next most effective biological agents that increased the dry root weights of cowpea plant were *T. pseudokoningii* (129.5%) and *T. koningii* (116.7%). It was worthy to note that these bioagents did not visualize any significant difference in their efficacy when compared with each other.

Aspergillus was found least effective in enhancing the dry root weight when compared in their efficacy with other bioagents. *A. flavus* exhibited only 84.6 per cent and *A. niger* exhibited 101.9 per cent increase in dry root weights of plants. All the bioagents were effective @15 g kg^{-1} seed dose and least @ 2 g kg^{-1} seed. The recommended dose of systemic fungicide, bavistin (@2g kg^{-1} seed) offered 148.7 per cent increase in dry root weight over untreated control. *A. flavus* proved least showing minimum dry root weights. Bavistin was also effective in increasing the dry root weights and was statistically *at par* with *T. harzianum.* Similar results were also obtained by many workers (Mukhopadhyay *et al.,* 1992; Mukhopadhyay, 1995; Harman *et al*., 2004). *Trichoderma* spp. stimulates growth and flowering of several plant species (Chang *et al*., 1986; Windham *et al*., 1986; Hanson and Howell, 2002). *Trichoderma* strains have been developed for promoting activities of non-pathogenic bacteria and mycorrhizal fungi, while others act as plant-growth promoters by increasing plant size, foliar surface area and weight. Recently more important role of *Trichoderma* spp. has been noticed. They induce localized and systemic resistance in susceptible plants. The dual roles of antagonistic activity against plant pathogens and promotion of soil fertility make *Trichoderma* strains most appealing alternatives to hazardous fumigants and fungicides. These findings are in accordance with the finding of the earlier workers (Datnoff, *et al*., 1995; Harman and Kupicek, 1998;Bhatnagar and Bansal, 2003; Harman *et al*., 2004; Patibanda and Sen, 2007). Mukhopadhyay (1996) and Zaidi *et al.* (2004) also demonstrated increase in growth and weight of crop plants with *T. harzianum* and *T. virens*. These responses may be due to suppression of deleterious root

Table 1: Efficacy of bioagents on dry root weight of cowpea infested with *M. phaseolina*

Treatments	Dry root weight* (gm) Doses (g kg^{-1})				Average	Increase over control (%)
	2g	5g	10g	15g		
A. flavus	0.70	0.71	0.73	0.74	0.72	84.6
A. niger	0.77	0.78	0.79	0.81	0.78	101.9
T. koningii	0.83	0.84	0.85	0.86	0.84	116.7
T. pseudokoningii	0.88	0.89	0.90	0.91	0.89	129.5
T. viride	0.92	0.94	0.95	0.96	0.94	141.7
T. harzianum	0.96	0.97	0.98	0.99	0.97	150.0
Bavistin (Treated check) @ 2g kg^{-1}	0.97	0.97	0.97	0.97	0.97	148.7
Control (untreated check)	0.39	0.39	0.39	0.39	0.39	-

CD 5% Treatment 0.01
Doses 0.01
Interaction 0.00

Each value is mean of three replicate.

micro-flora including those not causing obvious disease, Production of growth stimulating factors, Increased nutrient uptake through solublization and sequestration of nutrients and Enhanced root growth.

CONCLUSIONS

It was concluded that *Trichoderma* spp. (*T. harzianum*, *T. viride*, *T. pseudokoningii)* was effective in controlling the root rot disease and in enhancing the dry root weight of cowpea. *T. harzianum* at 15 g dose was significantly superior offering maximum fresh shoot weights. Hence, *Trichoderma* spp. can be exploited for the management of root rot disease of cowpea in place of bavistin (systemic fungicide) without disturbing the ecological balance. These bioagents are cheaper and can be multiplied on commercial scale on FYM, pressmud and other biological wastes and applied to the soil in disease prone areas to check soil borne pathogens especially the *M. phaseolinal*.

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Evaluation of indigenous seed treating materials in managing diseases of fennel (*Foeniculum vulgare* Mill)

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ABSTRACT

The field trial conducted at Agronomy farm, S.K.N. College of Agriculture, Jobner during rabi 2015-16 & 2016-17 in randomized blocked design (RBD), evaluating the efficacy of seed treatments viz., - cow urine (2.5%), asafoetida-heeng (1.2%), castor oil seven (5%), garlic clove extract (5%) - turmeric powder (5%), T6- turmeric extract (5%) and neem oil (5%) in the management of diseases of fennel indicated that all the treatments were found superior over control. The germination per cent was highest in garlic treated plots followed by neem oil treated plots. The neem seed oil treatment followed by garlic treatments recorded the maximum control of wilt diseases. The intensity of alternaria blight and powdery mildew diseases was found least in garlic-treated plots followed by neem and castor oil-treated plants.

Key Words: Seed borne pathogen, wilt, root rot, powdery mildew, management, fennel, indigenous technology, plant extract, eco-friendly

India is in the first position concerning producer, consumer and exporter of seed spices in the world. The seed spices primarily are grown in *rabi* season occupying about 51.79 per cent of the total area and 19.06 per cent of spices produced in the country. The total area under cultivation of seed spices was about 3970769 hectares, with about 9268081 tons (Anonymous, 2020a). The largest production center state of fennel in India is Gujrat and Rajasthan having more than 80 per cent area under fennel cultivation. Rajasthan stands in the second position in the production of the fennel. The export of fennel in 2018-19 was 26250 tonnes fetching Rs. 24412.50 lakhs (Anonymous, 2020b). Seed spices are used to impart flavour and taste to food and have huge medicinal properties for health benefits to treat multiple diseases. Though seed spices have been used in Unani and Ayurvedic medicines, seed spices are an essential crop and confined in semi-arid and arid parts of the country. In traditional medicine, the plant and its essential oil have been extensively used as carminative, digestive, galactagogue and diuretic and treat respiratory and gastrointestinal disorders. It is also used as a constituent in cosmetic and pharmaceutical products. The essential oil of *F. vulgare*, in particular anethole, exhibits antispasmodic, carminative, anti-inflammatory, estrogenic and anti-microbial activities. Fennel oil possesses antioxidant, anti-microbial, insecticidal, antithrombotic and hepatoprotective activities (Sharopov *et al*., 2017).

Fennel (*Foenicuilam vulgare* Mill), is well known for its essential oil having characteristic anise odour that makes it an excellent flavouring agent in baked goods, meat and fish dishes, ice cream and alcoholic beverages. The major components of *F. vulgare* seed essential oil have been reported to be trans-anethole, fenchone, estragol (methyl chavicol), and a-phellandrene (Rather *et al.*, 2016). Fennel is a multipurpose crop, and it is consumed as food, fodder, fuel, medicines, perfumery, cosmetic products, and used in religious rituals. Increasing health consciousness among the spice user, especially the European and American community, leads to the growing demand for organic crop products, including spices. It has led to the development of international trade for organic herbs. India is traditionally cultivating spices organically. However, there is a need to increase the area under the organic cultivation of fennel. To fulfill the international standard of making the consignments free from pesticide residue, our prime concern should to reduce the use of agrochemicals by promoting the application of agriculturally beneficial microorganisms as a safer and

potential alternative to traditional agricultural practices for crop health management (Abhilash *et al*., 2016). Exporters specializing in organic production have successfully achieved the international standards prescribed for spices (Malhotra and Vashishtha, 2007; Lal, 2018). In this WTO era, pesticide residue hazards in major seed spices have become the major concern due to the rejection of consignment for international export purposes. According to Shafique (2016), the chemical methods are easy, quick, and cost-effective; but, they create an imbalance in the environment, adversely affect human health, damage aquatic ecosystems, harm pollinators and reduce populations' benefits microorganisms in the soil. To reduce pesticide residue load, the adoption of alternative management strategies, which are eco-friendly and effective, should be prioritized (Chattopadhyay et al., 2018). Fennel crop have ample scope for expansion of areas under different agro-climatic zones.

A seed-borne pathogen growing externally, internally or associated with the seed as a contaminant cause seed abortion, rot, necrosis, reduction or elimination of its germination capacity. The seedling damage causes plant disease development at later stages of its growth (Salem *et al*., 2019). Many pests attacked seed spice crops due to their specific aroma, nectar, and pollen on crops. The population of insects increased with the increase of temperature in Rajasthan. The disease infestation in the fennel also fetches low income due to poor quality. Disease like leaf spot, leaf blight, damping-off, root rot, stem rot, Ramularia blight (*Ramularia foeniculaii*), wilt, downy mildew, powdery mildew in fennel frequently attack the crop, causing heavy losses of yield and deteriorating the product's quality (Jain and Jain, 1995). Salem *et al*., 2019 reported *Botrytis cinerea, Sclerotinia sclerotiorum, Cladosporium cladosporiodes, Cladosporium link* and *Bispora* sp. along with *Aspergillus niger* and *Penicillium digitatum* isolated from fennel as seed-borne fungi showed inhibition of radial growth above 50 per cent. Among them, the seed-borne pathogens reduce germination and stand establishment because of the slower growth rate. Seed treatment is one of the vital practice which can enhance the productivity of fennel.

A number of indigenous practices and materials were recommended in ancient and medieval times to manage seed health with brief information on their properties (Nene, 1999 (Singh and Sureja, 2008)). Among them honey, ghee/oils, cow dung, cow milk, extract of root/branches/leaves, ashes, chalk, rice flour, sesame & black gram, with wheat powder and turmeric powder were used for seed treatment to improve seed health by disinfecting the seed-borne microflora (Gahukar, 2011; Kumar,2014; Nene, 1999; Sridhar *et al*., 2013; Attri *et al*., 2020). Organic crops are the demand of the day, and spices are also preferred to be cultivated without chemicals. Many local practices which were used in yesteryears are disappearing. Auger *et al*., 2004 reported that the crushed *Allium* spp. containing thiosulfinates and disulfides that worked as the same spectrum of pesticide activity of methyl bromide, can be used in agriculture as a tool in effective integrated disease-management tool.

The present investigation aims at to evaluate and scientifically document the efficacy of indigenous seed treating materials (Kumar, 2014) in managing the significant diseases like wilt, powdery mildew and stem rot of fennel responsible for decreasing the productivity of fennel in the state. The peculiarity of this knowledge is that it is cheapest, eco-friendly, ecologically protective, socially acceptable, economically viable and sustainable. In appreciation of technical know-how of ethnic groups, an attempt was made to collect, document, and understand the rationale behind the traditional practices in general and pest management, mainly disappearing at a fast rate under the influence of high tech modern agriculture.

MATERIALS AND METHODS

Fennel seed samples were collected during the year 2014-15 & 2015-16 from Tonk, Kota, Baran, Bundi, Jhalawar Sirohi, Jalore Nagaur, and Sikar districts. Informations about indigenous materials used by farmers were also collected. The seed samples of each district, after proper recording and marked with separate accession number, were packed in paper envelops, labeled and kept in steel almirahs. For recording seed-borne mycoflora, the seed health testing procedures as prescribed by International Seed Testing Association (ISTA, 2005) were followed. Isolation of external and internal seed mycoflora of fennel was done using standard blotter and ages plate methods.

The standard blotter method involving three pieces of filter paper (Limonard, 1968), adequately soaked in sterilized water and placing these at the bottom of a 9 cm well labelled plastic petri dishes was used. Twenty seeds per petri dish were placed equidistantly under aseptic conditions. The lids of each petri dish were held in place with adhesive cello tape. The petri dishes containing seeds were incubated at room temperature (25 ± 2°C) for seven days under alternating cycles of light and darkness of 12 hours each (Ghosh *et al*., 2018).

In the agar plate method (Agarwal, 1976), 20 ml of potato dextrose agar (PDA) was distributed in sterile petri plates under aseptic conditions. After cooling, twenty seeds per Petri plate were kept at equidistance in a circle (Barnet and Hunter, 1999) and incubated at room temperature (25 ± 2°C)

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under 12 hours alternating cycles of light and darkness for seven days. Observation on the growth of fungi (ISTA, 2005) was recorded everyday. The per cent seed mycoflora and percentage frequency of various fungal species were calculated. The incidence per cent of fungi on seed was calculated as per formula mentioned below:

$$\% \text{ incidence} = \frac{\text{Number of infected seeds}}{\text{Number of plated seeds}} \times 100$$

Fungal species with maximum incidence on fennel seeds were further tested for pathogenicity. Isolated mycoflora were purified by the single spore technique and hyphal tip method. In the single spore technique, serial dilutions of spore suspension from 7-day old culture were made in sterilized double distilled water until a dilution containing 10-15 spores/ml was achieved. One ml of this diluted spore suspension was poured in petri plates containing solidified 2 per cent plain agar autoclaved at 15 lb pressure for 20 min. under aseptic conditions. The spore suspension was evenly distributed by tilting the petri plate in various directions. After few minutes, the excess suspension was removed from the petri plate. Inoculated petri plates were allowed to be incubated at 25±1°C for 24h. Germinating single spore was located and marked under the microscope with the help of dummy objective and transferred on 2 per cent PDA slants, aseptically. Inoculated slants were allowed to grow and sporulate. In hyphal tip method a single spore, a hyphal tip was marked and transferred on two per cent PDA slants. The identity of the *Alternaria spp* and *Fusarium spp*. obtained on PDA slants was confirmed by ITCC, New Delhi, with ID No. 9256.13 for *A. alternata* & ID No.8869.13 for *F. oxysporum*. The pathogenicity of both the fungus was proved.

Field trials to evaluate the efficacy of indigenous seed treating material against major diseases of fennel were conducted during *rabi* 2015-16 & 2016-17 in randomized blocked design (RBD) with three replications using local cultivar of fennel at Agronomy farm of S.K.N. College of Agriculture, Jobner. All the recommended agronomic practices were followed to raise the crop. The experiment consisted of eight indigenous seed treating materials viz., T_1- cow urine @ 2.5 per cent, T_2- heeng (asafoetida) @ 1.25 per cent, T_3- castor oil @ 5 per cent, T_4 – garlic clove extract @ 5 per cent, T_5 –turmeric powder @ 5 per cent, T_6- turmeric extract@ 5 per cent, T_7- neem oil@ 5%, T_8 – inoculated control. Root rot, powdery mildew, and downy mildew disease were recorded under natural conditions while the wilt incidence was recorded under artificial inoculation conditions. Data on per cent germination, disease incidence and intensity of all the diseases were recorded.

RESULTS AND DISCUSSION

The results presented in table 1 indicated that the mean germination per cent was best in garlic treated plots (90.54%) followed by neem oil treated plots (87.80%) compared to untreated control (69.93%). The incidence of wilt was minimum with neem oil (1.05%) followed by garlic (1.37%) as against (32.91%) in the control. Root rot disease was found least in garlic treated plots (5.5%) followed by neem oil treatment (9.89%) compared to control (40.66%). The intensity of powdery mildew disease was best managed with the garlic

Table 1: Evaluation of indigenous seed treating materials for managing the disease of fennel

Treatment		Germination (%)			Wilt(PD Incidence)			Blight(PDI)			PM(PDI)			Yield (q ha^{-1})		
		2015-16	2016-17	Mean	2015-16	2016-17	Mean	2015-16	2016-17	Mean	2015-16	2016-17	Mean	2015-16	2016-17	Mean
T_1	Cow urine	71.22 (57.56)	73.45 (58.98)	72.34 (58.27)	11.64 (19.95)	9.46 (17.91)	10.55 (18.95)	27.66 (31.73)	22.33 (28.20)	25.00 (30.00)	25.46 (30.30)	20.10 (26.64)	22.78 (28.51)	8.00	10.10	9.05
T_2	Heeng	72.05 (58.08)	76.88 (61.26)	74.47 (59.65)	9.45 (17.90)	7.54 (15.94)	8.50 (16.95)	24.50 (29.67)	19.45 (26.17)	21.98 (27.95)	24.27 (29.51)	20.50 (26.92)	22.39 (28.24)	8.90	10.50	9.70
T_3	Castor oil	80.00 (63.43)	82.40 (65.20)	81.20 (64.30)	5.17 (13.14)	3.71 (11.11)	4.44 (12.16)	21.10 (27.35)	12.11 (20.36)	16.61 (24.05)	18.90 (25.77)	22.75 (28.49)	20.83 (27.15)	9.50	11.75	10.63
T_4	Garlic extract	89.33 (70.93)	91.74 (73.30)	90.54 (72.08)	2.23 (8.59)	0.50 (4.05)	1.37 (6.71)	11.50 (19.82)	5.55 (13.63)	8.53 (16.98)	13.55 (21.60)	27.25 (31.47)	20.40 (26.85)	12.70	13.25	12.98
T_5	Turmeric powder	79.70 (63.22)	82.09 (64.96)	80.90 (64.08)	7.61 (16.01)	5.66 (13.76)	6.64 (14.93)	17.20 (24.50)	14.25 (22.18)	15.73 (23.36)	20.60 (26.99)	22.00 (27.97)	21.30 (27.49)	10.60	11.00	10.80
T_6	Turmeric extract	80.90 (64.09)	83.99 (66.41)	82.45 (65.23)	6.66 (14.96)	5.94 (14.11)	6.30 (14.54)	16.35 (23.85)	15.30 (23.03)	15.83 (23.44)	21.50 (27.62)	23.00 (28.66)	22.25 (28.14)	12.50	11.50	12.00
T_7	Neem oil	86.55 (68.49)	89.05 (70.68)	87.80 (69.56)	2.10 (8.33)	0.00 (0.00)	1.05 (5.88)	14.10 (22.06)	9.89 (18.33)	12.00 (20.26)	16.33 (23.83)	26.50 (30.98)	21.42 (27.57)	10.75	13.50	12.13
T_8	Control	69.93 (56.75)	69.93 (56.75)	69.93 (56.75)	34.49 (35.96)	31.33 (34.04)	32.91 (35.01)	41.32 (40.00)	39.99 (39.23)	40.66 (39.61)	48.60 (44.20)	15.10 (22.87)	31.85 (34.36)	7.00	8.40	7.70
S Em+		2.77	2.86	2.81	0.37	0.35	0.36	0.63	0.52	0.56	0.63	0.84	0.66	0.40	0.41	0.40
C.D. (5%)		8.52	8.80	8.66	1.14	1.08	1.11	1.95	1.60	1.73	1.93	2.57	2.02	1.24	1.26	1.25

seed treatment (20.40%) followed by neem oil (21.42%) compared to control (31.85%). The garlic treated plots also recorded the highest mean yield (12.98 q ha^{-1}).

The best seed treating materials so found were validated through field demonstrations on farmer's field (Table 2). Demonstrations were conducted on fennel at Ajmer, Nagaur (Moulaser) and Jaipur on farmer's field during rabi, 2016-17. Germination per cent in garlic-treated fields, followed by neem oil was found best across the locations. Seed treatment with garlic also reduced powdery mildew wilt and infestation. Yield level was also increased in garlic and neem treated areas compared to untreated control.

Studies conducted at NRCSS, Ajmer revealed that application of garlic extract and neem oil performed better to fetch higher growth, yield and net return of seed spices (Lal, 2018). Attri *et al*. (2020) assessed six plant origin pesticides and cow urine-based bio-resource under *in vitro* and field conditions against *Alternaria solani*. Their investigations revealed that cow urine and *Allium sativum* resulted in 80 per cent growth inhibition at 15 per cent concentration. Koch E and Groot, 2015 tested various plant extracts (ethereal oils) for seed treatment and supported our research.

Education of farmers on proper collection, storage, and preparation of plant-based preparations would help in popularizing plant products to manage pests and diseases of spice and condiment crops (Gahukar, 2011). Sridhar *et al*., 2013 worked on cumin seed-borne diseases management by indigenous technologies and reported that smearing the seeds with castor oil @ 2 litres 25 kg^{-1} of seeds before sowing is helpful in the prevention of wilt disease. He also reported that soaking ten kg seeds in a 5 litre brian solution of 5 percent before sowing increased the crop yield. Tanmay and Biswas,

Table 2: Validation of indigenous seed treating materials against diseases of fennel on farmers fields through FLD during Rabi-2016-17

Name of locations of demonstrations	Treatment	PDI			Yield (q ha^{-1})
		Wilt	Blight	PM	
Nagour (Maulaser)	Garlic extract	08.45	12.75	13.21	11.90
	Neem oil	06.20	14.25	19.22	11.20
	Control	15.90	23.22	33.33	07.00
Ajmer	Garlic extract	13.15	17.20	16.60	10.90
	Neem oil	09.95	22.44	18.90	09.70
	Control	24.90	37.70	36.40	06.40
Jaipur (Bobas)	Garlic extract	02.18	8.88	7.71	13.90
	Neem oil	02.90	11.80	8.13	13.70
	Control	04.60	17.50	14.15	08.40
Jaipur (Jobner)	Garlic extract	03.20	7.10	11.30	14.95
	Neem oil	06.80	12.70	13.75	14.60
	Control	11.11	23.50	27.90	08.90

2018 reported that cow urine is effective against seed-borne fungal species viz., *Aspergillus* sp., *Rhizopus* sp., *Mucor* sp., *Penicillium* sp., *Alternaria* sp., *Macrophomina* sp., and bacterial species viz., *Bacillus subtilis, Pseudomonas* sp., and *Streptococcus* sp. This study confirmed that fresh cow urine was more effective anti-microbial agent than photo-activated cow urine. Panchal et al., 2011 have controlled this disease with garlic extract treatment ; Carbendazim was significantly superior in reducing seedling mortality and also increasing germination percentage in fennel. A case study conducted by Tripathi *et al.* (2020) on traditional methods of insect-pest and plant diseases management in the Bundelkhand region of Madhya Pradesh reported that farmers regularly used cow urine, cow dung, garlic extract, onion extract and different cakes in many crops for controlling seed-borne, soil-borne and leaf diseases and pests and get better results. Jandaik *et al*., 2015 concluded that the cow urine has antifungal activities and the inhibitory activity can be used to control fungi. The nutritional effect of cow urine on plant growth was also tested with *Trigonella foenum-graecum* (fenugreek) and *Abelmoschus esculentus* (okra) plants and the chlorophyll and protein content was also estimated. Extracts from garlic have also been tested by Slusarenko *et al*., 2008 against various fungal and Oomycete diseases. *Allium* species emit a volatile compound, allicin, that has fungicidal properties.

CONCLUSION

The study concluded that the indigenous seed treating materials like garlic extract and neem oil could successfully be used as suitable alternatives to conventionally used chemicals in managing fennel diseases.

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Molecular characterization of the pigeon pea cyst nematode, *Heteroderacajani* Koshy, 1967 from India.

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ABSTRACT

Cysts of *Heteroderacajani*was characterized via molecular analysis using D2-D3 regionof 28S ribosomal DNA (rDNA).BlastN analysis of the amplified fragment in NCBI database revealed that *H.cajani* IARI isolate showed 100% similarity with *H.cajani* isolate 252 (Accession No. DQ328693). The phylogenetic relations of *H. cajani* were evaluated by neighbour joining method. Phylogenetic tree inferred from D2-D3 region of 28S rDNA usingvarious *H.cajani*populations revealedclose relationship of*H. cajani*IARI isolate with *H. cajani* isolate 252.

Keywords: Pigeon pea cyst nematode,*Heteroderacajani*, molecular characterization, phylogeny

Pigeon pea cyst nematode, *Heteroderacajani* is a major pest of pigeon pea (*Cajanus cajan*) and causes significant yield losses in India. The nematode is widespread on sandy loams in Northern India and vertisols of Southern India (Sharma *et al.*, 1992), and now reported in all the major pigeon pea producing states of India *viz*., Andhra Pradesh, Bihar, Gujarat, Haryana, Karnataka, Maharashtra, Punjab, Rajasthan, Tamil Nadu and Uttar Pradesh. It has a wide host range including other leguminous crops, such as green gram, black gram, moth bean, cowpea, cluster bean and horse gram, while sesame is the only non-leguminous host(Koshy and Swarup, 1972; Sharma and Nene, 1985; Jain *et al*., 1994; Balasubramanian and Vadivelu, 2004).

Heteroderacajani is an obligate sedentary endoparasite, causing severe reduction in the growth and yield of pulse crops (Sullivan *et al.*, 2014). In India, the yield loss caused by this nematode on cowpea, pigeon pea and green gram range between 25 to 67.5% even at low populations of 0.2-0.5 larvae/g soil (Sharma et al., 1993a; Sharma and Sethi, 1975; Reddy *et al.*, 1990). In greenhouse studies on pigeon pea, the crop loss was reported to range from 14-24% (Sharma *et al.*,1993b). Besides, it also adversely affects the nitrogen fixation by rhizobia bacteria (Walia and Bajaj, 2013).

This nematode was first reported by Koshy (1967) infesting the pigeon pea crop grown in the fields of IARI, New Delhi. Over theyears, several populations of *H. cajani*, prevalent across India, have been identified based on the morphological characters, isozyme patterns, random amplified polymorphic DNA (RAPD) andamplified fragment length polymorphism (AFLP)analysis. However, Subbotin*et al.* (2010) reported that sequencing of coding and non-coding regions of ribosomal DNA (rDNA) is more authentic and preferred tool for the identification of *Heterodera* species.The present study describes the characterization of the *H. cajani*New Delhi population based on the rDNA and its phylogenetic relationship with other reported populations from India.

MATERIALS AND METHODS

Pigeon pea cyst nematode (*H. cajani*) culture

Soil samples were collected from cowpea (*Vigna unguiculata* cv. Pusa Komal) plants growing in experimental plots of the Division of Nematology, Indian Agricultural Research Institute (IARI), New Delhi, India. Each soil sample was collected from the depth of 15-20 cm.The cysts were extracted from the collected soil samples on a 60mesh sieve using Cobb's sieving and decanting technique. White females of *H. cajani* were picked from the extracted samples and stored at -80 ^{o}C for DNA extraction.

DNA extraction, PCR and sequencing

White cysts of *H. cajani* were transferred to an eppendorf tube containing 20 µl lysis buffer (500mM KCl, 100 mM Tris-Cl pH 8, 15 mM $MgCl_2$, 10mM DTT, 4.5% Tween 20) (Subbotin*et al.*, 2001) and crushed mechanically with a crusher for 3 minutes. Five micro-litre of Proteinase K (20mg/ml) (HiMedia) was added and incubated at 65 ^{o}C for 1h followed by 95 ^{o}C for 10 minutes. After incubation, the tubes were centrifuged for 5 min at 13,000 rpm and the collected supernatant was kept at -20 ^{o}C until further use.

PCR amplification reaction was performed in 25 µl reaction volume containing 2.5 µl 10X PCR buffer (HiMedia),

1.5 µl 50mM MgCl2 (HiMedia), 0.5 µl 10mM dNTPs, 0.5 µl 25 µM each primer, 0.25 µl 5U of Taq polymerase (HiMedia), 18.25 µl sterile distilled water and 1 µl DNA.Two primer sets were used in the PCR reaction. A set of primers D2A (5′-ACAAGTACCGTGAGGGAAAGTTG-3′), D3B(5′-TCGGAAGGAACCAGCTACTA-3′) (De Ley *et al.,* 1999) were used for the identification of *H. cajani*. The PCR amplification conditions consist of 95 ºC for 3min; 35 cycles of 94 ºC for 1 min, 55 ºC for 2 min and 72 ºC for 1 min and followed by a final extension step of 72 ºC for 10 min.Seven micro-litreof amplified PCR product was run on 1% TAE bufferd agarose gel (90V, 45 min.), stained with ethidium bromide. Gel images were captured using Gel Doc system (Alpha Image Analyser, USA). DNA fragments of interest were excised from agarose gels, purified with HiPurA™ PCR product and gel purification combo kit (HiMedia) and send the samples for sequencing. Sequencing of the eluted products was performed by SciGenom Labs, India. The resulted sequence was analysed by using BlastN program in NCBI database.

Phylogenetic analysis

Newly obtained sequence of *H.cajani*, the available sequences of *H. cajani*and outgroup taxa (*Globoderarostochiensis, G. pallida* and*Punctodera punctata*) obtained from GenBank were used for phylogenetic analysis(Table 1). The evolutionary analysis was carried out by using MEGA7 software (Kumar *et al.,* 2016). The evolutionary history was inferred using the Neighbour-Joining method (Saitou and Nei, 1987). The percentage of replicate trees in which the associated taxa clustered together in the bootstrap test (1000 replicates) was shown next to the branches (Felsenstein, 1985). The tree was drawn to scale, with branch lengths in the same units as those of the evolutionary distances used to infer the phylogenetic tree. The evolutionary distances were computed using the p-distance method (Neiand Kumar, 2000) and were in the units of the number of base differences per site.For construction of a phylogenetic tree with respect to newly sequenced *H.cajani* isolate, the analysis involved 15 nucleotide sequences.

RESULTS AND DISCUSSION

The described *H. cajani*isolatewas characterized based on ribosomal sequences and the evolutionary relationship with the known species was tested. PCR amplification of D2-D3 regions of 28S ribosomal region yielded a fragment of 757 bp. The newly obtained sequence was submitted to GenBank database under accession numberKY660057.

Over the years, several authors have described the morphometrics of eggs, second stage juveniles, females, males and cysts of *H. cajani* (Koshy *et al.,* 1971; Walia and Bajaj, 2000; Abdollahi*et al.,* 2006, 2007; Subbotin*et al.,* 2010). Singh *et al.* (1998) used the isozyme patterns of esterases and malate dehydrogenases for the identification of two races of *H. cajani*. Abdollahi and Ganguly (2009) evaluated the genetic variation among Indian populations of *H. cajani* by RAPD, while Rao *et al.* (2011) reported cluster analysis of different populations using AFLP analysis. BLASTN analysis in NCBI showed that the nucleotide sequence was closely related to 28S rRNA gene of *H. cajani* isolate 252 from India (Accession No. DQ328693) with 100% identity. Phylogenetic relationship of the studied Indian isolate of *H. cajani* and the available sequences of *H. cajani* from GenBank are presented in Fig. 1. The optimal tree with the sum of branch length was 0.16.

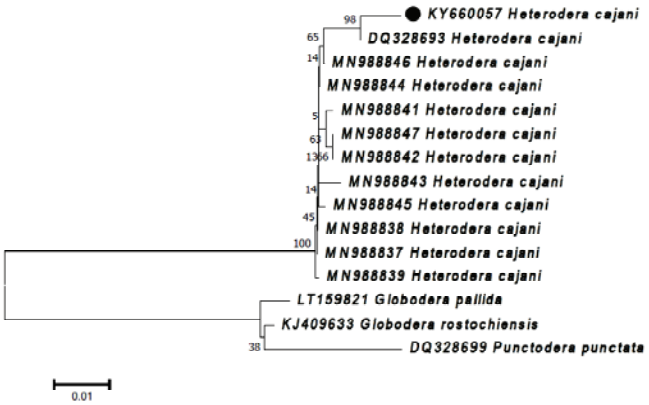

Fig. 1. Phylogenetic relationship of *H. cajani*based on D2-D3 region of 28S rDNA sequences using the Neighbor-Joining method. Numbers indicated on nodes are bootstrap values for each cluster based on 1000 replications. Bullet indicates the sequence produced in the study.

Table 1. Details of Heteroderacajani populations and outgroup taxa used in the present study for phylogenetic analysis

S. No.	Species	Isolate/Strain/Clone	GenBank accession number
1	*H. cajani*	Hc1	KY660057
2	*H. cajani*	252	DQ328693
3	*H. cajani*	Pasur	MN988847
4	*H. cajani*	Akola	MN988846
5	*H. cajani*	Delhi	MN988845
6	*H. cajani*	Hisar	MN988844
7	*H. cajani*	Bhiwani	MN988843
8	*H. cajani*	Gulbarga	MN988842
9	*H. cajani*	Coimbatore	MN988841
10	*H. cajani*	Indore	MN988839
11	*H. cajani*	Kanpur	MN988838
12	*H. cajani*	Hyderabad	MN988837
13	*G. rostochiensis*	GRRO40	KJ409633
14	*G. pallida*	28_34Gp	LT159821
15	*P. punctata*	318	DQ328699

In recent years, it has become a standard to study the plant-parasitic nematodes based on molecular data. Analysis of more Indian populations would be of value,especially, to correlate them based onthe differences with respect to the host-range involving other legume crops. This will providestrong scientific basis not only for discriminating populations to check their further distribution,but also help in designing future management programmes.

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Utilization of sugar syrup waste of aonla processing industry for flavored spicy beverages

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ABSTRACT

The article describes in detail the process of utilizing sugar syrup left over as waste in the aonla processing industries in preparing value added products like the mint flavored spiced squash and ginger-lemon flavored spiced ready-to-serve (RTS) drinks. The spicy squash was prepared using Indian spices with and without mint that can be consumed after diluting it with water whenever required. Both the beverages were stored at room and low temperature for nine months and evaluated for different chemical and sensory attributes at every three month storage intervals. Squash without mint and RTS drink stored at low temperature was organoleptically more acceptable even after nine months. Both the drinks showed good market potential during summer months. The technology may prove helpful in saving economic loss of aonla processing industry in the form of sugar syrup waste.

Key Words: Aonla, squash, RTS drink, spice, phenolics

Aonla (*Emblica officinalis* Gaertn) is an important minor fruit crop of India. Fruit has high nutritional as well as medicinal value due to fair amounts of fibre, vitamin-C and phenolics present in it. India produces huge quantum of aonla fruits. According to an estimate of National Horticulture Board, India produced 1046 thousand metric tonnes of aonla from 92 thousand hectares of cultivated land area during 2018-19. Uttar Pradesh and Himanchal Pradesh are the chief aonla growing states in India, though it is found in almost all parts of India. Aonla fruit, owing to its highly acidic-astringent taste, is not suitable for fresh consumption. Aonla fruits are processed into a number of food products like preserve, jam, jelly, candy, toffee, pickle, sauce, squash, juice, RTS beverage, cider, shreds, dried powder, etc. (Athawale and Akbari, 2017). Certain ayurvedic preparations like *Chyavanprash, Triphala, Ashokarishta,* and *Triphalamasin* are also made from aonla fruits (Jat *et. al*, 1988).

Aonla processing cluster located in eastern districts of Uttar Pradesh particularly Pratapgarh, Faizabad, Varanasi, etc., is known as aonla belt of Uttar Pradesh. Preserve and candy are the most popular products of aonla apart from pickle and juice. During preparation of aonla preserve and candy, a large amount of sugar syrup is left as by-product. According to Kocher *et al*. (2013), aonla fruit while being processed into candy by food processing industry generates a high brix candy syrup which otherwise is a waste and poses burden to environment. The syrup is turbid dark in colour as it undergoes repeated heating in order to raise the concentration of sugar at successive steps. It contains high amount of sugar, acid, phenolics and other compounds. The left-over syrup poses serious unhygienic condition coupled with problem of waste disposal. The sugar syrup is generally thrown away as waste material since it is not re-utilized or sold as animal feed supplement at very nominal price. Due to unavailability of suitable technologies for re-utilization of sugar syrup and, disposal of syrup as waste causes economic loss to aonla processing industry. Therefore, technologies for re-utilization of waste sugar syrup in the production of some value added products is highly needed. Trials run to prepare fermented beverages like wine, cider and vinegar through alcoholic/acetic acid fermentation were not successful due to presence of fair amount of preservative in the syrup, which inhibited growth of microbes responsible for fermentation. Therefore, attempts have been made to utilize this syrup waste to develop suitable non-fermented beverages that can well accommodate its sweetness, acidity and astringency with good acceptability in terms of sensory attributes.

MATERIAL AND METHODS

Sugar syrup waste collected from big aonla processing units of Pratapgarh, Uttar Pradesh was used for preparing spiced squash and ready-to-serve (RTS) drink. Two types of squashes one with mint ($Mint^+$) and the other without mint (Control), using Indian spices were prepared. Squashes were prepared by soaking desired quantities of spice materials including cumin and black pepper powders with mint and without mint powder in small amount of water. The mixture was left for overnight and then filtered through muslin cloth with gentle squeezing to obtain spice extract. The sugar syrup was added with common salt and black salt and heated till a temperature of 80 ^oC is achieved. After cooling sugar syrup

mixture, spice extract was added to it. A mixture of citric acid, potassium metabisulphite (KMS) and sodium benzoate, dissolved separately in little amounts of water, were added to the squash. The amount of citric acid added was standardized on the basis of taste of sugar-acid blend of the mixture, which may vary depending upon acidity of batch of sugar syrup. The product was filled in clean, dry, pre-sterilized glass bottles and sealed with crown corks. It was stored under ambient conditions at dry, cool place. The product was served in 1 : 4 dilution with chilled water.

The ready-to-serve (RTS) flavored drink was prepared using lemon and ginger along with spices and other ingredients. The sugar syrup was diluted to bring the total soluble solid (TSS) content to around 14 ^{o}B and added with required quantity of citric acid to adjust the acidity to around 0.3 percent. Ginger and black pepper powders were boiled in small amount of diluted syrup and filtered through muslin cloth with gentle squeezing to obtain spice extract. The spice extract along with lemon juice and salts was added to diluted syrup and the mixture was heated till a temperature of 95 ^{o}C is reached. Then, KMS was added as preservative and mixed well. The drink was filled in pre-sterilized glass bottles and sealed with crown corks. The filled bottles were then sterilized by keeping them in boiling water for 20 minutes. After cooling, the product was stored at low temperature or LT (10±2 ^{o}C) and room temperature or RT (20-30 ^{o}C).

The products so prepared were stored for nine months and analyzed at regular intervals of every three months. Microbiological quality of the squash and RTS drink was done as per the method of Speck (1985). Total soluble solids (TSS) was recorded in terms of degree brix using hand refractometer (Erma, Japan). Acidity, vitamin-C and total phenolics were determined as per the methods described by Ranganna (2000). Acidity of the samples was estimated in terms of citric acid by titration of sample solution against 0.1N NaOH solution using phenolphthalein as indicator. Vitamin-C in drinks was measured by titrating sample solutions against 2, 6-dichloro phenol indophenol dye solution. The total phenolics content was estimated spectrophotometrically using method evolved by Folin and Ciocalteu (1927). Non-enzymatic browning (NEB) was measured by recording optical density values of ethanol extracts of the samples at 440 nm wavelength using spectrophotometer. The sensory evaluation of squash and RTS drink was carried out by a panel of semi-skilled judges on composite scoring basis using hedonic scale as described by Amerine *et al*. (1965). The judges were given coded samples for scoring on the basis of colour, aroma and taste. Chilled RTS drink was served for direct consumption while squash was served after diluting it with 4 times of chilled water.

RESULTS AND DISCUSSION

Microbial examination of squash revealed no fungal or bacterial growth in any of the samples at any stage of storage. At zero day, TSS of control and mint^{+} squash samples were 63 and 65 ^{O}B, respectively, while titratable acidity were 2.91 and 3.06 per cent, respectively. The values of TSS and acidity remained almost unchanged during storage. Similarly, initial TSS of RTS drink was 13.8 ^{O}B and acidity 0.32 per cent, which remained same till the end of storage period at both LT and RT. The vitamin-C contents of control and Mint^{+} squashes were 42.2 and 44.4 mg 100 ml^{-1}, respectively (Table 1). A regular decrease in the vitamin-C content was observed after 9 months, both samples had 28.9 mg 100 ml^{-1} values. In case of RTS drink, initial vitamin-C content was 15.1 mg/100 ml in both samples (Table 2) which decreased regularly during storage. However, values remained same till 6 months. Thereafter, vitamin-C loss in RT sample was little higher than LT sample. The final retention values of vitamin-C in RT and LT samples were 0.9 and 1.3 mg 100 ml^{-1}, respectively. Bhattacherjee *et al.* (2011) also reported loss in vitamin-C content of aonla juice during storge. According to Sethi *et al*. (1980), oxidation of ascorbic acid into dehydroascorbic acid by oxygen during storage is responsible for reduction of vitamin-C. Addition of mint in the squash, resulted into increased level of phenolics (768 mg 100 ml^{-1}) as compared to control (627 mg 100 ml^{-1}). Total phenolics decreased continuously during storage and after 9 months levels came down to 454 and 538 mg/100 ml in controlt and Mint^{+} squashes (Table 1). Reduction in phenolics of RTS drinks (120.9 mg 100 ml^{-1}) stored at LT and RT was more or less parallel during storage. LT stored samples retained little more phenolics (67.2 mg 100 ml^{-1}) than samples stored at RT (64.9 mg 100 ml^{-1}) at the end of storage (Table 2). Break down of phenolic compounds during storage is responsible for decrease in these compounds. Gradual decline in polyphenol contents in sand pear and pear-apple juice beverage during storage was also reported by Raj *et al*. (2011). Addition of mint caused darkening of samples as revealed by higher optical density values (absorbance at 440 nm) of alcohol extracts of samples. The absorbance reading of Mint^{+} sample was 0.198 against 0.124 of control (Table 1). With the storage time, absorbance values increased continuously in both samples. After 9 months, absorbance of Mint^{+} sample reached to 0.351 while it was 0.287 in case of control. Breakdown of chlorophyll into some other colour compounds might be responsible for more rapid darkening of Mint^{+} squash. Increasing trend was also observed in RTS drinks. However, it was very slow in LT samples (Table 2). RT samples turned much dark after 9 months of storage, which might be due to non-enzymatic browning of the samples. Bharte and Bharte (2014) working

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on citrus juice reported that 2,5-Dimethyl-4-hydroxy-3(2H)-furanone (DMHF) is one of the promising marker formed during browning process.According to Jain and Khurdiya (2009) reaction between sugars and amino acids (Maillard reaction) might be responsible for browning. Shinoda *et al.* (2005) proposed that formation of furfural might result into browning of orange juice. Sensory evaluation of spiced squash revealed that Mint$^+$ sample (score 8.4/9.0) was liked more than control (score 8.1/9.0) sample (Table 1). Pleasant aroma of mint leaves led to enhanced acceptability of drink. However, in later stages of storage, its score went down might be due to loss in aroma coupled with development of some other compounds imparting slight bitterness. Control sample, though also displayed declining acceptability trend, it retained sensory qualities in better way. After 9 months of storage, control scored higher of 7.6 against 6.7 of Mint$^+$ sample. Among RTS drinks, LT samples obtained better score throughout the experiment (Table 2), might be due to light appearance and better retention of organoleptic qualities. RT samples turned darker with time coupled with more loss in nutritive qualities of drink as compared to LT samples.

Table 1: Changes in chemical and sensory attributes of mint flavoured spicy aonla squash prepared from sugar syrup waste during storage

Parameter	Treatments	0 day	3 month	6 month	9 month
Vitamin-C	Control	42.2	39.6	34.4	28.9
(mg 100 ml^{-1})	Mint$^+$	44.4	41.8	35.9	28.9
Total phenolics	Control	627	594	513	454
(mg 100 ml^{-1})	Mint$^+$	768	735	616	538
Non-enzymatic	Control	0.124	0.223	0.245	0.287
browning	Mint$^+$	0.198	0.284	0.318	0.351
(OD at 440 nm)					
Sensory score	Control	8.1	8.1	8.0	7.6
(Out of 9)	Mint$^+$	8.4	7.8	7.5	6.7

Table 2: Changes in chemical and sensory attributes of ginger-lemon flavored spicy aonla RTS drink prepared from sugar syrup waste during storage

Parameter	Treatments	0 day	3 month	6 month	9 month
Vitamin-C	RT	15.1	4.2	1.6	0.9
(mg 100 ml^{-1})	LT	15.1	4.2	1.6	1.3
Total phenolics	RT	120.9	111.4	73.9	64.9
(mg 100 ml^{-1})	LT	120.9	114.7	72.0	67.2
Non-enzymatic	RT	0.009	0.037	0.054	0.078
browning	LT	0.009	0.014	0.021	0.024
(OD at 440 nm)					
Sensory score	RT	7.4	6.8	6.6	6.2
(Out of 9)	LT	7.4	7.4	7.3	6.9

CONCLUSION

The study concluded that the sugar syrup waste of aonla processing units could successfully be utilized in preparing good quality of squash and RTS drink by adding spices and flavoring materials. It could be stored for 9 months without appreciable deterioration in nutritive and sensory qualities. Spiced squash without mint flavor was more suitable for long storage than squash with mint. However, lemon-ginger flavored spicy RTS drink stored at low temperature was more preferred than that stored at room temperature.

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Association of nutritional, cooking quality and yield traits in F_1 and F_2 population of rice (*Oryza sativa* L.)

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ABSTRACT

The magnitude of association between nutritional, cooking quality, yield and its attributing characters in nutrient rich rice genotype crosses studied during *Kharif* and *Rabi* seasons grain yield per plant exhibited significant correlations between the number of grains per plant, test weight, kernel length and L/B ratio in both F_1 and F_2 generations, respectively. The association analysis with cooking and nutritional quality traits indicated existence of a significant positive association between kernel length and volume expansion ratio and also with kernel elongation ratio. The study further confirmed that there is significant correlation between gelatinization temperature and cooking quality traits besides amylase and protein contents. Zinc and iron contents were negatively correlated with cooking quality traits *viz.,* kernel length, volume expansion ratio, kernel elongation ratio and with amylase content. The correlation between zinc and iron content is significantly positive.

Key words: *Rice, L/B ratio, amylase, protein, zinc and iron*

Rice is the most important staple food crop for more than half of the population in the world. The slogan 'Rice is life' is the most appropriate for India, as this crop is a livelihood for millions of households in India. Selection for yield *per se* is not reliable and indirect selection for yield component traits play an important role. Hence, studies on character association not only help to understand the nature of physical linkage but also provide information on the nature and direction of association existing between the traits. The efficiency of selection for yield mainly depends on the direction and magnitude of association between yield and its component characters and also among themselves. Character association provides information on the nature and extent of association between pairs of metric traits and helps in selection for the improvement of the character. The knowledge regarding relative contribution of individual traits to yield may be accomplished by correlation studies.

If the association of characters is due to manifold effects of a gene or genes, it is difficult to separate these characters by selecting a particular character so related. If the correlation is due to linkage it is possible to reverse the correlation, provided the linkage is not very close. Hence, it is important to establish genetic basis of correlations before launching any breeding programme. Further, the component characters of yield exhibit different associations among themselves and also with yield. Unfavorable associations between the desired attributes under selection may limit genetic advance. Therefore, the magnitude of association between quality, yield and its attributing characters, an essential parameter for planning sound breeding programme, was studied.

MATERIALS AND METHODS

The investigation was carried out at the Regional Agricultural Research Station, Warangal, (Professor Jayashankar Telangana State Agricultural University) located at an altitude of 304 M above MSL, 17.97° N latitude and 79.60° E longitude during 2 years 2014-15 and 2015-16 utilizing both *Kharif* and *Rabi* crop seasons in each year. The main crop seasons in Telangana State can be called as rainy (June-Dec) and post rainy (Nov-April) seasons. Entire study was conducted as two experiments. The experimental material comprised of 10 parents *viz.,* MTU 1010, WGL-32100, Ramappa, RP-Bio-5478-270, RP-Bio-5478-166, RP-Bio-5478-176, DRR Dhan-40, RP-Bio-5478-185, NH-686, NH-787, their 45 F_1 hybrids (generated from ten parents, crossed in diallel fashion without reciprocals during previous *Kharif*, 2014) and corresponding F_2's (seed obtained from F_1 generation raised during successive *Rabi*, 2014-15) and two promising check varieties *viz.* BPT 5204 and Chittimuthyalu. Sufficient seed of fresh crosses was preserved and half was sown to produce F_2 seed.

During *Kharif* 2014, ten parents were transplanted each in four rows in a crossing block at spacing of 20 x 15 cm and 4 sets were maintained. Crosses were effected in a 10 x 10 half diallel design to produce 45 F_1s. Selected parents were sown at staggered intervals of seven days to facilitate continuous availability of pollen during crossing. Twenty eight days old seedlings were transplanted at spacing of 30 x 15 cm. Hybridization was done by clipping method of emasculation as suggested by Jennings *et al*. (1981). Spikelets were emasculated in the afternoon and pollination was done

in morning of next day between 11 – 1.00 P.M. Parents were crossed in a half diallel fashion without reciprocals to generate 45 F_1 s hybrids. Adequate care was taken to produce sufficient seed required for studying different generations.

After the crosses were effected during *kharif*, 2014, in the next *rabi* 2014-15 itself, all the 45 F_1s without reciprocals along with parents and two check varieties were grown for producing sufficient F_2 seed. The material (Parents, F_1s and F_2s) was planted in randomized block design replicated thrice during final season *Kharif,* 2015 in a separate plot for studying combining ability, heterosis and inbreeding depression. Parents, hybrids and check varieties were planted in one row of 3.0 m length adopting a spacing of 20 cm between the row and 15 cm between the plants within a row with single seedling per hill. Each entry of F_2 was planted with same spacing maintaining 120 plants for cross in each replication (8 rows).

Five parent plants and F_1s, twenty from BC_1, BC_2 and forty from F_2 were tagged at random for each entry in each replication. The analysis for all quantitative data and quality data was done as per the standard procedures of IRRI. The mean data was used for final statistical analysis using Indostat to study the association between nutritional and cooking quality traits in rice.

RESULTS AND DISCUSSION

In the present investigation, simple correlation studies were estimated for yield and its components for F_1 and F_2 generation, whereas, grain quality traits *viz.,* kernel length, volume expansion ratio, kernel elongation ratio, gelatinization temperature, amylase content, protein content, iron and zinc contents in F_1 generation were recorded. The results are furnished in Table 1 & 2.

The data on days to 50 per cent flowering exhibited significant negative association in F_1 and F_2 generations with grain yield per plant as was reported by Chandrasekhar Haradari and Shailaja Hittalmani (2017). The days to 50 per cent flowering recorded a significant negative correlation with test weight (-0.3177, -0.3579), kernel L/b ratio (-0.2986, -0.6160) in F_1 and F_2 generations, respectively. Significant positive association was observed between this trait with total and productive tillers per plant in F_2 generation. (Table 1). The plant height recorded a significant negative correlation with grain yield as was earlier reported by Ratna *et al*. (2015). In F_1 generation, this character had non significant negative association with tillers per plant, number of grains per panicle, test weight, kernel length and kernel L/B ratio while non significant positive association with panicle length, spikelet fertility percent, kernel breadth.

Table 1. Simple correlations coefficients among grain yield, its components and grain quality in F_1 and F_2 generations

Character	Genera-tion	Days to 50 per cent flower-ing	Plant height (cm)	Total no. of tillers plant^{-1}	No. of produc-tive Tillers/ plant	Panicle length (cm)	No .of grains panicle^{-1}	SPF (%)	Test Weight (g)	Kernel length (cm)	Kernel breadth (cm)	L/B ratio (cm)	Grain yield plant^{-1} (g)
Days to 50 per cent flowering	F_1	1.0000	0.2006	0.0518	0.0416	-0.0341	-0.4008	-0.0871	-0.3177*	-0.2899	0.0194	-0.2986*	-0.4893**
	F_2	1.0000	0.3027*	0.3637*	0.3571*	0.1763	-0.3960*	0.1754	-0.3579*	-0.6710**	0.0497	-0.6160**	-0.4787**
Plant height (cm)	F_1		1.0000	-0.1568	-0.2215	0.2982	-0.2809	0.0410	-0.2932	-0.2006	0.0238	-0.2017	-0.3083*
	F_2		1.0000	-0.1092	-0.1306	0.0718	-0.3903**	0.1256	-0.4175**	-0.3548**	-0.0387	-0.2947*	-0.4170**
Total no. of tillers	F_1			1.0000	0.9125**	0.0602	-0.0273	-0.1839	-0.1452	-0.3660*	-0.0748	-0.3365*	-0.0258
	F_2			1.0000	0.5014**	-0.0662	-0.0693	0.1014	-0.1763	-0.3012*	-0.1756	-0.1994	-0.1073
No. of productive tillers	F_1				1.0000	0.0064	-0.0126	-0.1553	-0.0549	-0.1769	-0.0401	-0.1729	0.0726
	F_2				1.0000	-0.0706	-0.0932	0.2060	-0.0404	-0.3567*	-0.1490	-0.2427	-0.0330
Panicle length (cm)	F1					1.0000	0.0275	-0.0901	0.0687	-0.2766	0.3630*	-0.3796*	0.0475
	F_2					1.0000	0.1058	-0.0383	0.0894	-0.1453	0.3296*	-0.2991*	0.1001
No .of grains/plant	F_1						1.0000	0.1615	0.7305**	0.4368**	0.2214	0.3244*	0.8461**
	F_2						1.0000	-0.0502	0.6196**	0.3648*	0.0350	0.2946*	0.7618**
SPF (%)	F1							1.0000	0.1200	-0.0894	-0.0419	-0.0709	0.0784
	F_2							1.0000	-0.2410	-0.1989	0.0407	-0.1946	-0.1048
Test Weight (g)	F_1								1.0000	0.5440**	0.0620	0.4931**	0.7708**
	F_2								1.0000	0.4515**	-0.0487	0.4271**	0.6436**
Kernel length (cm)	F_1									1.0000	0.0291	0.9366**	0.6141**
	F_2									1.0000	-0.0006	0.8815**	0.5447**
Kernel breadth (cm)	F1										1.0000	-0.3156*	0.2379
	F_2										1.0000	-0.4629**	0.0336
L/B ratio (cm)	F_1											1.0000	0.4968**
	F_2											1.0000	0.4655**

* Significant at 5 per cent level; ** Significant at 1 per cent level

Table 2. Simple correlations coefficients among quality components in F_1 generation

Character	Kernel length (mm)	Volume expansion ratio	Kernel elongation ratio	Amylose content (%)	Protein content (%)	Iron content (%)	Zinc content (%)	Gelatinization temperature
Kernel length (mm)	1.0000	0.8560**	0.8154**	0.7948**	0.3727*	-0.2985*	-0.6264**	0.6351
Volume expansion ratio		1.0000	0.7644**	0.7007**	0.3010*	-0.2543	-0.5387**	0.5577
Kernel elongation ratio			1.0000	0.7930**	0.4705**	-0.3962**	-0.5516**	0.3917
Amylose content (%)				1.0000	0.4539**	-0.1759	-0.5257**	0.4502
Protein content (%)					1.0000	-0.0672	-0.0850	0.2008
Iron content (%)						1.0000	0.4281**	-0.0988
Zinc content (%)							1.0000	-0.5289

* Significant at 5 per cent level; ** Significant at 1 per cent level

Plant height had negative significant association with number of grains per panicle, test weight, kernel length and kernel L/b ratio in F_2 generation.

For total number of tillers per plant, correlations were found to be non significant negative association with grain yield, number of grains per panicle, test weight, kernel length, kernel breadth, kernel L/b ratio in both generations. This trait had significant positive association with number of productive tillers per plant in F_1 and F_2 generations, respectively.

The trait, productive tillers per plant, had positive correlation with yield per plant in F_1 generation while negative correlation in F_2 generation. This trait registered non significant negative association with number of grains per panicle, spikelet fertility percent, test weight, kernel length, kernel breadth, and kernel L/B ratio in F_1 and F_2 generation while in positive association with pikelet fertility percent. Positive association between number of productive tillers per plant with grain yield per plant earlier reported by Ashok *et al*. (2016), Edukondalu *et al.* (2017), Manjunatha *et al*. (2017) and Srikanth Thippani *et al.* (2017). Number of effective tillers plant^{-1} had negative significant correlation with panicle length in F_2 as was reported by reported by Ratna *et al*. (2015). The panicle length recorded significant positive association with kernel breadth (0.3630/0.3296) and positive association with grain yield, number of grains per panicle and test weight at both generations. It had negative association with spikelet fertility percent and kernel length. But, significant positive association of panicle length with grain yield plant^{-1} earlier reported by Vinoth et *al*. (2016) and Edukondalu *et al.* (2017) Rukmini Devi *et al*. (2017). The character number of grains per panicle registered significant positive correlation with grain yield, test weight, kernel length, kernel length, kernel L/B ratio in two generations. (0.3407, 1.2713) and significant positive phenotypic association with yield (0.4703). Significant positive association of number of grains per panicle with grain yield per plant earlier reported by Vinoth et *al.* (2016), Ashok *et al*. (2016), Dhurai *et al*. (2016) and Chandrashekhar Haradari and Shailaja Hittalmani (2017). Spikelet fertility percent had positive correlation was observed with grain yield per plant (0.0784) in F_1 generation while negative correlation in F_2 generation. It had non significant negative correlation with kernel length, kernel L/B ratio in both generations.

Test weight had significant positive association with yield per plant (0.7708, 0.6436), kernel length (0.5440, 0.4515) and kernel l/b ratio (0.4931, 0.4271) in both generations. Significant positive correlation of test weight with grain yield per plant reported by Ashok *et al*. (2016) and Rukmini Devi *et al*. (2017). Negative correlation of this trait with number of productive tillers/plant in F_1 and F_2 generations. These results are in accordance with the results of Moosavi *et al.* (2015). Grain yield per plant exhibited significant correlations with number of grains per plant, test weight, kernel length and L/B ratio in both F_1 and F_2 generations respectively. Productive tillers per plant exhibited non significant positive correlation in F_1 while negative in F_2. (Table 1). Rukmini Devi *et al.* (2017) reported the positive association of grain yield per plant with filled grains per panicle and effective tillers.

Kernel length had significant positive association with kernel L/b ratio (0.9366, 0.8815) (Vijay Kumar, 2015) and grain yield per plant (0.6141/0.5447) (Meena *et al.* (2016) and Srikanth Tippani *et al*. (2017) in two generations and it had negative association with kernel breadth in F_2 generation.(-0.0006). Similar result was reported with grain breadth by Patel *et al.* (2014). Kernel breadth had a non significant positive correlation with grain yield per plant, (Madhavi Latha *et al.* 2005). Kernel breadth had negatively significant association with kernel L/b ratio (-0.3156, -0.4629) irrespective of generations where similar result were reported by Meena *et al*. (2016), Suman Rawte and Ritu R Saxena (2017). Kernel L/B ratio registered significant positive correlation with grain yield (0.4968/0.4655) in F_1 and F_2 generation as was reported by Madhavilatha *et al.* (2005).

The study on simple correlation suggested that selection of plants with more number of productive tillers

per plant, number of grains per panicle and test weight, which had significant positive association with yield, may be taken in to account in rice breeding program for yield improvement. In addition, kernel length, L/B ratio also had significant positive correlation with grain yield per plant.

Relationship among nutritional and cooking quality traits

The overall results of correlation analysis with cooking and nutritional quality traits indicated that a significant positive association existed between kernel length and volume expansion ratio and also with kernel elongation ratio. Hence, these three traits are interrelated positively. Mahmuda Khatun *et al*., (2003) reported a significant correlation between kernel length and kernel length after cooking. There existed significant positive correlation between amylose content and other quality traits *viz*., kernel length, volume expansion ratio, kernel elongation ratio and protein content. Further, amylose content has significant positive association with zinc content. In similar way, protein content also possessed a significant positive association with kernel length, volume expansion ratio, kernel elongation ratio and also the amylase content. However, its association with iron and zinc content was negative but not significantly. It is interesting to note that both zinc, iron contents were negatively correlation with cooking quality traits viz., kernel length, volume expansion ratio, kernel elongation ratio and with amylase content. The correlation between zinc, iron content is significantly positive, this indicates that it is possible to develop pure lines or hybrids with high iron and zinc content simultaneously. (Table 2)

The negative association between micronutrients (iron and zinc) with important cooking quality traits like volume expansion ratio, kernel elongation ratio is quite disappointing to note that it is difficult to develop best cooking quality traits with high iron and zinc contents. The study further indicated that higher protein content and moderate to high amylase content is required for developing rice with best volume expansion ratio and kernel elongation ratio. The study also confirmed that there is significant correlation between gelatinization temperature and cooking quality traits besides amylase and protein contents. EL-Hissewy *et al*., (1992) has also reported significant positive correlation of gelatinization temperature and amylase content with kernel elongation after cooking and cooking time which is in agreement with our findings. They also reported positive association between protein content and kernel expansion which is in support of our present findings. Oko *et al*., (2012) also reported a significant positive correlation between gelatinization temperature, kernel length after cooking and amylase content, though not at significant level. They also reported a negative correlation between amylase content and kernel elongation which is contrary to our present study.

CONCLUSION

The study conclusively suggested that selection of plants with more number of productive tillers per plant, number of grains per panicle and test weight having significant positive association with yield may be used in the rice breeding programs. Kernel length, L/B ratio also had significant positive correlation with grain yield per plant, where as the zinc and iron contents were negatively correlated with cooking quality traits viz., kernel length, volume expansion ratio, kernel elongation ratio and amylase content.

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Black soybean (*Glycine max* (L.) Merrill): Effect of soaking, boiling and germination on its nutritional quality

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ABSTRACT

Black soybean (*Bhatt*) (*Glycine max* (L.) Merrill) is one major crop of Uttarakhand state. Besides nutraceutical potential, of this crop plays a vital role in ensuring nutritional and livelihood security of rural population of hills. In the present research, effect of various processing techniques i.e. soaking, boiling and germination on the nutritional properties of black soybean flour was studied. Based on the findings, it can be concluded that all the three processing methods are significant in improving the nutritional quality of black soybean. Germination was found to be the most valuable technique for the efficient utilization of black soybeans which makes them rich in mineral content, dietary fibre and *in-vitro* nutrient availability with less content of antinutritional factors.

Keywords: Black soybean, processing, germination, oligosaccharides, *in-vitro* protein and iron bioavailability

Soybean (*Glycine max* (L.) Merrill) is an important legume crop belonging to botanical family, *Leguminosae* and the subfamily *Papilionideae*. It an ideal crop for improving nutrition, food security, sustainable crop and livestock production systems. Black soybean, locally known as *Bhatt/ Bhatmash/kala bhatt*, produced in 5734 hectare area of Uttarakhand **(Hipparagi *et al.*, 2017)** is considered as a "treasure house of medicinal and nutritional properties". It reduces cholesterol level, inhibit growth of cancerous cells and have protective effect against a number of fungal, viral and chronic diseases. It has high protein, crude fibre and crude fat content.

Black soybean is grown in Garhwal and Kumaon hills of Uttarakhand as a pulse crop. The only processing treatments given to black soybean include overnight soaking of grains and roasting. As per the local people of Uttarakhand, soaking is done to lessen the time of cooking and roasting is done to prepare a snack called "*bhujwa*", which is commonly consumed during winters. Bhatt is traditionally prepared in an iron kadhai to make "*churkani*" and "*dubka*".

Various studies have reported that nutritional quality of legumes can be improved using different processing treatments (**Joshi and Varma, 2016)**. Black soybean is rich in protein and minerals but their bioavailability is reduced due to the presence of antinutritional factors viz trypsin inhibitors, phytates, tannins, oxalates and oligosaccharides. Some of processing techniques like pressure cooking, roasting, boiling, fermentation and germination have been reported to reduce the antinutritional factors to a considerable levels **.**

Despite of having numerous nutritional health benefits, black soybean is consumed on a very limited scale due to the lack of information on its nutritional quality as well as the effect of various processing treatments. Therefore, present study was undertaken to assess the impact of processing treatments like soaking, boiling and germination on the proximate composition, mineral composition, antinutrients and antioxidant activity of black soybean.

MATERIALS ANS METHODS

Duration of the study

The present study was conducted in the year 2017-18.

Procurement of raw materials

Black soybean (VL-63 variety) was procured from Tarai Development Corporation (TDC), Haldi, District U.S. Nagar (Uttarakhand).The seeds of black soybean were cleaned manually to remove foreign materials (dust, dirt, grit and other impurities), broken and immature soybeans.

Flour development

Flour was prepared using three processing treatments i.e. soaking, germination and boiling (Fig. 1).

Nutritional composition of black soybean flours

Proximate composition

All four flour samples obtained from black soybean seeds were analyzed for moisture, crude protein, crude fibre, ash and crude fat contents using the standard procedures given by **AOAC (1995)**. The crude fat was estimated in Sox

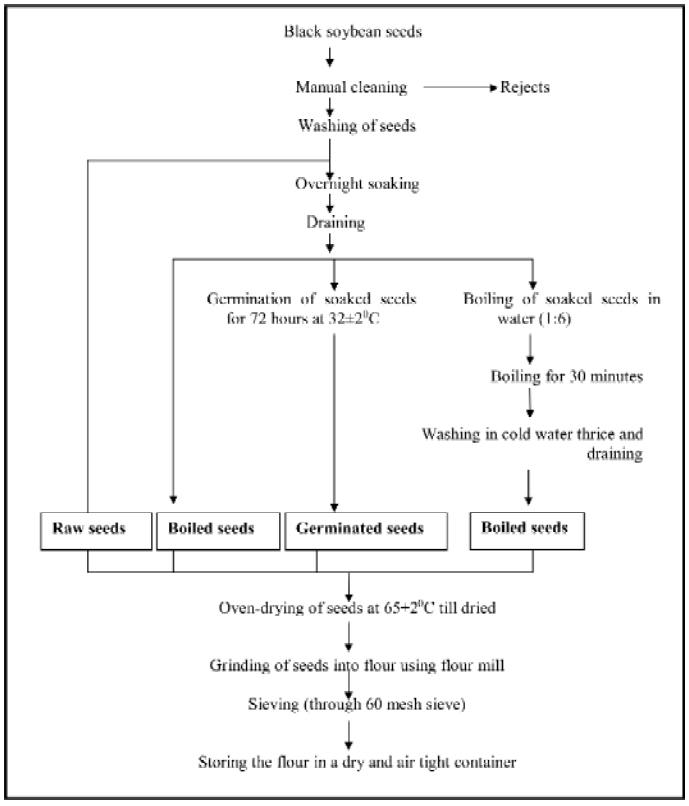

Fig. 1. Flow chart for developing black soybean flour by different techniques

Plus SPS 20 (Pelican Bioinnovations Pvt. Ltd) and the crude protein was estimated by Kjeldahl method. Carbohydrate content was determined by difference. Physiological energy was calculated in kcal per 100g using the formula:

(9 x % fat) + (4 x % protein) + (4 x % carbohydrate)

Minerals estimation

Calcium, iron, zinc, copper and magnesium in the black soybean flour samples were analyzed by Atomic Absorption Spectrophotometry using A7040 SensAA spectrophotometer. The ash solution for mineral estimations was prepared using wet-ashing procedure given by Raghuramulu *et al.* (2003).

Antinutritional factors

The antinutritional factors analyzed in the present research included trypsin inhibitor activity, tannins, phytate, oxalate and oligosaccharids. Trypsin inhibitor activity (TIA) was also determined (Fig. 2) using benzoyl-DL-arginine-p-nitroanilide (BAPNA) as substrate. Tannins were estimated. Phytate and oxalates were measured. The oligosaccharides viz raffinose and stachyose were estimated in the black

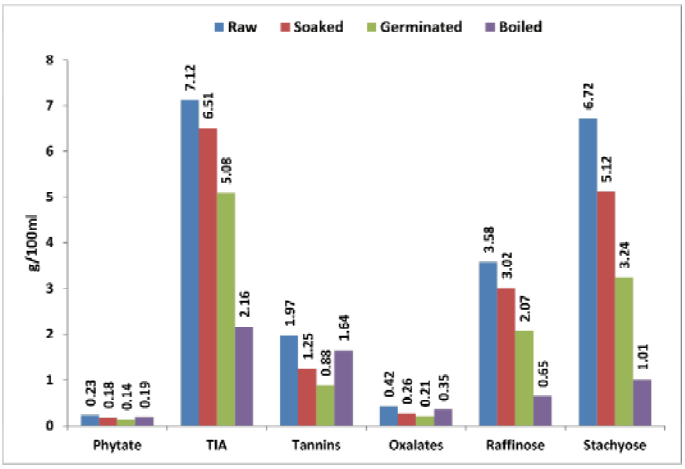

Fig. 2 Antinutritional content of black soybean flours (Dry weight basis)

soybean flour sample by the procedure given by **Tanaka *et al.* (1975)**.

Other dietary parameters

Dietary fibre

Total dietary fibre content was estimated using method of **Asp *et al.* (1983)** which included gelatinization and enzymatic digestion of flour followed by filtration, washing and titrimetric estimation of dietary fibre, respectively.

***In-vitro* protein digestibility**

In-vitro protein digestibility was analyzed using the procedure given by **Akeson and Stahman (1964)** where protein was extracted by method of **Degroot and Stump (1961)**. The digested protein of the black soybean flour samples was calculated by subtracting residual protein from total protein of the sample and protein digestibility was calculated using the formula:

$$\text{Protein digestibility (\%)} = \frac{\text{Digested protein}}{\text{Total protein}} \times 100$$

***In-vitro* iron bioavailability**

In-vitro iron bioavailability of black soybean flours was determined by the method given by **Rao and Prabhavati (1978)**, which is based on the release of the ionisable iron from food subjected to treatment with pepsin–HCl at pH 1.35 and subsequent adjustment of pH to 7.5, respectively. Ionizable iron was determined in the filtrate by the á-á-dipyridyl method.

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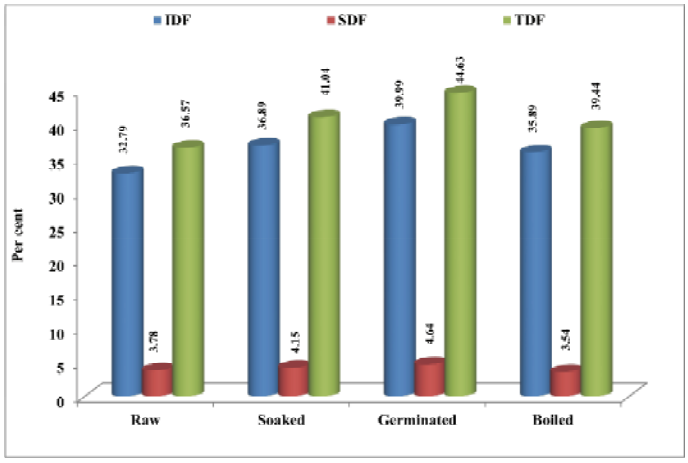

Fig. 3 Dietary fibre content of black soybean flours (Dry weight basis)

DPPH radical scavenging activity and total antioxidant capacity (TAC)

DPPH radical scavenging activity of black soybean flours was estimated by the 1, 1-diphenyl-2-picryl-hydrazyl (DPPH) method of **Brand-William *et al.* (1995).** Further, total antioxidant capacity (TAC) was estimated in terms of milligram of trolox equivalent/ 100 g sample.

Statistical analysis

All the experiments were carried out in triplicates. All the quantitative data was computed in terms of mean and standard deviation. Data were subjected to one-way ANOVA (F-test) to determine significance of difference among different flours of black soybean with respect to different attributes. Critical difference (CD) was calculated at 5% level of significance where p-value of less than 0.05 indicated the presence of significant differences.

RESULTS AND DISCUSSION

Proximate composition

The findings of proximate composition of raw, soaked, germinated and boiled black soybean flours are presented in table 1. Moisture content of a product represents the total amount of water present in the product either as free water or bound to gluten, fibre and gums. In the present study, moisture content varied from 8.52-10.16% with highest value in germinated black soybean flour to lowest in raw black soybean flour. The moisture content of soaked as well as germinated black soybean flour was found significantly higher (pd"0.05) than the moisture content of raw black soybean flour. This finding is similar to the results reported by **Dobhal and Raghuvanshi (2018)** showed that higher hydration capacity permits the black soybean seeds to absorb more amount of water leading to high moisture content (Table 1, Fig. 3).

The findings on crude fat content showed a significant difference (pd"0.05) between raw (19.25%), germinated (17%) and boiled (15.92%) black soybean flour. Decrease in fat content of legume flour due to germination has also been reported in various studies. The decrease in the crude fat content could be due to the fact that the pericarp and seed coat of black soybean which are mainly composed of the fatty component got affected with processing.

Table 1. Proximate composition of raw and processed black soybean flours (dry weight basis)

Parameter	Raw	Soaked	Germinated	Boiled	CD at 5%	f-value
Moisture (%)	8.52±0.08^{a}	9.64±0.05^{b}	10.16±0.63^{c}	9.46±0.25^{b}	0.35	30.07*
Crude fat (%)	19.25±0.82^{a}	17.95±0.46^{ab}	17.00±1.71^{b}	15.92±0.58^{c}	1.91	14.73*
Crude protein (%)	43.62±1.04^{a}	44.87±0.77^{a}	47.27±0.29^{b}	46.27±0.24^{b}	1.27	39.10*
Total ash (%)	5.96±0.45^{a}	5.70±0.48^{a}	7.40±0.53^{b}	5.49±0.10^{a}	0.80	23.46*
Crude fibre (%)	5.28±0.17^{a}	5.76±0.25^{b}	8.23±0.09^{c}	4.97±0.09^{d}	0.31	26.03*
Carbohydrate by difference (%)	25.89±0.26^{a}	25.72±1.36^{a}	20.10±2.00^{b}	27.35±0.51^{a}	1.34	18.77*
Physiological energy (kcal)	451.29±3.94^{a}	443.91±1.88^{a}	422.48±9.01^{b}	437.76±4.90^{a}	1.48	15.55*

*Different alphabets in superscript in each row show significant difference between values

Table 2. Mineral composition of black soybean flours (mg/100g) (dry weight basis)

Mineral	Raw	Soaked	Germinated	Boiled	CD at 5%	f-value
Calcium	268.79±1.72^{a}	316.99±1.99^{b}	336.19±1.28^{bc}	310.69±1.50^{bd}	1.34	18.80*
Iron	9.84±1.56^{a}	11.43±1.32^{a}	15.58±1.12^{b}	11.05±0.55^{a}	1.25	12.99*
Magnesium	264.54±1.36^{a}	281.65±2.79^{b}	290.42±2.97^{c}	274.04±0.39^{d}	1.05	78.35*
Zinc	4.79±0.24^{a}	5.18±0.09^{b}	5.42±0.10^{bc}	5.75±0.11^{d}	0.28	22.47*
Copper	2.35±0.03^{a}	2.29±0.10^{a}	2.51±0.13^{a}	2.49±0.15^{a}	0.21	2.78^{NS}

Different alphabets in superscript in each row show significant difference between values

Proteins carry out a wide range of functions essential for sustenance of life and thus become one of the most of the important nutrients required by the body. In the present study, highest crude protein content was found in germinated black soybean flour as 47.27% followed by boiled, soaked and raw black soybean flour as 46.27, 44.87 and 43.62%, respectively. The reason for the increase in protein content during germination could be the mobilization of stored nitrogen to produce nutritionally high-quality proteins needed by the sprouts for their growth. Contrary to our results, **Raghuvanshi *et al.* (2011)** reported no change in the crude protein, crude fibre, ash and crude fat content of mung bean after 24-hours germination.

Total ash provides a measure of the total amount of minerals within a food. Ash content in the present study varied from 5.49 to 7.4% with significantly higher ash content in germinated black soybean flour. Soaking and boiling didn't show any significant increase in the ash content of raw black soybean flour.

Crude fibre content in the present study varied between 4.97 to 8.23% with highest content in germinated and lowest in boiled black soybean flour. All four flours were found significantly different (pd"0.05) from each other.

As depicted in table 1, carbohydrate content varied from 20.10 to 27.35% with highest content in boiled black soybean flour to lowest in germinated black soybean flour. The decrease in carbohydrate content may be due to the active respiration process during germination, leading to the breakdown of number of carbohydrate molecules in order to allow increased protein synthesis. Physiological energy of black soybean flours in the present study ranged from 422 (germinated) to 451 kcal/100g (raw), which is higher than the value reported by **Longvah *et al.* (2017)** for yellow soybean.

Mineral composition

The findings on mineral composition (table 2) showed that all the three processing techniques viz soaking, boiling and germination led to significant increase in calcium, iron, magnesium and zinc content of raw black soybean flour. Germination increased the zinc content of black soybean by 13%. The copper content of flours in the present study varied from 2.29 to 2.51mg/100 g with a non-significant difference between all processing techniques (Table 2).

Antinutritional factors

Antinutritional factors are defined as "natural or synthetic compounds that interfere with the absorption and metabolism of nutrients". Trypsin inhibitors are low molecular weight, heat-labile proteins which inhibit the activity of trypsin and chymotrypsin in the gut and interfere with the digestibility of dietary proteins and reduce their utilization **(Gopalan *et al.*, 2007).**

The findings of the present study (fig. 2) showed that all the four flours of black soybean were significantly different to each other with lowest activity in boiled flour and highest activity of trypsin inhibitors in raw flour. TIA was reduced by 8.6% with soaking, 28.68% with germination and markedly reduced by 69.7% with boiling. Soaking, germination and boiling increase the permeability of cell membrane and thus, increase the amount of anti-nutrient leaching. Similar findings with respect to decrease in the trypsin inhibitor activity with soaking and germination has been reported in various studies (**Ramakrishna *et al.*, 2006).**

Germination, soaking and boiling, all the three treatments showed significant reduction in trypsin inhibitor activity, oxalate, phytate and tannin content of the raw sample. This finding of the present study is in accordance to the findings of **Chandrashekhraiah (2013)** and **Gawad (2016).** Decrease in oxalates may be due to activation of oxalate oxidase enzyme during processing which breaks down oxalic acid into carbon dioxide and hydrogen peroxide consequently releasing calcium **(Murugkar *et al.*, 2013).**

Oligosaccharides i.e. raffinose, stachyose and verbascose are thought be the major producers of flatulence in soybean leading to its reduced utilization as human foods. Processing viz soaking, germination and boiling led to significant reduction in raffinose content by 15.64, 42.18 and 81.8%, respectively. Similarly, stachyose content was also reduced significantly in soaked (23.81%), germinated (51.78%) and boiled (84.97%). The decrease in the level of raffinose and stachyose during boiling might be attributed

Table 3. Other nutritional parameters in black soybean flour (dry weight basis)

Nutritional parameter	Raw	Soaked	Germinated	Boiled	CD at 5%	f-value
In-vitro protein digestibility (%)	48.02±1.52^{a}	56.61±1.64^{b}	60.72±2.01^{c}	54.74±1.11^{bd}	1.59	34.31*
In-vitro iron bioavailability (%)	8.64±1.35^{a}	31.28±1.37^{b}	44.65±2.09^{c}	21.98±1.42^{d}	1.80	19.35*
DPPH radical scavenging activity (%)	65.52±0.99^{a}	40.76±0.56^{b}	52.77±0.51^{c}	31.20±0.32^{d}	0.94	135.6*
TAC (mg TE/100 g)	294.06±1.39^{a}	182.98±2.13^{b}	228.02±0.77^{c}	95.93±0.24^{d}	1.10	430.6*

Different alphabets in superscript in each row show significant difference between values

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to hydrolysis of these oligosaccharides into disaccharides and monosaccharides or to the formation of other compounds which are subsequently leached out.

Other dietary parameters

Dietary fibre

Insoluble dietary fibre (IDF) and soluble dietary fibre (SDF) in the present study ranged from 32.79-39.9% and 3.54-4.64%, respectively (fig. 3). The total dietary fibre (TDF) content in the present study ranged from 36.57 to 44.63% with highest value in germinated black soybean flour followed by soaked (41.04%), boiled (39.44%) and raw black soybean flour (36.57%), respectively.

***In-vitro* protein digestibility**

Combination of traditional food processing and preparation practices can improve the nutritional profile of soybean as well as increase the bioavailability of protein and micronutrients. In the present study, all the three processing treatments i.e. soaking, germination and boiling lead to significant increase in *in-vitro* protein digestibility (IVPD) of raw black soybean flour (Table 3). Soaking increased IVPD by 17.89%, germination by 26.45% and boiling increased IVPD by 13.9%.

Sprouting increases protein digestibility of pulses by increasing metabolism of seed proteins and catabolism of antinutritional factors including protease inhibitors, phytate, polyphenols etc.

***In-vitro* iron bioavailability**

The *in-vitro* iron bioavailability of black soybean flour samples in the present study ranged from 8.64-44.65% with highest value in germinated flour followed by soaked (31.28%), boiled (21.98%) and raw black soybean flour (8.64%), respectively, with significant (pd"0.05) differences among all flours. The findings of present study is in accordance to the observations of **Nakitto *et al.* (2015)** who showed that processing techniques viz soaking, roasting and germination lead to significant increase in the *in-vitro* iron bioavailability of mung bean, common bean and yellow soybean, respectively.

DPPH radical scavenging activity and total antioxidant capacity (TAC)

Soybeans are considered to be an intact source of phytochemicals like á-tocopherol, isoflavones, flavonoids and anthocyanins, possessing high biological activity. In the present study, highest DPPH radical scavenging activity was recorded in raw black soybean flour (65.52%), followed by germinated (52.77%), soaked (40.76%) and boiled (31.2%) black soybean flours, respectively (table 3). Observations related to decrease in free radical scavenging activity of soaked, germinated and boiled soybean.

Total antioxidant activity (TAC) of raw, soaked, germinated and boiled black soybean flour in the present study was 294, 182.98, 228.02 and 95.93TE mg/100g. reported TAC of yellow soybean as 5246.81µmol TE/100g using ORAC assay. They also reported that pressure cooking of soybean for 10 minutes without soaking led to decrease of TAC by 34.8% (Table 3).

CONCLUSION

Black soybean (*Bhatt*) is one major Kharif crop of Uttarakhand state. In the present research, effect of various processing techniques i.e. soaking, boiling and germination on the nutritional properties of black soybean flour was studied. Based on the findings, it can be concluded that all the three processing methods are significant in improving the nutritional quality of black soybean with germination to be the most effective method in utilization of black soybeans which makes them rich in mineral content, dietary fibre and *in-vitro* nutrient availability with less content of antinutritional factors. It is recommended to include in the take-home-rations (THR) supplied in the ICDS centres to reduce PEM among children in India.

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