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Twelfth Annual Day Function of "Journal of Eco-friendly Agriculture"

On the completion of 12 years of successful publication, 12th Annual Day Function of Journal of Eco-friendly Agriculture was organized by Doctor's Krishi Evam Bagawni Vikas Sanstha (Doctor's Agricultural and Horticultural Development Society), Lucknow on 8th August, 2017, in collaboration with ICAR-Indian Institute of Sugarcane Research, Lucknow. Dr. A.D. Pathak, Director, ICAR-Indian Institute of Sugarcane Research, Dilkusha, Lucknow, graced the occasion as chief guest and Dr. M.D. Pathak, Chairman, CRIWMI, Lucknow and Former Director, Training and Research, IRRI, Manila, Philippines was the Guest of Honour.

During the occasion an Annual Key Note Address was delivered by Dr. Manu Dutt Pathak, Chairman, Centre for Research and Development of Waste & Marginal Land, Lucknow, former DG, UP Council of Agricultural Research and former Director, Training and Research, IRRI, Manila, Philippines on the topic "Bio-rejuvenation of sodic waste land by a chemical free, ecofriendly and cheap technology". He shared his experience about conversion of waste land without chemical with ecofriendly approach, which can save large amount of state exchequer.

In his 15 years of research experience in sodic waste land at two locations, i.e. Pipersand near Lucknow and village Kursi in Barabanki District, where he conducted experiments in waste land, finally able to convert both farms from waste land to productive areas. He is so much interested in the subject that he purchased the above sodic waste land farms for this experiment. He informed this while delivering his key note address. In his deliberation, he emphasized that soil sodicity has been a major worldwide problem and especially in Uttar Pradesh limiting agricultural production. Most of the earlier work carried out to ameliorate these soils primarily included chemical measures. Since, these chemicals are expensive, governments have been subsidizing it. For the first time Dr. M.D. Pathak showed that an environment-friendly and inexpensive approach to tackle this problem.

In this technology first sodic soils with irrigation facility, if left undisturbed, get reclaimed by itself, provided no cattle grazing is done or removal of vegetation from the area takes place. Secondly, to exploit the use tolerant or resistant crops for soil sodicity. There are many tolerant varieties of different crops, which can be grown on these soils and in this process the land is reclaimed without any extra cost.

In the first exploratory experiment, 300 germplasm of 22 different crops were sown after minimum soil tillage. One germplasm of each, barley, linseed, mustard and rice crops could germinate normal with normal growth and yield, while others either failed to germinate or their seedlings did not survive. Subsequently 4,194 germplasm of 21 different crops were evaluated for their germination and survival at the three different levels of soil sodicity. Several crop lines were selected which were tolerant to sodicity and could grow normally.

Four years after starting these experiments, the entire 15-acre farm became fully fertile without any symptoms of soil sodicity and its soil pH dropped to normal sodicity levels, which was repeated in other farm.

The approach will lead to a major saving to the government and farmers, increase agricultural production substantially and not cause environmental contamination.

His findings were published in the "Journal of Ecofriendly Agriculture" which is published by Doctor's Krishi Evam Bagawni Vikas Sanstha (Doctor's Agricultural and Horticultural Development Society), Lucknow.

During the function on 8th August at ICAR-Indian Institute of Sugarcane Research, Lucknow, the recent issue was released and Dr. M.D. Pathak was honoured with a shawl and memento by chief guest Dr. A.D. Pathak, Director, Sugarcane Research Institute, Lucknow. Chief Guest and Guest of Honour were also honoured by society.

It is relevant to mention that Dr. MD Pathak is an internationally recognised scientist, known for his rice variety IR 20, which was released from IRRI, Phillipines for multiple resistance againsed two different species of leaf hopper and plant hopper pests, besides tungro and grassy stunt viruses, bacterial leaf blight, bacterial leaf streak diseases and resistance to several other insect pests and diseases and also recorded as tolerant to iron toxicity and to iron, phosphorus and zinc deficiencies. This variety occupied the largest area in the world and Dr. Pathak was awarded Borlaug award during 1973 and the first prize of the Research Accomplishment Award of the International Year of Rice, 2004.

The function was attended by large number of scientists from different institutes and universities of Lucknow.

Glimpses of 12th Annual Day Function of Journal of Eco-friendly Agriculture

Sodic soil waste land prior to start of the experiment in 1994.

The undisturbed plots developed thick weed cover and good crop in a period of 3 years.

Normal germination and plant growth of spinach, garlic and mustard varieties in sodic land.

Performance of different pigeon pea genotypes in soil pH 9.4-9.9.

Honouring guest of honour Dr. M.D. Pathak by Dr. A.K. Mishra, Chief Editor of the Society

Dr. A.D. Pathak, chief guest of the function honouring Dr. M.D. Pathak with a memento and shawl.

Release of recent issue of Journal of Ecofriendly Agriculture.

Honouring chief guest Dr. A.D. Pathak by Dr. Rajeev Dutta, Vice Prisident of the Society

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Review article

Microbial consortium in biological control: An explicit example of teamwork below ground

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ABSTRACT

Biocontrol strategy has been approved as the most acceptable and sustainable approach to moderate the crop losses due to phytopathogens and pests. Though the screening of biocontrol agents (BCA) is done meticulously to select the best among the lot, still a single strain proves inept to fight the numerous opponents present below ground, around the plant system. Under natural conditions, microbes do live in harmony supporting two to several different genera together utilizing the available nutrients and thereby creating a team of beneficial cluster acting against their negative counterparts. This team of helpful microbes acting as a team to protect the plants from the pathogens is termed as "consortia". Though there exist certain parameters to be kept in mind before designing an effective consortium against a particular target pathogen in a definite habitat of the host plant. Microbes tolerant to environmental shock with longer shelf life and sustainability, possessing higher enzymatic activity with higher rate of metabolism along with being non-pathogenic to the host plant should be preferred which should incur lower cost of mass multiplication. Though the mechanisms of action remain the same as that for a single biocontrol agent, but in consortium the synergism between the microbes is the most essential character that calls for the higher rate of success in field, comprising different genera of BCA. The present review overviews the studies that have been carried out reporting the successful effects of applying microbial consortia against different phytopathogens and pests along with giving a brief account of the mechanisms undertaken by the BCAs to combat the same.

Key words: Biocontrol, microbial consortia, synergism, phytopathogen

Towards the increasing inclination for a safer and sustainable approach against the use of chemicals, biocontrol strategy has taken a surge for managing phytopathogens proving effective management to crop losses for different plant families. Biological control has already proved to be feasible alternative to chemicals and therefore various eco-friendly products containing wide genera of microbes have been successfully commercialized (Punja and Utkhede, 2003). Another attractive aspect of utilizing the beneficial microbes for pest and pathogen management is the sustainability of the strategy which aids in improving the prevalent homeostasis in the environment along with providing a healthy and acceptable commodity quality (Anonymous, 1987, 1989). The success for this strategy lies in the synergism between the microbial community of different genera working together to protect the host plant from the diverse abiotic and biotic stresses (Saxena *et al*., 2013). Also, it largely depends on the size of the active populations of such microbes maintained at the site of action for their enhanced performance (Savazzini *et al*., 2009).

PGPR (plant growth promoting rhizobacteria) and PGPF (plant growth promoting fungi), the beneficial microbes have also been shown effective in enhancing plant growth of numerous agronomic crops including legumes, cereals *etc*. (Guo *et al*., 2004; Saxena *et al*., 2014). Increasing the accessibility of nutrients to the host plant thereby reducing the ethylene level within the plant along with augmented production of stimulatory compounds like plant growth regulators have been few of the important strategies utilized by the group of beneficial microbes to restrict the growth of phytopathogens (Whipps, 2001). The basic mechanisms employed by the BCAs include production of anti fungal compounds and competing with the target pathogen for space and nutrients (Vinale *et al*., 2008; Keswani *et al*., 2014).

An appropriate and calculated quantity of everything is beneficial, while the excess of the same cause detrimental effect also (Fig. 1). Similar is the case with the BCAs as well, significant larger populations of a specific BCAs at the target site may pose danger for the native non-target species thereby resulting in alternation of the prevalent micro habitat of host plant (Singh *et al*., 2013). Care must be taken into account while preparing microbial formulation to avoid possibilities of competitive displacement and toxicity to non-target microorganisms existing naturally in environment (Arora *et al*., 2008). Apart from screening an efficient BCA to target the host pathogen ecosystem, development of economic and efficient delivery systems for successful applications of selected BCAs on the field in another very important

component of the biocontrol technology (Bashan, 1998). Delivery systems like dusts, starch granules, extruded granules, alginate pellets and seed coatings have been utilized for preparing formulations of the screened BCAs in order to manage pests and pathogens which can be prepared under laboratory conditions (Gasic and Tanovic, 2013).

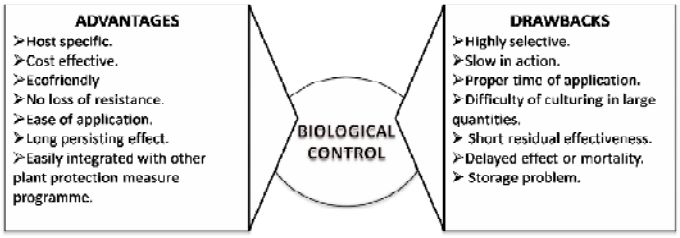

Fig. 1. Positive and negative prospects of biological control

Biological control in plant disease management

Biological control is regarded as an eco-friendly and sustainable approach to disentangle the prevalent scenario in crop disease management along with the aim of quality crop production. Originally, biological control was referred to as 'natural control' wherein the pests and pathogens were controlled by employing their natural enemies or antagonist in order to manage or eradicate their populations (Baker and Cook, 1983). But as the term is vast including the action of all environmental factors either biotic or abiotic, so biological control was regarded as a phase of natural control. According to Garret 'Biological control of plant disease may be precisely defined as any condition or practice whereby survival or activity of a pathogen is reduced through the agency of any living organism with the result that there is reduction in incidence of the disease caused by the pathogen'. However, Cook (1988) defined biological control as 'use of natural or modified organisms, genes, or gene products to reduce the effects of pests and diseases'. Wilson (1997) again defined biological control as 'The control of a plant disease with a natural biological process or the product of a natural biological process'.

According to Sharma *et al.* (2009), two basic strategies could be employed to apply the antagonist, one by utilizing the anti-microbial property of the already existing microbes in the field or another by introducing the efficient strains artificially. In plant pathology, biological control refers to the suppression of pathogenic microbes apart from controlling the weed populations by utilizing the microbial antagonists both under field and green house conditions, generally being referred to as BCAs. Application of BCAs proves to be beneficial in maintaining plant health and hygiene along with enhancing the crop yield (Singh, 2006) (Fig. 2). Combinations of several agents with numerous antimicrobial activities give a better possibility of functionality of at least one of the released bio-agents. PGPR is a broad category belonging to diverse genera and most of them comprise of different species of *Pseudomonas*, *Bacillus*, *Rhizobium*, *Trichoderma* and *Serratia*. The fungal genera *Trichoderma* explicit multifarious applications in various fields besides biological control (Keswani *et al.*, 2014).

Application of single antagonistic strain often results in inconsistent control. The best way to overcome such hindrance is to combine the application of different microbes in a single unit. Such combinations may result in more extensive colonization of the rhizosphere along with expression of defense responses in varying ecosystems (Bashan, 1998). The successful outcome of such combinations has resulted in development of formulations involving microbial consortium (Duffy *et al.*, 1996). Multiple organisms boost the potential and constancy of control through numerous mechanisms, and finally provide a better stability over wide range of environmental conditions (Pandey and Maheshwari, 2007).

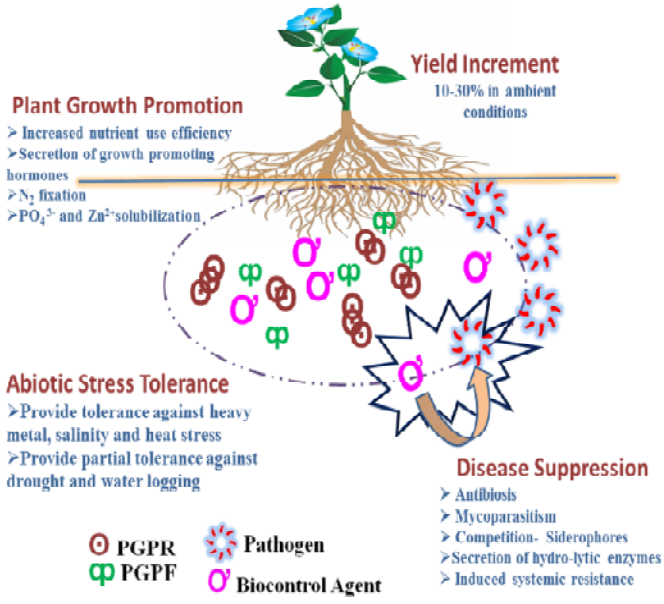

Fig. 2. Beneficial effects of agriculturally important microorganisms

Microbial consortium

Microbial consortium may be stated as a group of different microorganisms that have the ability to act together in a community. The rhizosphere provides residence to a large number of microbial populations, which imparts various functions to plants that may be beneficial or detrimental to their growth (Jain *et al.*, 2012). In general, the

beneficial microbes tend to live in diverse communities and get bound to the root surface. The cross talk between the plant and microbe render them to work and live as a community, thus exhibiting excellent symbiosis (Koornneef and Pieterse, 2008). The basic idea behind using microbial consortium is that a single BCA may not prove to be active under various soil conditions or against all the pathogens attacking a particular host plant, thus combination of microbes may prove to be more relevant in the long run to provide better and quick results (Bashan, 1998). Thus, the focus on the use of multiple microorganisms has gained momentum and lead to the development of microbial consortium. The use of microbial consortium not only facilitates disease suppression but also have a positive impact on plant growth promotion (de Boer *et al.*, 2003). The prime advantage of microbial consortium is that if by any chance one microbe fails to show its potential, then it is presumed that the other one will show its activity to its full extent. Moreover, application of microbes in a consortium improves consistency, efficiency and reliability of microbes under different soil conditions (Stockwell *et al.*, 2011). In several studies conducted across the globe, it has been revealed that use of various combinations of bioagents provided a higher level of protection compared to sole application (Dunne *et al.*, 1998). The different mixtures of biocontrol agents are used to control numerous plant diseases caused by bacteria and fungi. Previous studies of combinations of biological agents for plant diseases have included mixtures of fungi (de Boer *et al.*, 1997), mixtures of fungi and bacteria (Leeman *et al.*, 1996) and mixtures of bacteria (de Boer *et al.*, 2003).

Need for microbial consortium

The constant use of chemicals for decades has leaded a negative impact on the environment. The growing concern regarding the ecosystem has advocated the farmers to shift management practices towards biological control. Biological control proved to be an efficient management practice for a sufficient period till the targeted organism developed resistance against a particular bioagent (Pal and Gardner, 2006). These hindrances have forced the scientists to opt for microbial consortium in which more than one bioagent was used. The success of consortium have fascinated scientists to develop different types of consortium in which different organisms were used *i.e.,* fungi and bacteria, bacteria and actinomycetes, bacteria and mycorrhiza *etc* (Fig.3). Thus, microbial consortium has proved to be an efficient tool of biological control, which has being widely used for controlling serious diseases of crop plants (Pandey and Maheshwari 2007 a, b; de Jansen *et al.*, 2002; Anjaiah *et al.*, 2003).

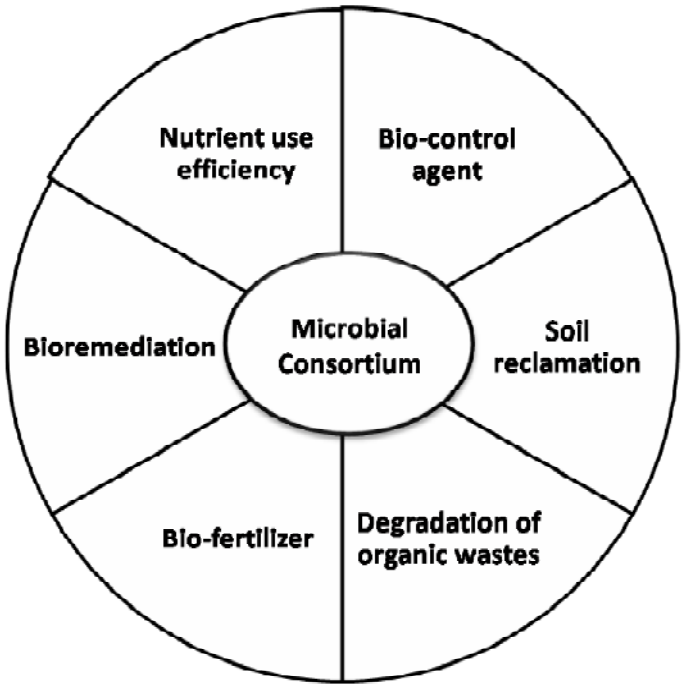

Fig. 3. Various applications of microbial consortium

Prerequisite for preparing microbial consortium

Development of microbial consortium demands certain characteristics from the constituent microorganisms. The microbes should be selected which are compatible with each other and are resistant to environment fluctuations (Singh *et al.*, 2012). Moreover, they must be fast acting, easy to handle and store, non pathogenic, longer shelf life and have high efficacy. The production of consortium should be cheap and economical. Most of the reported biological agents have lead to provide high levels of disease suppression compared with the individual antagonists (Dandurand and Knudsan, 1993).

Types of microbial consortium

Consortium comprising fungi and bacterial strains

Combinations of fungal and bacterial strains are being widely used for preparation of microbial consortium. *Trichoderma* and *Pseudomonas* strains are the most popular and frequently studied bioagents for developing consortium as they have been reported to suppress disease incidence along with imparting growth promotion in different crops (Raja *et al.*, 2013; Yadav *et al.*, 2017). It is reported that arbuscular mycorrhiza are usually an important source of carbon and have the ability to provide energy source to the rhizospheric bacteria like *P. fluorescens* for biocontrol. It was reported that a bacterial strain *P. fluorescens* BBc6 which could

synergistically combine with mycorrhizal strains and improve the efficiency of *Laccaria bicolor* S238N.

Combined application of *T. viride* and *P. fluorescens* reduces incidence of sheath blight disease compared to control (Mathivanan *et al*., 2005). Dandurand and Knudsen (1993) reported that incidence of root rot of pea caused by *Aphanomyces euteiches* f.sp. *pisi* was considerably reduced on application of combined treatment of *T. harzianum* and *P. fluorescens* strain 2-79RN10 compared to single *T. harzianum* treatment. *Rhizobium* and *Glomus intraradices*, when applied together demonstrated an adverse effect on the pathogens by increasing availability of N and P to plants and ultimately depriving it for the pathogens. Raja *et al*. (2013) recorded highest height of chilli plant with *T. harzianum* + *T. viride* and followed by *T. harzianum* + *P. fluorescens*. Application of *P. putida* WCS358 and *F. oxysporum* strain Fo47 resulted in suppression of Fusarium wilt of carnation and flax. Yadav *et al*. (2017) observed that *Trichoderma viride*, *T. asperellum* and *T. harzianum* alone individually gave inhibition of 37.8, 30.9 and 25.7 per cent. When it is mixed with *Pseudomonas fluorescens,* it gave inhibition of 23.5 per cent. This combination also gave inhibition of growth of *Fusarium oxysporum*, the chick pea wilt.

Similarly, *F. oxysporum* f. sp. *radicis-lycopersici* can be effectively controlled by application of fluorescent *Pseudomonas* sp. with a nonpathogenic *F. oxysporum* thereby making the pathogen deprive of C and Fe (Lemanceau *et al.* 1993). A successful control of *F. oxysporum* f. sp. *cucumerinum* was achieved by combined effect of *P. putida* with saprophytic strains of *F. oxysporum* (Park *et al*., 1998). *Bacillus mycoides* and *Pichia guilermondii* altogether demonstrated a successful control of *Botrytis cinerea* infection on strawberry (Guetsky *et al*., 2001).

Consortium comprising only bacterial strains

The application of rhizospheric bacteria such as *Bacillus, Pseudomonas* and *Streptomyces* spp. has been reported to control several important plant diseases in numerous crops (Chung *et al*., 2005; Hass and Defago, 2005). There are various examples of management of plant diseases with combination of different bacteria or different strains of same bacterium. Muthukumar *et al.* (2010) reported that treatment of chilli seeds with endophytic strains of *P. fluorescens* in combination (EBC 5 and EBC 6) resulted in lower incidence of pre- and post emergence damping-off caused by *Pythium aphanidermatum*. The combined application of *Paenibacillus* and *Streptomyces* sp. suppresses Fusarium wilt of cucumber. In addition to it, combination of *Stenotrophomonas maltophila* and *P. fluorescens* improves protection against Pythium-mediated damping-off in sugar beet compared to their single application (Dunne *et al*., 1998).

A consortia of *Sinorhizobium meliloti* PP3 and *Burkholderia* sp. MSSP and have been reported to promote growth of pigeon pea due to their ability to enhance IAA production and solubilise phosphate (Pandey and Maheshwari 2007 a, b). Similarly, *P. fluorescens* NBRI-N and *P. fluorescens* NBRI-N6 in a consortium controlled collar rot in betelvine caused by *S. rolfsii* (Singh *et al*., 2003). The dry root rot of bean caused by *F. solani* f. sp. *phaseoli* has found to be controlled successfully by application of *B. subtilis* MBI600 and *Rhizobium tropici* with considerable increase in yield (de Jansen *et al*., 2002).

A mixture of *B. amyloliquefaciens* strain IN937a and *B. pumilus* strain IN937b successfully inhibit soil and air borne pathogens such as *Ralstonia solanacearum*, *R. solani*, *S. rolfsii*, and cucumber mosaic virus (Jetiyanon *et al*., 2003). The mixture triggered a set of defense enzymes leading to significant disease reduction in four plant pathosystems, *viz*., tomato with *S. rolfsii* and *Ralstonia solanacearum* and pepper with *S. rolfsii* and *C. gloeosporioides*. It is reported that a cluster of bacteria producing antibiotics and chitinase have the ability to suppress sheath blight disease of rice caused by *R. solani* compared to a single microbial treatment. Similarly, combined application of *P. fluorescens*, *Sinorhizobium fredii* and *Azobacter chroococcum* reduces incidence of fusarium wilt of *Cajanus cajan* (Choure and Dubey, 2012).

Studies conducted on microbial consortium

The use of microbial consortium may provide defense against a particular pathogen or a series of pathogens attacking a host plant. Apart from providing defense against the pathogens, they also facilitate plant growth promotion. The microbial consortium may either comprise of only bacterial strains or a mixture of fungal and bacterial strains. These may be further categorized into dual, triple or quadruple based on number of microbes present in the consortium. In literature, there are various examples citing use of microbial consortium for disease control and plant growth promotion.

M. incognita and *M. phaseolina* are responsible for causing root rot disease in chickpea, and is considered as a serious problem which limits the production of the crop (Siddiqui and Husain, 1992). However, it is reported that the disease can be minimized to a considerable extent by applying a consortium of *Pseudomonas striata*, *Rhizobium* sp. and *Glomus intraradices*. Application of consortium has resulted in an increase in root colonization and reduction in nematode multiplication rate was found compared to individual application. The increased availability of

nutrients such as P and N quenched by *Rhizobium* and *P. striata*, has been regarded to play a negative role towards growth and development of the nematode (Pant *et al.*, 1983).

The successful application of consortium comprising of *Bacillus*, *Pseudomonas* and *Streptomyces* sp. have been documented for management of several plant diseases in numerous crops (Chung *et al.*, 2005; Hass and Defago, 2005). The dry root rot of bean caused by *F. solani* f. sp. *phaseoli* has found to be managed successfully by application of *B. subtilis* MBI600 and *Rhizobium tropici* with considerable increase in yield (de Jansen *et al.*, 2002). Similarly, consortia of *Sinorhizobium meliloti* PP3 and *Burkholderia* sp. MSSP have been reported to promote growth of pigeon pea due to their ability to enhance IAA production and solubilise phosphate (Pandey and Maheshwari, 2007 a, b). Sung and Chung (1997) reported that a cluster of bacteria producing antibiotics and chitinase have the ability to suppress sheath blight disease of rice caused by *R. solani* compared to a single microbe. *P. fluorescens*, *Sinorhizobium fredii* and *Azobacter chroococcum* when applied together, reduces the incidence of fusarium wilt of *Cajanus cajan* (Choure and Dubey, 2012). De Boer *et al.* (2003) reported that Fusarium wilt of Radish can be effectively controlled by using a combination of *P. putida* strains RE8 and WCS358. Similarily, *P. fluorescens* NBRI-N and *P. fluorescens* NBRI-N6 demonstrated a superior control over collar rot in betelvine caused by *S. rolfsii* (Singh *et al.*, 2003).

Raaijimakers *et al.* (1995) reported that in a consortium comprising two different strains of pseudomonas, one strain of *Pseudomonas* act as siderophore, while another strain showed ability to induce disease resistance in the plant. The action of two pseudomonads was effective only when they are applied in combination however their single treatment does not have any impact disease control. In another example, mixtures of fluorescent pseudomonads have demonstrated an enhanced protection against take-all disease of wheat caused by *Gaeumannomyces graminis* var. *tritici* along with significant increase in yields (Pierson and Weller, 1994). However, another consortium comprising *Trichoderma koningii* and a nonpathogenic strain of *Fusarium oxysporum*, resulted in significant enhancement in the control of take-all disease of wheat (Duffy *et al.*, 1996) and Fusarium wilt of tomato (Lemanceau *et al.*, 1992). Stockwell *et al.* (2011) stated that combined efficacy of *Pseudomonas fluorescens* A506, and *Pantoea vagans* strain C9-1 or *Pantoea agglomerans* strain Eh252 is higher than individual strains. While A506 plays a key role in suppressing growth of the pathogen by limiting its chances on floral colonization, C9-1 and Eh252 produces peptide antibiotics which aids in disease control. The management of *F. oxysporum* f. sp. *cucumerinum* could be achieved by the combined effect of bacterium *P. putida* with saprophytic strains of *F. oxysporum* (Park *et al.*, 1998).

Mathivanan *et al.* (2005) reported that application of *P. fluorescens* and *T. viride* significantly reduces rice sheath blight disease in rice. The incidences of root rot of pea caused by *Aphanomyces euteiches* f. sp. *pisi* was considerably reduced on application of combined treatment of *T. harzianum* and *P. fluorescens* strain 2-79RN10 compared to single *T. harzianum* treatment (Dandurand and Knudsen, 1993). *Bacillus mucoides* and *Pichia guilermondii* has showed tremendous potential in suppressing gray mold of strawberry caused by *Botrytis cinerea* (Guetsky *et al.*, 2002). The consortia comprising *Pseudomonas aeruginosa* (MBAA1), *Bacillus cereus* (MBAA2), *Bacillus amyloliquefaciens* (MBAA3) and *Trichoderma citrinoviride* (MBAAT) has found to successful control various fungal diseases of *Glycine max* L. (Thakkar and Saraf, 2014).

Apart, from role of microbial consortium in biocontrol of various diseases, they are also responsible for plant growth promotion. It is reported that the bioagents in combination produces various growth promoting substances like IAA, GA and cytokinins along with high nutrient uptake ability which facilitates in better growth of crop plants. There are reports on combination of *P. fluorescens* strains EBC5 and EBC6 found to increase germination percentage, shoot length, and root length of chilli plant (Muthukumar *et al.*, 2010). Naseby *et al.* (2000) reported that three strains of *T. harzianum* increased fresh shoot weight, root weight, root length in pea more than single strain. Jain *et al.* (2017) reported that 12 isolates of *Trichoderma* spp. Inhibit the mycelial growth of *Rhizoctonia solani* causing banded leaf and sheath blight of little millet.

Plant microbe interaction

Plants always confront a battle with the environment surrounding them. Biotic stress is a natural element in plant lifecycle and thus the plants develop certain defense strategies to counter biotic stress (Durrant and Dong, 2004). Defense against pests is usually brought either by mechanical defense or production of chemicals, which may be toxic or in other ways not desired by the invading organisms. However, for defense against the pathogens, the hosts have evolved certain mechanisms such as microbe associated molecular pattern (MAMP) triggered defense, hormone signaling, innate immunity, systemic acquired resistance (SAR), chemical warfare, hypersensitive response and programmed cell death (Boller and Felix, 2009; Zipfel, 2009). In spite of possessing such phenomenal features, the plant still undergo pathogen attack and suffers losses in terms of quality and quantity.

Biological control has constantly evolved as an

Table 1. Microbial consortia effective against different plant pathogens along with their mode of action

Microbial consortium	Target pathogen	Mode of action	References
Trichoderma viride and *P. fluorescens*	*Rhizoctonia solani*	ISR and plant growth promotion	Mathivanan *et al*., 2005
Trichoderma viride, T. asperellum and T. harzianum	*Fusarium oxysporum*	-do-	Yadav *et al*., 2017
Trichoderma spp. (12 isolates)	*Rhizoctonia solani*	-do-	Jain *et al.*, 2017
Trichoderma viride	*Alternaria* spp.	Seed treatment and foliar spray	Shakywar *et al*., 2013
Trichoderma viride	*Fusarium oxysporum and Pythium irregualre*	Seed treatment and foliar spray	Jamwal and Jamawal, 2012
Fusarium oxysporum strain F047 and *P. putida*	*Fusarium oxysporum*	Siderophore mediated competition for carbohydrate	Lemanceau *et al*., 1993
P. fluorescens strain 2-79RN10 and *T. harzianum*	*Aphanomyces euteiches* f. sp. *pisi*	Siderophore, ISR and plant growth promotion	Dandurand and Knusden, 1993
Gaeumnnomyces graminis var. *graminis* and mixture of fluorescent pseudomonads	*Gauemannomyces graminis* var. *tritici*	Antibiosis	Dufy *et al*., 1995
P. aeruginosa, *Mesorhizobium* sp. and *T. harzianum*	*S. rolfsii*	ISR	Singh *et al*., 2012
P. aeruginosa, Bacillus subtilis and *T. harzianum*	*S. sclerotiorum*	ISR	Jain *et al.*, 2012
Trichoderma harzianum Tr6 and *Pseudomonas* sp. Ps14	*Cucumis sativus, A. thaliana*	Primed expression of a set of defense-related genes	Alizadeh *et al*., 2013
T. harzianum, *P. fluorecsens* and *G. intraradices*	*Fusarium oxysporum*	Plant growth promotion	Srivastava *et al.*, 2010
Rhizobium and *P. striata*	Nematode	Increase N and P availability	Pant *et al.*, 1983
P. fluorescens and *Stentrophomonas maltophila*	*Pythium* spp.	ISR	Dunne *et al.,* 1998
Pichia guilermondi and *B. mycoies*	*B. cinerea*	Competition	Guetsky *et al*., 2001; 2002
P. fluorescens NBRI-N6 and *P. fluorescens* NRI-N	*Sclerotium rolfsii*	ISR	Singh *et al*., 2003
P. strita and *Rhizobium* sp.	*Meloidogyne incognita*	Plant growth promotion	Siddique and Singh, 2005
Bacillus sp. strain mixture IN937b + SE49 and T4 + INRN	*Colletotrichum gleosporoides* and Cucumber mosaic virus	ISR	Jetiyanon *et al*., 2003
Burkholdria OSU 7, *Bacillus* OSU 142 and *Pseudomonas* BA 8	Brown rot of apricot *Moniliana laxa* Ehr.	Antibiosis	Altindag *et al*., 2006
P. fluorescens EBC5 and *P. fluorescens* EBC6	*Pythium apanidermatum*	ISR	Muthukumar *et al*., 2010
P. chlororaphis PCL1391 and *P. fluorescens* WCS365	*Colletotrichum lindemuthianum*	Plant growth promotion, Reduced sporulation and conidial germination	Bardas *et al*., 2009
P. fluorescens Aur 6 and *Chryseobacterium balustinum* Aur 9	*Pyricularia oryzae*	ISR	Lucas *et al*., 2009
B. subtilis MBI600 and *Rhizobium tropici* UMR 1899	*Fusarium solani* f. sp. *phaseoli*	Siderophore production	Estevez de Jensen *et al*., 2002
Rhizobia, B. cereus strain BS03 and *P. aeruginosa* RRLJ04	*Fusarium udum*	Higher PAL, PO and PPO activities	Dutta *et al*., 2008
Trichoderma harzianum DB11 and *Gliocladium catenulatum* Gliomix	*Phytophthora cactorum* and *P. fragariae*	Possible antimicrobial activities	Vestberg *et al*., 2004

important strategy for management of plant disease. It is not only an important tool for managing phytopathogens but also aids in plant growth promotion (Vacheron *et al*., 2014). The plant microbe interactions can further be classified as neutral, beneficial or detrimental. The beneficial microbes colonizing the rhizospheric zone, suppress plant disease and facilitate its growth through various mechanisms, such as antibiosis, high nutrient uptake, production of enzymes and toxins as well as release of pathogen inhibiting compounds (Whipps, 1997). The symbiotic association between various microbes has found to display elevated defense compared to individual application.

Induced systemic resistance

Plants are known to produce various defense related compounds in response to stress conditions. The time at which defense has been initiated decides the fate of the plant *i.e*., whether it can withstand the pathogen attack or may succumb (Choudhary *et al*., 2007). If defense mechanisms are triggered in the plant prior to pathogen attack, disease can be reduced to a considerable extent. Two types of resistance have been reported to trigger inside the plants *i.e*., Induced Systemic Resistance (ISR) and Systemic Acquired Resistance (SAR). Induction of ISR in plants is through non pathogenic microorganisms residing in the plant

rhizospheric zone prior to pathogen challenge. However, SAR is elicited in the plant in response to local infection and leads to systemic expression of long lasting resistance against a broad range of phytopathogens. Both resistance triggers a defense response inside the host, which protects them from the invading pathogen and their subsequent attack. The beneficial rhizobacteria usually do not cause necrosis in the host plants, which may strengthen the possibility that there is considerable variation in the eliciting factors produced by them from that of pathogen elicitors (Somers *et al*., 2004; Garbeva *et al*. 2004; Persello-Cartieaux *et al.* 2003).

Three pathways leading to induction of systemic resistance in *Arabidopsis* have been reported so far out of which two lead to production of pathogenesis-related (PR) proteins (Neuhaus, 1999). In the first pathway, PR proteins are formed in response towards attack of pathogenic microorganisms, whereas second pathway leads to production of PR proteins as a result of wound or necrosis. However, in both the pathways, alternate mechanisms for induction of PR proteins are present. The expression of PR-proteins can be used as an indicator of ISR (Van Loon, 1997). The pathogen-induced pathway uses salicylic acid (SA) as the primary signaling molecule, whereas the defense signal is triggered in wound induced pathway by Jasmonic acid (JA) and ethylene (Raskin, 1992; Mauch-Mani and Metraux, 1998; Cameroon, 2000). When applied exogenously, these compounds and their analogues induce similar responses along with a substantial cross talk between the pathways (Pieterse *et al*., 2001).

Cross talk between salycylic acid and jasmonic acid/ ethylene dependent pathways

The induction of systemic resistance in plants is regulated by complex signaling pathways including salicylic acid, or jasmonic acid and ethylene (Dong, 1998). These pathways have neither synergistic nor antagonistic effect on each other. This cross talk between the signaling molecules offers great regulatory potential to the plant. Depending upon the invading microorganism, the plant can differentially use a pathway that can best halt the pathogen. The plant can block one or the other signal, guided by the defense compound required. The defense compounds are SA induced PRPs, plant defensin, thionins and protease inhibitors (Van Peer *et al*., 1991).

Resistance conferred by SA dependent pathway may be effective against certain pathogens on which the JA/ ethylene pathway may be ineffective and *vice-versa*. Thus, it entirely depends upon the plant to block the unwanted signal. This has been confirmed using *Arabidopsis* mutant genotypes that do not respond to JA/ethylene signal (coil mutant) or SA signal (*npr1* mutant or transgenic *nahG* plants (Bowling *et al*., 1994). The basal resistance of JA mutant against the fungal pathogens *Alternaria brassicola* and *Botrytis cinerea* was lost, whereas there was no change in resistance against *Perenospora parasitica*. In contrast *npr1* and transgenic *nahG* plants, which are both blocked in response to SA, show a lower level of basal resistance against *P. parasitica*, whereas basal resistance against *A. brassicola* and *B. cinerea* remain unaffected (Parker *et al*., 1996). This clearly demonstrates that the defense compounds produced by SA dependent or SA independent pathways have different specificities against pathogens.

The ISR mediated pathway has been studied in *Arabidopsis thaliana*, in which JA and ethylene are the main signaling molecule involved. *Pseudomonas* spp. have been reported to induce resistance in several plants *viz.,* tobacco, cucumber, radish, carnation and *Arabidopsis*, which ultimately enhances the defense potential of the host plant upon pathogen challenge (Bakker *et al*., 2007; Weller *et al*., 2012). The combined activity of ISR and SAR lead to enhance protection against the pathogens which defy the two pathways or through ISR/SAR alone. Apart from *Pseudomonas*, ISR is also triggered by various *Bacillus* sp. such as *B. subtilis, B. pumilus, B. pasteurii, B. cereus, B. amyloliquifaciens, B. sphaericus* and *B. mycoides* which elicit significant disease reduction in a wide range of hosts (Chaudhary and Johri, 2008; Kloepper *et al*., 2004).

Role of antioxidants

Reactive oxygen species (ROS) are produced in plants in response to recognition of a pathogen attack. They are also potential regulators of various cellular processes such as growth, development and other defense related pathways. Superoxide ion (O_2), or its dismutation product hydrogen peroxide (H_2O_2), plays a vital role in recognition of different types of pathogens. Plants possess the ability to regulate the levels of ROS in such a manner that the levels are detrimental to the pathogen but not to the host. There are two phases of ROS accumulation by avirulent pathogens- first which is small and transient followed by a constant phase having direct correlation with resistance. However, reports where ROS accumulation have taken place in three phases have also exists *i.e. Blumeria graminis* f. sp. *hordei* infecting barley and *Septoria tritici* infecting wheat. Induction of ROS in plants leads to strengthening of host cell walls *via.* a cross linking of glycoproteins.

ROS, especially H_2O_2, is an important component mediating primary defense responses triggered by the host (Custers *et al*., 2004; Walters, 2003). However, the actual toxicity of ROS in a given plant–pathogen interaction is

dependent on sensitivity of the pathogen towards a particular level of the ROS induced. The amount of extracellular H_2O_2 formed relies on various factors such as type of plant species, age or developmental stages of the plant cells and nature of the elicitor (Legendre *et al.*, 1993). It is reported that micromolar concentrations of H_2O_2 inhibited spore germination of a number of fungal pathogens *in vitro*. Thus, a concentration of 0.1 mM H_2O_2 completely inhibited growth of *Pectobacterium carotovorum* sub sp. *carotovorum* and also resulted about 95 per cent inhibition in growth of *Phytophthora infestans*.

In response to stress, there is an increase in the synthesis of antioxidants. However, it has been found that the formation of antioxidants is quite high in case of plants treated with microbial consortium. Singh *et al.* (2013) reported that in respect of single microbial treatment, the antioxidant activities increased 1.8-3.3 folds in the triple microbe consortium consisting of *T. harzianum* THU0816, *P. aeruginosa* PHU094 and *Mesorhizobium* sp. RL091 treated chickpea plants under *S. rolfsii* challenge. Apart from the ROS, other defense related enzymes such as PAL and PO are also enhanced in a consortium of *Trichoderma* and *Pseudomonas* (Kartikeyan *et al.*, 2006)

Drawbacks of microbial consortium

The role of microbial consortium in plant growth promotion and disease suppression has been widely documented (Chandanie *et al.*, 2006; Raimam *et al.*, 2007). However, there are several reports which reveal that consortium is unable to demonstrate minimal effect compared to their single application (Schisler *et al.* 1997; Schmidt *et al.*, 2004). *Bacillus subtilis* and *Fusarium oxysporum* strains when applied together did not provide control over Fusarium wilt of chickpea (*F. oxysporum* f. sp. *ciceri*), as compared to single treatment (Hervas *et al.*, 1997). Co-inoculation with *B. subtilis* 101 and *Azospirillum brasilense* Sp245 had no significant effect on plant growth whereas their individual application showed opposite result (Felici *et al.*, 2008). In another example, Walker *et al.* (2012) reported that co inoculation of maize roots with a mixture of *Pseudomonas*, *Glomus* and *Azospirillum* do not play any role in activation of secondary metabolites as compared with single application of *Glomus*. This may be due to non compatibility of the two strains which show antagonistic activity against each other (Siddiqui and Shaukat, 2002; Whipps, 2004). The non compatibility of isolates in microbial mixture may be attributed to their independent signalling pathways, which do not show a synergistic effect. For example, competition for iron or different substrates limit the colonization ability of introduced biocontrol strains. One of the key points in preparing a successful microbial consortia is to identify and screen different BCAs with diverse activities whose combined effect leads to enhance plant's performance otherwise the main objective behind its development will all go in vain.

Future prospects

In microbial consortium one of the chief hindrances lies in the maintenance of the viable number of spore count of the microorganisms after their formulation and prior to application. When the consortium is tested for experimental purposes, it is possible to prepare the inoculum within 1-2 days before its application, but in case of their commercial application they must survive long transportation run. The future research which lay an important thrust is the formulation of biocontrol agents in such a way so that they can thrive the long gap between their manufacture and application. Bioagents used in microbial consortium must be safe and sound to human health as few of them are responsible for causing dreadous human infection. For example, *Pseudomonas aeruginosa*, which shows high antagonistic activity towards *Pyricularia grisea* causing gray leaf spot of turf, occur as a virulent opportunistic human pathogen causing surgical wounds with severe burns. Moreover *Burkholderia cepacia*, a popular biocontrol agent of pea root rot has found to cause opportunistic lung infections in patients with cystic fibrosis. Cost, efficacy, convenience and consistency of biological controls are important factors that must be considered during formulation of microbial consortium. Crop based microbial consortium may be developed to meet specific requirement. Work on gene and on gene products of bioagent of microbial consortium to be done for specifying mode of action. Statistical procedure and mathematical model to be developed for assessing the interaction between constituents of microbial consortium, interaction with pathogen and for assessing their biocontrol and growth promotion ability separately. The study on the role of different microbes in consortia apart from plant disease management could also be a new field of research. In addition to it, different metabolites from the culture of microbial mixes could be assessed for their potential in boosting crop production and regulation of some specific genes.

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Yield and quality parameters of guava fruits grown in conventional and organic farming system

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ABSTRACT

The yield and quality profile of organic and conventionaly grown guava fruits in new alluvial zones of West Bengal were compared. In total, 400 fruits (from both growing systems) were analyzed for a range of quality parameters. Data were evaluated by ANOVA and means were compared by using t-test. Compared with conventional system, organic fruits were rich in minerals like phosphorus, potassium, zinc, manganese and calcium. More fruit weight and yield were obtained in conventionally grown system, while quality parameter and shelf life were better in organically grown fruits. An important constituent *i.e.* vitamin C content of guava fruits was also higher (139.27 mg $100g^{-1}$) in organically grown fruits as compared to conventional once. Hence, organic system can be recommended for quality produce of guava.

Key words: Guava, quality, organic sysytem, conventional system

Guava (*Psidium guajava* L.) is one of the important fruit crops of West Bengal, particularly in the alluvial zone of West Bengal. The fruits are rich in minerals, vitamin C and pectin. Guava is such a horticultural crop, where fruits are consumed fresh after harvest along with peel, hence for safe nutrition, there is need of organic cultivation of guava. Vitamin C content in guava fruit is an important ingredient, which act as antioxidant and as agent that contributes to anticarcinogenic or cardioprotective properties (Rice-Evans *et al*., 1996, Rapisarda *et al*., 1999). This vitamin C is also considered as a very important water soluble antioxidant, as it protects compounds in extracellular and intracellular spaces in most biological systems and reduces tocopherol radicals back to their active form at the cellular membranes. It can also directly scavenge superoxide radicals singlet oxygen, hydrogen peroxide and hydroxyl radicals (Kaur and Kapoor, 2001; Klimezak *et al*., 2007). So, the usefulness of guava fruit is important for the purpose of health benefits. The quality of organic fruit is often higher as compared to conventionally grown once. Greater vitamin C content in citrus in organically grown fruits compared to conventionally grown system was also reported by Tarozzi *et al*. (2006). Scanty information is available in guava fruit on this aspect. Keeping this, the present investigation was undertaken.

MATERIALS AND METHODS

The study was conducted at the farmer's field, nearby Regional Research Station, Bidhan Chandra Krishi Viswavidyalaya, Gayeshpur during 2012-2014 on 8 years old guava trees, cv. L-49, having uniform growth and vigour. The two growing systems were studied. First in the orchard, where recommended dose of chemical fertilizers (N-260 g, P_2O_5–320 g and K_2O-260 g) $plant^{-1}year^{-1}$ were applied *i.e.* conventional system, and the second was organic system. In organic system trees were grown with *Azotobacter* (200 g) + *Azospirillum* (100 g) + VAM (100 g) + potassium mobilizer (100 g) + 2.5 kg vermicompost, all $plant^{-1}$ $year^{-1}$. In this system, plant protection measures were also by organic means only. Yield and fruit physical quality were recorded at maturity at harvest. The mature fruits from each system harvested were used for physico-chemical analysis following all standard methods as described by Ranaganna (2000). Mineral content of the fruits were also estimated following all standard methods.

RESULTS AND DISCUSSION

Mineral contents of guava fruits varied significantly in two growing systems (Table 1). Phosphorus, potassium, zinc, manganese and calcium content of fruits were higher in organically grown fruits, while nitrogen and iron content of fruits were relatively higher in fruits grown conventionally *i.e.* with chemical fertilizers. Similar results were also obtained by Neuhoff *et al*. (2011) in oranges, Shankar *et al*. (2012) in tomato and Dutta and Talang (2014) in mango. The absorption of micro-nutrient such as iron and zinc from soil is significantly influenced by the application of organic manures and bio-fertilizers. Soil, that has been managed organically has more micro-organism, which produces many compounds that influences the plant to absorb more micronutrients from soil. It is also reported that substances, such as citrate and lactate combine with the soil minerals

Table 1. Mineral content of organically and conventionally grown guava fruits

Parameter	Organic	Conventional	Level of significant
Nitrogen (% dry weight)	0.49	0.51	P≤0.01
Phosphorus (% dry weight)	0.11	0.09	P≤0.01
Potassium (% dry weight)	0.75	0.61	P≤0.01
Zn (ppm)	41.00	23.00	P≤0.01
Fe (ppm)	57.00	58.00	N.S
Mn (ppm)	17.90	11.20	P≤0.01
Ca (% dry weight)	0.074	0.069	P≤0.01

and make them more available to plant roots. Particularly for iron, it is especially important because many soils contain adequate iron but not in an available form. The presence of these micro-organisms explains the trend showing a higher mineral content of organic food crops (McClintock, 2004).

Yield and quality parameters are also influenced by different growing systems (Table 2). Fruit weight and yield were recorded relatively more in conventional farming system as compared to organic farming, while biochemical parameters like total soluble solids, total sugar and ascorbic acid content of fruit were more in fruits grown organically compared to conventional system. Organically grown fruits also exhibited more shelf life (9 days) at ambient room temperature. The increased fruit quality may be explained from the fact that organic growing system enhanced the nutrient availability by enhancing the capability of plant for better uptake of nutrients from rhizosphere. The results are in close conformity with the findings reported by Korwar *et al*. (2006) and Pathak *et al.* (2005) in aonla. Dutta and Talang (2014) also found similar results in mango. Finally, it is concluded that organic growing system can be recommended in guava for obtaining better fruit quality including vitamin C content, which is an important constituent.

Table 2. Effect of growing systems on yield and quality parameters of guava fruits

Parameter	Organic	Conventional	Level of significance
Fruit weight (g)	138.10	142.33	P≤0.01
Fruit yield (kg tree^{-1})	36.77	41.27	P≤0.01
Fruit length (cm)	7.22	7.31	N.S.
Total sugar (%)	6.97	6.11	P≤0.01
Acidity (%)	0.31	0.37	P≤0.01
Ascorbic acid (mg 100g^{-1})	139.27	123.97	P≤0.01
Shelf life (days)	9	5	P≤0.01
Total soluble solids (°Brix)	10.20	9.80	P≤0.01

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Response of Knol khol (*Brassica oleracea* L. var. *gongylodes*) to different organic amendments and microbial consortium

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ABSTRACT

The performance of Knol khol (*Brassica Oleracea* L. var. *gongylodes*) var. White Vienna was evaluated in different organic amendments along with microbial consortium during 2014-15 and 2015-16. There were eight treatments consisting of T_1 (Microbial consortium + Rock phosphate), T_2 (T_1 + 2.5 t ha^{-1} compost), T_3 (T_1 + 5 t ha^{-1} compost), T_4 (T_1 + 2.5 t ha^{-1} Vermicompost), T_5 (T_1 + 5 t ha^{-1} Vermicompost), T_6 (Enriched compost 2.5 t ha^{-1}), T_7 (Enriched compost 5 t ha^{-1}) and T_8 (Recommended dose of fertilizer). The highest yield of 191.45 q ha^{-1} was recorded in conventional treatment, while among the organic treatments, T_7 (Enriched compost 5 t ha^{-1}) recorded the highest yield (169.73 q ha^{-1}) and other yield attributing parameters. The lowest yield attributing characters were exhibited by T_1 (Rock phosphate + Consortium). On quality parameters, the organic treatments exhibited superior results, when compared to inorganic treatments.

Key words : Knol khol, carotene, ascorbic acid, ash, enriched compost, protein

Knol khol is characterized by the formation of knob, which arises from thickening of stem tissues above the cotyledons. It is a good source of carbohydrate, protein, vitamin C and minerals like Ca, P and K. Growth, yield and quality depend upon nutrient availability in soil, which is related to judicious application of manures and fertilizers. To increase the productivity and profitability, the use of chemical fertilizers, pesticides and other chemical inputs are increasing day by day resulting in economic, environmental and ecological problems, which adversely affected the sustainability of agricultural system as well as human health hazards. The continuous use of chemical fertilizers badly affect the texture and structure of soil, reduce soil organic matter content and decreases microbial activity of soil (Alam *et al*., 2007). Hence, the vegetables that are produced organically are gaining importance because of less chemical residues. The organic manures give better quality produce as compared to those grown with inorganic sources of fertilizers.

MATERIALS AND METHODS

The present experiment was carried out in the Experimental Farm, Department of Horticulture, during 2014-15 and 2015-16 in Assam Agricultural University, Jorhat in Randomized Block Design with three replications. The experimental site is located 26°47' N latitude, 94°12 E longitude and 86.8 m above mean sea level of Upper Brahmaputra Valley Agro Climatic Zone of Assam. The experiment was laid out with eight treatments in RBD with three replications. The treatments were T_1 : Rock phosphate + Consortium; T_2 : T_1 + Compost @ 2.5 t ha^{-1}; T_3 : T_1 + Compost @ 5 t ha^{-1}; T_4 : T_1 + Vermicompost @ 2.5 t ha^{-1}; T_5 : T_1 + Vermicompost @ 5 t ha^{-1}; T_6 : T_1 + Enriched compost @ 2.5 t ha^{-1}; T_7 : T_1 + Enriched compost @ 5 t ha^{-1} and T_8 : Recommended dose of fertilizer @ 80 : 60 : 60 NPK hectare^{-1}. The consortium used in the experiment was the mixture of *Azotobacter, Azospirillum,* Phosphate solubilising bacteria and *Rhizobium* and the variety was White Vienna. There were 24 plots, each having 36 plants with a spacing of 40 x 30 cm, within rows and plants respectively. Individual plot size was 4.32 m^2 and the total area of the experimental site was 250 m^2. Yield, quality and soil health parameters were studied for two consecutive years and statistically analyzed.

RESULTS AND DISCUSSION

The present study was targeted with the aim to find out the effect of biofertilizer consortium (*Rhizobium, Azotobacter, Azospirillum* and PSB) alone or in combination with other organic fertilizers like vermicompost and enriched compost at different doses on growth, yield and quality of Knol khol and post soil fertility status.

The knob diameter is an important determinant of yield in Knol khol, which was greatly influenced by different sources (Table 1). In the present study the highest knob diameter of 8.41 cm was recorded in T_8 (RDF) followed by 7.89 cm in the treatment receiving Enriched compost 5 t ha^{-1}. The increase in the knob diameter might be due to the more photosynthesis rate from larger photosynthetic area and their translocation towards knob resulting in increase in the knob diameter. The present findings are in close conformity with the findings of Sharma and Singh (2003) and Chaurasia *et al*. (2001). The increased knob diameter in organic treatment

might be due to the better plant stand and direct contribution of organic inputs with consortium in improving the fertility condition of the soil because of microbial activities.

A significant influence of treatment on knob yield plant^{-1} and yield hectare^{-1} was observed with a maximum of 211.76 g plant^{-1} and 191.45 q ha^{-1} respectively in T_8 i.e. RDF (80 : 60 : 60 kg NPK + 10 t FYM ha^{-1}). This could be due to the rapid availability and utilization of nitrogen for various internal processes in the plant. The higher leaf number provided by this treatment facilitates larger photosynthetic area coupled with increased uptake of water and nutrients from soil might have resulted in increased production of photosynthates, leading to better filling of knobs and thereby increasing yield. Irrespective of the types of nutrient sources in all cases, the treatments receiving enriched compost 5 t ha^{-1} recorded more yield of 203.70 g plant^{-1} and 169.73 kg ha^{-1} than other treatments. Increase in the yield is due to the supply of additional nutrient through organics as well as improvement in the physical and biological properties of soil (Sharma *et al*., 2005). The increase also might be due to fact that these nutrients are being important constituents of nucleotides, proteins, chlorophyll and enzymes, involved in various metabolic process, which have direct impact on vegetative and reproductive phase of the plants. It seemed that organic manure need more time for nutrients to be available for plant absorption. However, the beneficial effect of organic manure on yield may be due to an increase in organic matter rate caused by the generation of carbon dioxide during compost decomposition (Wilkinson, 1979) and improvement of the soil physical conditions, which encouraged the plant to have a good root development by improving the aeration of the soil (Arisha *et al*., 2003). Application of organic manure increases microbial population in soil that helps the soil to release various immobile nutrients. These microbes also produce PGR that are important for plant growth and photosynthetic activity (Levy and Taylor, 2003). These results are in consonance with Raja *et al*. (2006) and Zaki *et al*. (2009).

Effect of organics on quality parameters

Table 2 shows the qualities of knobs and vitamin C (ascorbic acid) content. The present study shows that the highest ascorbic acid content (64.22 mg 100 g^{-1}) was recorded in T_7 (Enriched compost 5 t ha^{-1}) and the lowest ascorbic acid content of 35.01 mg 100g^{-1} was observed in treatment receiving RDF. The findings are in close agreement with those earlier reported by Guo *et al*. (2004) in cabbage and Sable and Bhamare (2007) in cauliflower. A negative correlation between vitamin C content and level of applied nitrogen is often reported (Lee and Kader, 2000).

Ash content represented the total amount of non-combustible substances *i.e.* minerals present in the plant product. In the present study highest ash content of 4.66 per cent in knob was found in T_5 (T_1 + Vermicompost 5 t ha^{-1}). This might be due to the increase in quality parameters, because soil that has been managed organically has more microorganisms, which produce many compound that influence the plant to absorb more micro nutrients from soil.

In the present study, it has been observed that the protein content was highest under T_8 (RDF) treatment and the lowest was under T_1 *i.e.* Rock phosphate + Consortium (Table 2). The increase in protein content with higher dose of nitrogen content might be due to the fact that nitrogen is a major contributor of protein synthesis. When nitrogen is adequate, proteins are formed from the manufactured carbohydrate. A positive correlation between protein content and level of applied nitrogenous fertilizer was also found by Khorr and Vogtmann (1983). Similar results has been clearly demonstrated by Kumpulainen (2001) in potato and Shelke *et al.* (2001) in brinjal.

Carbohydrate content in Knol khol seems to be affected by different treatments significantly. From Table 2 it is seen that maximum carbohydrate content (6.74 %) was recorded

Table 1. Knob yield of Knol khol (*Brassica oleracea* L.var. *gongylodes*)

Treatment	Knob diameter (cm)			Knob yield plant^{-1} (g)			Knob yield ha^{-1} (q ha^{-1})		
	2014-15	2015-16	Pooled	2014-15	2015-16	Pooled	2014-15	2015-16	Pooled
T_1 : Rock phosphate + Consortium	5.15	5.26	5.21	58.27	56.07	57.17	46.49	47.48	46.99
T_2 : T_1 + Compost (2.5 t ha^{-1})	6.06	6.14	6.10	83.38	88	85.87	87.57	98.26	92.92
T_3 : T_1 + Compost (5 t ha^{-1})	6.71	6.50	6.60	125.24	130.04	127.64	104.54	106.71	105.66
T_4 : T_1 + Vermicompost (2.5 t ha^{-1})	7.11	7.13	7.12	153.35	155.33	154.34	127.94	129.65	128.80
T_5 : T_1 + Vermicompost (5 t ha^{-1})	7.53	7.54	7.53	171.83	176.15	173.99	143.29	148.48	145.88
T_6 : Enriched compost (2.5 t ha^{-1})	7.77	7.65	7.71	182.13	185.90	184.02	151.81	155.99	153.90
T_7 : Enriched compost (5 t ha^{-1})	7.88	7.91	7.89	201.44	205.96	203.70	168.07	171.45	169.73
T_8 : RDF (80:60:60 kg ha^{-1} NPK + 10 t ha^{-1} FYM)	8.38	8.45	8.41	209.04	214.33	211.76	189.35	195.56	191.45
S. Ed (±)	0.19	0.23	0.22	2.69	1.94	1.65	1.44	1.03	0.99
CD (5%)	0.45	0.52	0.50	6.45	5.35	4.57	4.00	2.85	2.75

in T_7 (Enriched compost 5 t ha^{-1}) and minimum carbohydrate content (5.03 %) was recorded in T_8 (RDF). It might be due to the fact that when a plant is exposed to more of nitrogen, it increases protein production and reduces carbohydrate concentration .

Carotene content of Knol khol under different treatments showed significant differences among themselves (Table 3). The highest carotene content (4.73 µg g^{-1}) was observed in T_7 (Enriched compost 5 t ha^{-1}) and lowest carotene content (2.66 µg g^{-1}) was recorded in T_1 (Rock phosphate and consortium). Increased availability and uptake of nutrients, particularly micronutrients may be one of the reasons for improved carotene content in organically treated Knol khol. Similar results were reported by Borgohain (2012) in tomato. Table 3 represents the mineral content of knob, such as Ca, P and K, which were recorded more in organic treatments. Increased mineral content in organic treatments might be due to more beneficial micro organisms in the soil, which produce many compounds that influence the plant to absorb more micronutrients from soil. Similar observations also reported by Shelke *et al*. (2001) in brinjal.

Physiological weight loss (Table 4) was found highest

Table 2. Biochemical characters of Knol khol (*Brassica oleracea* L.var. *gongylodes*)

Treatment	Ascorbic acid (mg 100g^{-1})			Carbohydrate (%)			Protein (%)			Ash (%)		
	2014-15	2015-16	Pooled	2014-15	2015-16	Pooled	2014-15	2015-16	Pooled	2014-15	2015-16	Pooled
T_1	35.35	35.07	35.21	5.03	5.05	5.04	1.68	1.81	1.74	4.97	6.05	6.01
T_2	39.16	40.05	39.60	5.33	5.38	5.35	1.75	1.82	1.78	6.18	6.21	6.19
T_3	43.50	44.66	44.08	5.23	5.29	5.26	2.15	1.93	2.04	4.92	4.94	4.93
T_4	46.85	48.23	47.55	5.42	5.45	5.44	2.25	2.50	2.37	6.45	6.58	6.51
T_5	51.66	53.84	52.75	6.05	6.19	6.12	2.33	2.64	2.48	8.61	8.72	8.66
T_6	62.48	60.37	61.42	6.30	6.35	6.32	2.51	2.61	2.56	8.21	8.20	8.20
T_7	64.22	62.87	63.54	6.69	6.80	6.74	2.64	2.82	2.75	8.24	8.36	8.30
T_8	33.37	36.65	35.01	5.02	5.05	5.03	2.92	3.05	2.98	6.87	6.90	6.88
S. Ed (±)	1.08	0.47	0.62	0.09	0.05	0.03	0.06	0.08	0.07	0.08	0.10	0.06
CD (5%)	2.99	1.30	1.73	0.22	0.12	0.19	0.18	0.26	0.22	0.22	0.26	0.16

Table 3. Calcium, phosphorus, potassium and carotene content in different treatments

Treatment	Calcium (%)			Phosphorus (%)			Potassium (%)			Carotene (µg g^{-1})		
	2014-15	2015-16	Pooled	2014-15	2015-16	Poold	2014-15	2015-16	Pooled	2014-15	2015-16	Pooled
T_1	1.51	1.55	1.52	0.21	0.26	0.23	2.50	2.56	2.53	2.67	2.65	2.66
T_2	1.11	1.18	1.14	0.18	0.26	0.22	2.69	2.60	2.64	3.05	3.06	3.05
T_3	1.33	1.39	1.36	0.56	0.51	0.53	3.06	2.87	2.96	3.40	3.25	3.32
T_4	1.28	135	1.31	0.59	0.64	0.61	3.58	3.90	3.74	3.86	3.94	3.90
T_5	1.77	1.70	1.74	0.61	0.66	0.63	4.08	4.19	4.13	4.55	4.71	4.63
T_6	1.89	2.04	1.96	0.48	0.49	0.48	4.13	4.21	4.17	4.12	4.08	4.10
T_7	2.00	2.12	2.06	0.66	0.72	0.69	4.58	4.66	4.62	4.65	4.82	4.73
T_8	0.86	0.92	0.89	0.60	0.65	0.62	3.92	3.91	3.91	3.35	3.38	3.36
S. Ed (±)	0.03	0.03	0.02	0.03	0.02	0.02	0.04	0.05	0.03	0.18	0.28	0.25
CD (5%)	0.09	0.08	0.06	0.07	0.06	0.04	0.17	0.19	0.15	0.45	0.63	0.53

Table 4. Physiological weight in loss (%) in different treatments

	Knob wt. (g)					
Treatment	Day 1	Day 3	Day 5	Day 7	Day 9	Day 11
T_1	58.36	8.16	18.12	22.02	-	-
T_2	83.10	7.31	15.17	19.60	26.00	-
T_3	124.10	7.58	15.18	18.34	25.60	-
T_4	150.25	6.72	12.24	16.71	25.22	-
T_5	173.67	6.33	10.63	15.33	24.14	24.18
T_6	185.42	5.50	11.71	15.01	19.57	26.98
T_7	200.42	4.71	9.38	13.88	19.66	24.05
T_8	212.63	10.12	21.12	30.36	-	-
S. Ed (±)		1.16	1.22	2.35	2.12	1.09
CD (5%)		3.42	3.67	5.79	4.86	2.32

in conventional treatment and significantly low under the organic treatments. Low PLW in organic treatments might be due to higher availability of antioxidants and growth retarding substances which interferes with metabolic activities within the vegetables (Mc Sheehy, 1977). Moreover, the availability of all macro and micro nutrients from the organic sources in moderate amounts might have also helped in enhancing the storage period. The higher moisture content in inorganic Knol khol leads to increased respiration, rotting and decaying and ultimately decreases the shelf life. This corroborate with the results of Mali (2004) in cucumber and Kumar (2011) in cabbage.

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Response of brinjal (*Solanum melongena* L.) to organic manures and foliar nutrition

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ABSTRACT

An investigation was undertaken at Agricultural Research Station, Palghar, District Palghar, Maharashtra State during *rabi* season of 2010-11 to study the effect of organic manures and foliar nutrition on growth and yield of brinjal. The application of vermicompost and spraying of 0.5 per cent at 30 and 45 days after transplanting in brinjal recorded the highest plant height (113.50 cm), 4.56 branches per plant. with large sized fruits. The plant spread, fruit weight, length and diameter did not differ significantly due to combined effect of organic manures and foliar spray. The significantly maximum fruit yield (1.54 kg plant^{-1} and 346.65 q ha^{-1}) was recorded in M_2T_3 treatment *i.e.* application of vermicompost and spraying of 0.5 per cent 19:19:19 NPK at 30 days and 45 days after transplanting.

Key words: Brinjal, vermicompost, foliar nutrition, yield

The brinjal (*Solanum melongena* L.) is popular, high productive and principal vegetable crop and known as poor man's crop. It is a widespread vegetable and can be grown throughout the year in almost all the states of India except at higher altitudes.

Plant nutrition plays an important role for enhancing yield and quality in brinjal. In recent years, the role of foliar nutrition and organic manures are gaining more importance to boost not only the productivity but also to improve the quality of the produce. Application of some portion of organic source of manure as a source of required nutrients play positive impact on soil physical and chemical properties, which ultimately increase the productivity. FYM and vermicompost are locally available and cheap sources of organic manures. Subbiah *et al*. (1985) obtained higher yields of tomato and eggplant with combined use of FYM and fertilizers. Maintenance of soil fertility is a prerequisite for the long term sustainable agriculture, where organic manures can play a vital role. Moreover, the foliar nutrition is greatly beneficial, when used as in addition to soil fertilization. Absorption of nutrients through foliage is known to affect the plant metabolism faster than their absorption through roots. Hence, an investigation was undertaken to study the effect of organic manures and foliar nutrition on growth and yield of brinjal.

MATERIALS AND METHODS

A field experiment was carried out at Vegetable Improvement Scheme Farm, Agricultural Research Station, Palghar, District Palghar, Maharashtra State during *rabi* season of 2010-11. The soil properties of experimental site are given in table 1. The experiment was laid out in factorial RBD with four replications. The brinjal variety, CHES-309 was selected for the study, as it is recommended for cultivation in the region. The main plot treatments were of two sources of organic manures *viz*. farm yard manure (FYM) @ 25.0 t ha^{-1} and vermicompost @ 12.5 t ha^{-1}. The subplot treatments were the different levels of foliar nutrition of 19:19:19 NPK as no spray, 0.5 per cent at 30 days after transplanting, 0.5 per cent at 30 days and 45 days after transplanting and 1.0 per cent at 30 days after transplanting. Recommended tillage practices for land preparation were carried out and plots of 4.5 m X 3.6 m were prepared and specified dose of organic manures were incorporated in the plots as per the main plot treatments. The seedlings of six weeks old were transplanted at a spacing of 75 cm X 60 cm. All the experimental plots received NPK fertilizers @150 : 50 : 50 kg ha^{-1}. Full dose of P, K and 1/3rd dose of N was given at the time of transplanting of seedlings and remaining N was given in two equal doses at three weeks interval. The foliar spraying of 19:19:19 NPK (granular foliar fertilizer) was done at scheduled time and concentration. The recommended cultural practices like irrigation, weed control, plant protection *etc*. were followed equally to all the experimental plots. Observations on growth (at 75 days after

Table 1. Physico-chemical properties of experimental sites

Properties	Value
pH	8.6
EC (ds/m)	0.36
Organic Carbon (%)	0.99
Available Nitrogen (N) kg ha^{-1}	148.96
Available Phosphorus (P_2O_5) kg ha^{-1}	2.04
Available Potassium (K_2O) kg ha^{-1}	176.17
Calcium Carbonate	3 to 7 %

transplanting), fruiting parameters and yield were recorded. Data were analyzed statistically (Panse and Sukhatme, 1985).

RESULTS AND DISCUSSION

The data on effect of organic manures and foliar nutrition on growth of brinjal is presented in table 2. The organic manures and foliar nutrition showed the significant effect on growth of brinjal plants.

The highest plant height (101.03 cm) was recorded in vermicompost treatment, which exhibited 6.54 cm more growth than FYM treatment. In case of foliar nutrition, the maximum plant height (105.46 cm) was observed in spraying of 0.5 per cent 19:19:19 NPK at 30 days and 45 days after transplanting (T_3) followed by 1.0 per cent at 30 days after transplanting (T_4). The plant height also significantly differed in interaction. The significantly highest plant height (113.50 cm) was recorded in M_2T_3 treatment combination, application of vermicompost and spraying of 0.5 per cent 19:19:19 NPK at 30 days and 45 days after transplanting followed by application of vermicompost and spraying of 1.0 per cent 19:19:19 NPK at 30 days after transplanting (M_2T_4). The lowest height was recorded in application of FYM without foliar nutrition (M_1T_1).

The significantly maximum plant spread (47.65 cm) was recorded in vermicompost treatment, which exhibited 1.81 cm more growth than FYM treatment. In case of foliar nutrition, the maximum plant spread (49.19 cm) was recorded in spraying of 0.5 per cent 19:19:19 NPK at 30 and 45 days after transplanting (T_3), which was at par with 1.0 per cent 19:19:19 NPK at 30 days after transplanting (T_4). The plant spread also significantly differed in interaction and significantly maximum plant spread (50.98 cm) was recorded in application of vermicompost and spraying of 1.0 per cent 19:19:19 NPK at 30 days after transplanting (M_2T_4), which was at par with M_2T_3 treatment combination, *i.e.* application of vermicompost and spraying of 0.5 per cent 19:19:19 NPK at 30 days and 45 days after transplanting. The lowest plant spread (42.15 cm) was recorded in application of FYM without foliar nutrition. (M_1T_1). The increased growth parameters might be associated with combined effect of organic manures and foliar nutrition.

The significantly higher number of branches plant^{-1} (4.28) was recorded in vermicompost treatment than FYM treatment. The maximum branches (4.44 plant^{-1}) was observed in plants with spraying of 0.5 per cent 19:19:19 NPK at 30 and 45 days after transplanting (T_3), which was at par with 1.0 per cent 19:19:19 NPK at 30 days after transplanting (T_4). In interaction, significantly maximum branches (4.56 plant^{-1}) was recorded in M_2T_3 treatment combination *i.e.* application of vermicompost and spraying of 0.5 per cent at 30 days and 45 days after transplanting. The minimum number of branches plant^{-1} (3.13) was noticed in application of FYM without foliar nutrition (M_1T_1). This may be attributed to higher levels of nutrients enhancing photosynthetic and other metabolic activities leading to an increase in various plant metabolites responsible for cell division. The results are analogues with Rahman *et al.* (1998) and Narayanamma *et al*. (2006).

The data on effect of organic manures and foliar nutrition on weight and size of brinjal fruits are presented in table 3. The organic manures and foliar nutrition showed the significant effect on weight, length and diameter of brinjal fruits, while interaction showed the non significant differences. In case of organic manures, the highest fruit weight, length and diameter 90.70 g, 14.14 cm and 4.92 cm, respectively, was recorded in vermicompost treatment. Among the different foliar nutrition treatments, the maximum fruit weight, length and diameter 92.84 g, 14.82 cm and 5.07 cm, respectively was recorded in spraying of 1.0 per cent at 30 days after transplanting (T_4), followed by 0.5 per cent 19:19:19 NPK at 30 days and 45 days after transplanting (T_3). The increase in fruit weight and fruit volume might be due to the better utilization of photosynthates and increased allocation of photosynthates towards the economic parts. These findings are also in conformity with the results of Narayanamma *et al.* (2006) and Ullah *et al.* (2008).

Table 2. Effect of organic manures and foliar nutrition on growth of brinjal

Treatment	Plant height (cm)			Average plant spread (cm)			No. of branches plant^{-1}		
	M_1	M_2	Mean	M_1	M_2	Mean	M_1	M_2	Mean
T_1	85.75	90.81	88.28	42.15	44.33	43.24	3.13	4.25	3.69
T_2	92.95	95.63	94.29	46.48	45.23	45.85	4.38	4.00	4.19
T_3	97.43	113.50	105.46	48.30	50.08	49.19	4.31	4.56	4.44
T_4	101.83	104.19	103.01	46.45	50.98	48.71	4.19	4.31	4.25
Mean	94.49	101.03	97.76	45.84	47.65	46.75	4.00	4.28	4.14
For comparing means of	SE±	C.D. (5%)		SE±	C.D. (5%)		SE±	C.D. (5%)	
Organic manures (M)	0.95	3.04		0.54	1.63		0.08	0.25	
Foliar nutrition (T)	0.45	1.39		0.44	1.35		0.13	0.40	
M X T	0.90	2.77		0.87	2.69		0.16	0.49	

Table 3. Effect of organic manures and foliar nutrition on fruit weight and size of brinjal

Treatment	Fruit weight (g)			Fruit length (cm)			Fruit diameter (cm)		
	M_1	M_1	Mean	M_1	M_2	Mean	M_1	M_2	Mean
T_1	85.94	87.31	86.63	12.71	13.16	12.94	4.12	4.61	4.37
T_2	88.00	89.81	88.91	13.30	13.81	13.56	4.53	4.87	4.70
T_3	90.75	92.25	91.50	13.81	14.55	14.18	5.00	5.10	5.05
T_4	92.25	93.44	92.84	14.59	15.05	14.82	5.04	5.10	5.07
Mean	89.23	90.70	89.97	13.60	14.14	13.87	4.67	4.92	4.80
For comparing means of	SE±	C.D. (5%)		SE±	C.D. (5%)		SE±	C.D. (5%)	
Organic manures (M)	0.45	1.39		0.12	0.38		0.09	0.29	
Foliar nutrition (T)	1.40	4.48		0.26	0.84		0.06	0.19	
M X T	0.90	NS		0.25	NS		0.12	NS	

Data on yield plant^{-1} and hectare^{-1} as influenced by the organic manures and foliar nutrition and their interaction are furnished in table 4. The application of FYM and vermicompost showed the significant difference in yield of brinjal. The significantly higher fruit yield (1.22 kg plant^{-1} and 275.08 q ha^{-1}) was recorded in vermicompost treatment (M_2), where 10.70 per cent higher yield was found than FYM treatment. Among the different foliar nutrition treatments, the significantly higher fruit yield (1.40 kg plant^{-1} and 323.11 q ha^{-1}) was recorded in spraying of 0.5 per cent 19:19:19 NPK at 30 days and 45 days after transplanting (T_3), followed by1.0 per cent 19:19:19 NPK at 30 days after transplanting (T_4). The interaction effect was also significant and significantly maximum fruit yield (1.54 kg plant^{-1} and 346.65 q ha^{-1}) was recorded in M_2T_3 treatment combination *i.e.* application of vermicompost and spraying of 0.5 per cent 19:19:19 NPK at 30 days and 45 days after transplanting. The minimum fruit yield (0.88 kg plant^{-1} and 197.44 q ha^{-1}) was noticed in application of FYM without foliar nutrition (M_1T_1). This might be due to synergic effect of organic manure and foliar nutrition. Higher yield may also be attributed to satisfactory nutrient availability and increased plant metabolism, which ultimately lead to more carbohydrates accumulation. Besides these, increased vegetative growth at peak growth stage might have resulted in higher yield. This result is in agreement with the observations of Devi *et al.* (2002) and Ullah *et al.* (2008).

From the above findings, it can clearly be inferred that application of vermicompost and spraying of 0.5 per cent 19:19:19 NPK at 30 days and 45 days after transplanting in brinjal recorded the highest plant growth characters like height, spread and branching with large sized fruits. This treatment combination was also beneficial for maximizing the yield of brinjal fruits.

Table 4. Effect of organic manures and foliar nutrition on yield of brinjal

Treatment	Yield plant^{-1} (kg)			Yield (q ha^{-1})		
	M_1	M_2	Mean	M_1	M_2	Mean
T_1	0.88	1.00	0.94	197.44	225.31	211.38
T_2	1.02	1.04	1.03	231.00	233.44	232.22
T_3	1.26	1.54	1.40	299.56	346.65	323.11
T_4	1.17	1.31	1.24	266.01	294.91	280.46
Mean	1.08	1.22	1.15	248.50	275.08	261.79
For comparing means of	SE±	C.D. (5%)		SE±	C.D. (5%)	
Organic manures (M)	0.04	0.12		7.84	25.10	
Foliar nutrition (T)	0.02	0.06		3.03	9.33	
M X T	0.04	0.11		6.06	18.66	

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Interaction effect of consortium of *Azospirillum*, PSB and AM fungus with reduced levels of N & P fertilizers on growth of direct seeded rice

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ABSTRACT

The efficient strains of *Azospirillum*, phosphate solubilizing bacteria (PSB) and AM fungus were used to inoculate as single, dual and combined in direct seeded rice (DSR). The field experiment was laid out in randomised block design (RBD) with 10 treatments and 3 replications. Among all the treatments, the combined inoculation of *Azospirillum*, PSB and AM fungus with 75% of recommended doses of N and P performed better with respect to number of leaves (95.67), root dry matter (15.87 g hill^{-1}), stem dry matter (11.75 g hill^{-1}), leaves dry matter (11.75 g hill^{-1}), total dry matter (39.37g hill^{-1}), root length density (7.53 g/cm^3) and biomass yield (242.00 g hill^{-1}) at harvest while the other growth parameter *viz*., leaf area (831.67 cm^2 hill^{-1}) and leaf area index (4.153) showed maximum at 90 DAS of the DSR compared to other inoculated treatments, RDF application and uninoculated control.

Key words: AM fungi, *Azospirillum,* DSR and PSB.

Rice (*Oryza sativa* L.) is the second most important cereal in the world after wheat, and the principal crop in Asia, serving as food for about 50 per cent of the world's population. Among the rice growing countries, India ranks first in area followed by China and Bangladesh. In Karnataka, rice is cultivated in an area of 1.33 m ha with an annual production of 3.76 m t and productivity of 2827 kg ha^{-1} (Anon, 2014).

Currently, growth of rice plants uses a lot of chemical fertilizer and chemical pesticide. Moreover, rice is grown in flooded conditions and uses lots of irrigated water. Over the years this has led to serious environmental problems such as depletion of soil quality and health, increasing salinity, emergence of resistant pathogens and elimination of soil microbes. The increase of production of rice must be achieved through improvement in agricultural productivity. Microbes are the beneficial key factor in maintaining soil quality by decreasing the dose of chemical fertilizers and increasing the crop production. The current scientific challenge is to find physiological and agronomic systems in crop establishment under direct seeded rice (DSR).

Azospirillum represents the main group of microaerophilic associative nitrogen fixing bacteria (Dobereiner and Day, 1976). Associative nitrogen fixing bacteria colonize endorhizosphere, and fix atmospheric nitrogen. They have significant potential for commercial applications as biofertilizers (Bashan and Holguin, 1997). The phosphorus solubilising microorganisms (PSB) include different groups of microorganisms, which not only assimilate phosphorus from insoluble forms of phosphates, but also cause a large portion of soluble phosphates to be released in quantities in excess of their requirements. Arbuscular mycorrhizal (AM) symbiosis is a mutualistic association between vast range of terrestrial plants and a class of fungi (Glomeromycota) which occurs in the root zone of plants. This association aid in plant mineral nutrition and plant health with a wide range of applications in sustainable agricultural systems. With the use of AM fungi, it is possible to increase plant water acquisition and/or drought tolerance (Ferrol *et al*., 2002).

The success of the microbe-microbe-plant interaction depends on the survival and persistence of the microorganisms in soil and the effective colonization in the rhizosphere. If two or more microorganisms are used, may lead to synergistic effect, which may have direct reflection on plant growth and yield. Further, DSR is irrigated in rows as done in field crops. Interaction between *Azospirillum*, PSB and AM fungi may stimulate plant growth in a positive manner like plant growth promotion, root colonization and disease suppression in direct seeded rice. Further, the evaluation was carried out with 75 per cent of recommended doses of fertilizers in biofertilizer inoculated treatments and compared with RDF alone and uninoculated control.

MATERIALS AND METHODS

Collection and maintenance of pure culture of bioinoculants

Pure cultures of *Azospirillum* (*A. brasilense,* ADSR-9), PSB (*Bacillus megaterium* var. *phosphaticum*) and AM fungus

(*Glomus* sp., MDSR-3) were maintained and obtained from Department of Agricultural Microbiology, College of Agriculture, Bheemarayangudi, University of Agricultural Sciences, Raichur. The efficient strains of *Azospirillum* and AM fungus have been isolated from rhizosphere of DSR and screened *in vitro* for their efficiency in the department earlier.

Field experiment

The field experiment was conducted to know the influence of efficient strain of *Azospirillum*, phosphate solubilizing bacteria (PSB) and AM fungus on growth of direct seeded rice. The experiment was laid out in randomized block design (RBD) with 10 treatments and 3 replications.

The treatment details were as follows:

Treatment	Treatment detail
T_1	Uninoculated Control
T_2	Recommended dose of fertilizers (100% NPK)
T_3	75% N + 75% P + 100% K
T_4	*Azospirillum* + 75% N + 75% P + 100% K
T_5	PSB + 75% N + 75% P + 100% K
T_6	AMF + 75% N + 75% P + 100% K
T_7	*Azospirillum* X PSB + 75%N+75%P+100%K
T_8	*Azospirillum* X AMF + 75% N + 75% P + 100% K
T_9	PSB X AMF + 75 % N + 75% P + 100% K
T_{10}	*Azospirillum* X PSB X AMF + 75% N + 75% P + 100% K

Growth attributes

Five plants were randomly selected in each treatment for recording various observations on growth parameters at 45, 90 DAS and at harvest.

Dry matter distribution in different plant parts

Five plants were uprooted at random from adjacent to net plot area excluding two border rows in the field. Leaves, stem and root portions were separated. The samples were dried in hot air oven at 65°C for 72 hours until constant weight. The completely dried samples were weighed and the dry weight of different plant parts was expressed in g hill^{-1}.

Total dry matter production and its distribution in different plant parts

The total dry matter production hill^{-1} was obtained with the summation of leaves, stem and root dry weight and was expressed in g hill$^{-1.}$

Number of leaves hill^{-1}

Total numbers of fully opened leaves from five tagged plants or hills were counted and mean hill^{-1} was computed.

Leaf area hill^{-1}

For measuring leaf area, number of leaves hill^{-1} was counted. The length and maximum width of each leaf on the middle tiller was measured and leaf area of each leaf was computed as follows.

Leaf area = K x L x W

Where, K = constant factor; L = length of leaf (cm); W = maximum width of leaf (cm)

The value of constant K was taken as 0.75 upto 90 days and 0.67 during maturity stage (Gomez, 1972). The leaf area hill^{-1} was then calculated as follows.

Leaf area hill^{-1} = total leaf area of middle tiller x total number of tillers hill^{-1}.

It was recorded for five hills separately and averaged to get leaf area in cm^2 hill^{-1}.

Leaf area index

Leaf area index is defined as the leaf area per unit land area. It was worked out by the formula given by Watson (1952).

$$LAI = \frac{A}{P}$$

Where,
LAI = Leaf area index
A = Leaf area (cm^2)
P = Land area (cm^2)

Root length density

Root length density was recorded at 45, 90 DAS and at harvest by uprooting plants carefully and measuring the root length density from tip of the longest root to the neck region and expressed in g/cm^3.

Biomass yield per hill

All the plants from 1m row length were uprooted at 45, 90 DAS and at harvest and fresh plants were weighed to determine the total biomass yield and expressed in g/hill.

RESULTS AND DISCUSSION

Dry matter distribution in different plant parts

The maximum dry weight of root (15.87 g hill^{-1}), stem (11.75 g hill^{-1}), leaf (11.75 g hill^{-1}) and total dry matter (39.37 g hill^{-1}) of DSR at harvest were recorded in treatment T_{10} (*Azospirillum* X PSB X AMF + 75% N + 75% P + 100% K) followed by T_9 (PSB X AMF + 75% N + 75% P + 100% K), which recorded root dry matter of 14.80 g hill^{-1}, stem dry

matter of 10.65 g hill^{-1}, leaf dry matter of 10.65 g hill^{-1} and total dry matter of 36.10 g hill^{-1}. The uninoculated control recorded the lowest root dry matter of 4.70 g hill^{-1}, stem dry matter of 8.10 g hill^{-1}, leaf dry matter of 7.52 g hill^{-1}, and total dry matter of 20.32 g hill^{-1} (Table 1). Similar results were noticed on growth parameters by several workers. Goussous and Mohammad (2009) reported that the greater dry weight of onion plants which were colonized by AM1, a mixture of indigenous mycorrhizal fungi and AM2, *Glomus intraradices*. The synergistic interaction between AMF and N_2 fixers in the rhizosphere soil of DSR under pot culture condition might help in increasing the availability of N and P. Further, it may also help in production of growth promoting hormones. Sivakumar and Thanizhiniyan (2012) reported the influence of AM fungi and *Azospirillum* on the growth, yield and nutritional potential of tomato. The results showed highest root length, shoot length, fresh weight, dry weight at various stages of growth (35, 50 and 75 days).

Number of leaves

Combined inoculation of *Azospirillum*, PSB and AM fungus with 75% N, 75% P and 100% K (T_{10}) recorded highest number of leaves (95.67) followed by T_9 which recorded 89.33 leaves at harvest. The lowest number of leaves (61.67) was recorded in uninoculated control (Table 2). Similar results were noticed on growth by Chandrashekar (2003) who observed that the plant growth parameters *viz*., shoot and root length and number of leaves per plant in green gram plants at 45 days after sowing significantly increased due to inoculation of P-solubilizing fungal strains along with rock phosphate application as compared to rock phosphate alone (control).

Leaf area hill^{-1} and Leaf area index

At 90 DAS, combined inoculation of *Azospirillum*, PSB and AM fungi with 75% N, 75% P and 100% K significantly recorded maximum leaf area hill^{-1} and leaf area index. During this time, the seeds treated with T_{10} recorded highest leaf area hill^{-1} (831.67 cm^2) and leaf area index (4.153) followed by T_9, which recorded leaf area of 823.67 cm^{2}hill^{-1} and leaf area index of 4.113. The lower leaf area (754.67 cm^2 hill^{-1}) and leaf area index (3.776) was recorded in uninoculated control (Table 2). The declining trend was observed after 90 DAS. Priya and Geetham (2015) reported that co-inoculation of *Azospirillum* and phosphate solubilizing bacteria showed a significant performance in number of leaves, length and breadth of the leaves (leaf area), shoot length, plant height, root length, dry weight and number of pods (yield) when compared with single inoculations and control.

Root length density

The seeds treated with T_{10} recorded highest root length density (7.53 g/cm^3) followed by T_9 which recorded 6.91g/cm^3 at harvest. The lowest root length density (2.10g/cm^3 at harvest) was recorded in uninoculated control (Table 3). Vasanthakumar (2003) reported that combined inoculation of *Azospirillum* (AZUS10) and PSB isolate (PSB7) produced synergistic effect, resulting in increased root length, shoot length, stem girth, number of leaves and number of branches in solanaceous crop plants.

Biomass yield

The seeds treated with T_{10} recorded highest biomass yield (242.00 g hill^{-1}) followed by T_9 which recorded 235.70 g hill^{-1} at harvest. The lowest biomass yield (139.60 g hill^{-1} at

Table 1. Effect of *Azospirillum*, PSB and AM fungi on dry matter distribution of roots, stem, leaves and total dry matter in direct seeded rice

Treatment	Dry matter of roots g hill^{-1}			Dry matter of stem g hill^{-1}			Leaves dry matter g hill^{-1}			Total dry matter g hill^{-1}		
	45 DAS	90 DAS	At harvest	45 DAS	90 DAS	At harvest	45 DAS	90 DAS	At harvest	45 DAS	90 DAS	At harvest
T_1	1.44^{e}	2.50^{d}	4.70^{d}	1.32^{g}	3.86^{j}	8.10^{j}	1.35^{f}	3.45^{j}	7.52^{h}	4.10^{i}	9.81^{j}	20.32^{g}
T_2	1.69^{cde}	2.90^{d}	5.60^{d}	1.44^{f}	5.13^{h}	8.62^{h}	1.40^{f}	5.10^{h}	8.25^{g}	4.53^{gh}	13.12^{h}	22.47^{f}
T_3	1.58^{de}	2.80^{d}	4.93^{d}	1.43^{f}	4.79^{i}	8.16^{i}	1.36^{f}	4.43^{i}	7.67^{h}	4.37h^{i}	12.02^{i}	20.76^{g}
T_4	1.70^{cde}	3.10^{d}	5.63^{d}	1.44^{f}	6.02^{g}	9.11^{g}	1.64^{e}	5.66^{g}	8.57^{f}	4.78f^{g}	14.78^{g}	23.31^{f}
T_5	1.94^{cd}	4.10^{c}	8.10^{c}	1.47^{f}	6.53^{f}	9.33^{f}	1.68^{e}	6.15^{f}	9.10^{e}	5.09^{f}	16.78^{f}	26.52^{e}
T_6	2.14^{c}	4.60^{bc}	9.27^{b}	1.71^{e}	6.61^{e}	9.52^{e}	2.02^{d}	6.60^{e}	9.47^{d}	5.86^{e}	17.81^{e}	28.26^{d}
T_7	2.81^{b}	5.30^{b}	10.10^{b}	3.07^{c}	9.23^{c}	10.28^{c}	2.78^{b}	8.90^{c}	10.37^{b}	8.66^{c}	23.43^{c}	30.75^{c}
T_8	2.64^{b}	4.60^{c}	9.43^{b}	2.31^{d}	8.02^{d}	9.62^{d}	2.43^{c}	7.45^{d}	9.77^{c}	7.38^{d}	20.07^{d}	28.82^{d}
T_9	3.64^{a}	7.20^{a}	14.80^{a}	3.70^{b}	9.76^{b}	10.65^{b}	2.84^{b}	9.26^{b}	10.65^{b}	10.18^{b}	26.22^{b}	36.10^{b}
T_{10}	3.90^{a}	7.80^{a}	15.87^{a}	4.71^{a}	10.26^{a}	11.75^{a}	4.72^{a}	9.95^{a}	11.75^{a}	13.33^{a}	28.01^{a}	39.37^{a}
SE.M±	0.03	0.06	0.13	0.03	0.10	0.13	0.03	0.09	0.13	0.10	0.26	0.39
CD (0.05)	0.10	0.19	0.39	0.10	0.29	0.41	0.10	0.28	0.40	0.31	0.78	1.17

Note: DAS: Days after sowing; Values are mean of three replications; Mean values followed by the same letter are not significantly different based on Duncan's multiple range test ($p<0.05$), a> b > c.

Table 2. Effect of *Azospirillum*, PSB and AM fungi on number of leaves and leaf area

Treatment	No. of leaves hill^{-1}			Leaf area hill^{-1} (cm^2)			Leaf area index		
	45 DAS	90 DAS	At harvest	45 DAS	90 DAS	At harvest	45 DAS	90 DAS	At harvest
T_1 -	19.00^{d}	41.67^{e}	61.67^{d}	145.14^{bcd}	754.67^{j}	215.67^{g}	0.713^{f}	3.776^{c}	1.056^{g}
T_2 -	30.00^{c}	47.67^{d}	64.00^{cd}	159.03^{bcd}	766.33^{h}	222.67^{f}	0.796^{e}	3.840^{c}	1.176^{e}
T_3 -	23.00^{d}	43.33^{de}	63.67^{cd}	156.00^{d}	761.67^{i}	221.67^{f}	0.786^{e}	3.806^{c}	1.103^{fg}
T_4 -	30.00^{c}	56.00^{c}	68.33^{cd}	161.67^{cd}	771.33^{g}	225.67^{e}	0.803d^{e}	3.846^{c}	1.133^{ef}
T_5 -	31.00^{bc}	56.33^{c}	68.67^{cd}	161.67^{bcd}	776.00^{f}	229.00^{d}	0.806d^{e}	3.886^{bc}	1.173^{e}
T_6 -	35.67^{abc}	59.33^{bc}	71.00^{c}	167.67^{abc}	781.00^{e}	231.67^{cd}	0.836^{cd}	3.906^{bc}	1.393^{d}
T_7 -	37.33^{a}	64.00^{a}	85.67^{b}	173.00^{abc}	803.00^{c}	234.00^{bc}	0.856^{bc}	4.056^{ab}	1.393^{d}
T_8 -	36.00^{ab}	62.67^{ab}	71.33^{c}	169.67^{abc}	792.00^{d}	234.00^{bc}	0.836^{cd}	4.056^{ab}	1.596^{c}
T_9 -	38.33^{a}	64.67^{a}	89.33^{ab}	177.33^{ab}	823.67^{b}	235.00^{b}	0.886^{b}	4.113^{a}	1.766^{b}
T_{10} -	38.67^{a}	65.00^{a}	95.67^{a}	185.33^{a}	831.67^{a}	239.00^{a}	0.926^{a}	4.153^{a}	1.896^{a}
SE.M±	0.47	0.79	1.06	0.91	1.05	0.96	0.012	0.059	0.020
CD (0.05)	1.42	2.36	3.17	18.57	3.11	2.86	0.034	0.179	0.050

Note: DAS: Days after sowing; Values are mean of three replications; Mean values followed by the same letter are not significantly different based on Duncan's multiple range test ($p<0.05$), a> b > c and leaf area index in direct seeded rice

Table 3. Effect of *Azospirillum*, PSB and AM fungi on root length density and biomass yield in direct seeded rice.

Treatment	Root length density (g/cm^3)			Biomass yield (g hill^{-1})		
	45 DAS	90 DAS	At harvest	45 DAS	90 DAS	At harvest
T_1	0.39^{d}	1.20^{e}	2.10^{f}	26.90^{e}	70.33^{c}	139.60^{e}
T_2	1.30^{bc}	2.92^{cd}	5.20^{d}	44.11^{cde}	83.20^{c}	167.50^{cde}
T_3	1.10^{cd}	2.10^{de}	3.01^{e}	36.80^{de}	70.80^{c}	144.90^{de}
T_4	1.39^{bc}	3.30^{bc}	5.30^{d}	45.90^{cd}	91.94^{bc}	182.60^{bcd}
T_5	1.40^{bc}	3.54^{abc}	5.50^{cd}	46.83^{cd}	92.40^{bc}	183.50^{bcd}
T_6	1.47^{bc}	3.64^{abc}	6.00^{cd}	47.80^{cd}	92.56^{bc}	193.00^{bc}
T_7	1.80^{abc}	4.03^{ab}	6.90^{ab}	58.80^{bc}	94.30^{bc}	213.40^{ab}
T_8	1.80^{abc}	3.70^{abc}	6.32^{bc}	48.60^{cd}	92.70^{bc}	194.80^{bc}
T_9	1.90^{ab}	4.40^{a}	6.91^{ab}	69.44^{ab}	120.20^{ab}	235.70^{a}
T_{10}	2.50^{a}	4.55^{a}	7.53^{a}	80.80^{a}	128.50^{a}	242.00^{a}
SE.M±	0.30	0.40	0.30	6.10	9.50	13.60
CD (0.05)	0.79	1.11	0.87	18.21	28.30	40.15

Note: DAS: Days after sowing; Values are mean of three replications; Mean values followed by the same letter are not significantly different based on Duncan's multiple range test ($p<0.05$), a> b > c.

harvest) was recorded in uninoculated control (Table 3). Similar result was given by the response of rice plants to inoculation with an Arbuscular Mycorrhizal (AM) fungus *Glomus intraradices*, *Azospirillum brasilense* or combination of both microorganisms, when assayed under well-watered or drought stress conditions. Water deficit treatment was imposed by reducing the amount of water added, but AM plants, with a significantly higher biomass, received the same amount of water as non-AM plants, with a poor biomass (Ruiz-Sancheza *et al*., 2011).

The concept of biofertilizers was developed based on the observation that these microorganisms can have a beneficial effect on plant and crop growth and keep the soil healthy for future. Based on these reports, it can be assumed that biofertilizers could offer an opportunity for DSR farmers to increase productivity, and resource use efficiency. And, the increasing availability of biofertilizers in many countries and regions and the sometimes aggressive marketing brings ever more farmers into contact with this technology.

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Microbial population and nutrient dynamics in rhizosphere soil of direct seeded rice

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ABSTRACT

A field experiment was undertaken to investigate the microbial population and nutrient dynamics in rhizosphere soil of direct seeded rice (DSR). The research was conducted at ARS, Dhadesugur during the year 2015-16. The experiment was laid out in randomized block design with ten treatments and three replications. Among all the treatments, combined inoculation of *Azospirillum* + PSB + AMF + 75% N + 75% P + 100% K recorded significantly higher soil microbial population with respect to bacteria (6.04 x 10^6 cfu g^{-1} dry soil), fungi (7.89 x 10^3 cfu g^{-1} dry soil), actinobacteria (7.11 x 10^4 cfu g^{-1} dry soil), *Azospirillum* (39.30 x 10^2 cfu g^{-1} roots), PSB (8.01 x 10^3 cfu g^{-1} dry soil) compared to individual and dual inoculation. Similarly, per cent root colonization, spore count of AM fungi, soil biomass carbon and chemical properties of soil performed better with combined inoculation of efficient strains of *Azospirillum,* PSB and AM fungus as compared to other treatments and uninoculated control.

Key words: *Azospirillum*, PSB, AM fungi, DSR

Rice (*Oryza sativa* L.) is the second most important cereal in the world after wheat and the principal crop in Asia, serving as food for about 50 per cent of the world's population. It occupies an area of 153.76 m ha with an annual production of 598.85 mt with a productivity of 3895 kg ha^{-1} in the world. Asia produces and consumes 90 per cent of world's rice. Among the rice growing countries, India ranks first in area followed by China and Bangladesh with an area of 43.95 m ha and production of 106.54 mt with an average productivity of 2424 kg ha^{-1}. In Karnataka, rice is cultivated in an area of 1.33 m ha with an annual production of 3.76 mt and productivity of 2827 kg ha^{-1} (Anonymous, 2014).

Microorganisms play major role in the availability of nutrients. Among the major nutrients, nitrogen and phosphorus are most vulnerable for microbial transformation (Alexander, 1961). The success of the microbe-plant interaction depends on the survival and persistence of the microorganisms in soil and the effective colonization in the rhizosphere. If two or more microorganisms are used in combination, it may lead to synergistic effect, which may have direct reflection on plant growth and yield. Hence, the present investigation aims at the field evaluation of efficient *Azospirillum*, phosphate solubilising bacteria and AM fungus on growth and yield of direct seeded rice (DSR), with an objective to know the microbial population and nutrient dynamics in rhizosphere soil of DSR.

MATERIALS AND METHODS

A field experiment was carried out at Agricultural Research Station, Dhadesugur, University of Agricultural Sciences, Raichur, Karnataka, India during 2015-16 to know the microbial population and nutrient dynamics in rhizosphere soil of DSR. All the parameters were analyzed before sowing and during crop growth (45, 90 days after sowing and at harvest). The isolates of *Azospirillum* and AM fungi, which fared best under pot cultural studies in DSR carried out earlier in the department *viz*., ADSR-9 (*Azospirillum brasilense*) and MDSR-3 (*Glomus* sp.) along with PSB strain *Bacillus megatarium* var. *phasphaticum* were used individually and in combination with 25 per cent reduced levels of recommended N and P were evaluated using DSR under field conditions.

The following were the treatments and details:

Treatment		Treatment detail
T_1	-	Uninoculated Control
T_2	-	Recommended dose of fertilizers (100% NPK)
T_3	-	75% N + 75% P + 100% K
T_4	-	*Azospirillum* + 75% N + 75% P + 100% K
T_5	-	PSB + 75% N + 75% P + 100% K
T_6	-	AMF + 75% N + 75% P + 100% K
T_7	-	*Azospirillum* X PSB + 75% N + 75% P + 100% K
T_8	-	*Azospirillum* X AMF + 75% N + 75% P + 100% K
T_9	-	PSB X AMF + 75 % N + 75% P + 100% K
T_{10}	-	*Azospirillum* X PSB X AMF + 75% N + 75% P + 100% K

Biological properties

Soil samples collected from experimental field were used for enumeration of common soil microorganisms like bacteria, fungi, actinobacteria and different physiological groups such as *Azospirillum*, phosphorus solubilising bacteria (PSB) and AM fungi.

Enumeration of bacteria, fungi and actinobacteria

Each soil sample was sieved through the 2 mm sieve to remove the bigger particles and were used for isolation of bacteria, fungi and actinobacteria (NA, MRBA and KA media were used respectively) by serial dilution agar plate technique and the plates were incubated for 24 hrs., 4 days and 6 days respectively at 28°C. Colonies that appeared on media were enumerated and expressed in terms of CFU per gram of soil on dry weight basis.

Enumeration of *Azospirillum*

Before sowing, *Azospirillum* was enumerated from soil of experimental plots by serial dilution pour plate technique using malate medium with ammonium chloride. After sowing of DSR, the enumeration of *Azospirillum* was carried out using DSR root samples. Here fresh root samples of DSR were collected and washed thoroughly in running tap water and surface sterilized by dipping in 0.1 per cent $HgCl_2$ solution for three minutes followed by dipping in 70 per cent ethyl alcohol for one minute. The roots were finally washed in six times of sterile distilled water and cut into bits of 1 cm length. The root bits were then placed at subsurface level in screw capped tubes containing sterilized semisolid N free malate medium under aseptic conditions. The tubes were then incubated at 28° C for a period of one week and tubes that show growth (white, translucent, undulating, subsurface pellicles) of *Azospirillum* were selected for isolation and all the samples were serially diluted by 5th fold series and analysed for the *Azospirillum* by MPN method using semi solid malate medium. The MPN counts of *Azospirillum* were taken for root samples collected from every replication and mean value of three replications was taken in the table 2.

Enumeration of PSB

The phosphate solubilizing bacteria (PSB) were isolated from rhizosphere soil of DSR by dilution plating technique on Pikovskaya's agar medium (Pikovskaya, 1948) containing tricalcium phosphate (TCP). The plates were incubated at 28°C for two to seven days. Detection and estimation of the phosphate solubilisation ability of microorganisms have been possible using plate screening methods. Phosphate solubilizers produce clear halo zones around the microbial colonies in media supplemented with insoluble mineral phosphates such as tricalcium phosphate or hydroxyapatite.

Per cent root colonization and spore count of AM fungi

The soil samples were collected at different stages for per cent root colonization studies. From each treatment, individual plant was uprooted and washed with water and root part was taken for estimation of root colonization by AM fungi and it was expressed in percentage. Likewise, the number of spores were counted treatment wise at different stages and was expressed as number of spores 50^{-1} g of soil.

Soil biomass carbon

Soil biomass carbon was determined by chloroform fumigation cum incubation method (Jenkinson and Ladda, 1981). Soil biomass carbon estimation was based on the principle that moist soil was exposed to ethanol free chloroform for twenty four hours, the fumigant was removed by repeated evacuation and then the soil was inoculated and incubated at 28°C for ten days at fifty per cent of its water holding capacity. Based on the difference between the quality of CO_2 evolved by fumigated and that of un-fumigated soils, the soil biomass carbon was estimated before sowing and at different stages of DSR crop growth.

Organic carbon

A composite soil sample from the field experimentation were dried under shade, powdered using pestle and mortar and passed through 0.2 mm sieve for analysis (Jackson, 1973). A known weight of soil was treated with excess volume of potassium dichromate solution in the presence of concentrated H_2SO_4 and organic carbon in the soil was oxidized to CO_2. The excess of potassium dichromate unused was titrated back against ferrous ammonium sulphate in the presence of concentrated phosphoric acid and diphenyl amine indicator.

Estimation of available nitrogen, phosphorus and potassium

The alkaline potassium permanganate method of Subbaiah and Asija (1956) was used for the estimation of available N content in soil. A known weight of soil was treated with excess of alkaline 0.32 per cent potassium permanganate (made alkaline with 25% NaOH solution). The liberated ammonia was trapped in boric acid and determined by titration against standard H_2SO_4. The available N content in kg ha^{-1} was computed using titre value. The Phosphorus content was determined by chloromolybdic blue colour method using UV spectrophotometer (Jackson, 1973). Available phosphorus content of the soil was extracted by using 0.5M $NaHCO_3$ and intensity of blue colour was determined at 660 nm. The available phosphorus content was computed and expressed in kg ha^{-1}. Available potassium content from soil was extracted by using neutral N NH_4OAC as described by Jackson (1973). The concentration of potassium in the extractant was determined by flame photometer.

RESULTS AND DISCUSSION

General and beneficial soil microflora

The initial soil microbial populations *viz*., bacteria, fungi, actinobacteria, *Azospirillum*, PSB, AM fungi and soil biomass carbon in the experimental site were non-significant in different treatment plots.

During the growth stages of DSR, microbial population increased with the age of host upto 90 DAS but showed declining trend at the harvest stage. The results indicated that, at 90 DAS, combined inoculation of efficient strains of *Azospirillum*, PSB and AM fungus (T_{10}) has recorded highest population of bacteria (6.04 x 10^6 cfu g^{-1} dry soil), fungi (7.89 x 10^3 cfu g^{-1} dry soil), actinobacteria (7.11 x 10^4 cfu g^{-1} dry soil) (Table 1), *Azospirillum* (39.30 x 10^2 cfu g^{-1} roots) and PSB (8.01 x 10^3 cfu g^{-1} dry soil) (Table 2).

Percentage root colonization and spore count of AM fungi

The per cent root colonization showed an increasing trend upto 90 DAS but it showed a decline at harvest stage. The percentage root colonization in the rhizosphere soil of DSR were significantly highest due to combined inoculation of *Azospirillum*, PSB and AM fungus (45.30, 59.40 and 35.00 at 45, 90 DAS and at harvest respectively) as compared to dual treatment combinations, individual inoculation and uninoculated control (Table 3).

Table 1. Effect of inoculation of *Azospirillum*, PSB and AM fungus on viable population of bacteria, fungi and actinobacteria in the rhizosphere soil of direct seeded rice

Treatment	Viable bacteria (CFUx10^6 g^{-1} of dry soil)			Viable fungi (CFUx10^3 g^{-1} of dry soil)			Viable actinobacteria (CFUx10^4 g^{-1} of dry soil)		
	45 DAS	90 DAS	At harvest	45 DAS	90 DAS	At harvest	45 DAS	90 DAS	At harvest
T_1	3.44^{c}	4.10^{f}	3. 05^{d}	3.70^{d}	4.08^{h}	3.05^{g}	2.98^{g}	4.18^{e}	3.11^{g}
T_2	3.90^{b}	4.15^{e}	3. 98^{b}	3.87^{d}	5.51^{ef}	4.22^{f}	3.92^{f}	5.54^{d}	4.89^{ef}
T_3	3.83^{b}	4.11^{f}	3.50^{c}	3.80^{d}	5.40^{f}	4.06^{f}	3.78^{f}	5.43^{d}	4.73^{f}
T_4	4.07^{b}	5.45^{d}	4.65^{a}	4.89^{c}	5.75^{de}	4.68d^{e}	5.02^{e}	6.42^{bc}	5.73^{b}
T_5	4.01^{b}	5.54^{cd}	4.55^{a}	4.90^{c}	5.82^{d}	4.58^{e}	5.20^{de}	6.29^{bc}	5.70^{b}
T_6	4.10^{b}	5.69^{cd}	4.54^{a}	4.98^{c}	5.10^{g}	4.80^{d}	5.29^{cd}	6.25^{c}	5.12^{cd}
T_7	4.05^{b}	5.65^{cd}	4.61^{a}	4.97^{c}	5.95^{d}	4.75^{de}	5.35^{cd}	6.53^{b}	5.04^{de}
T_8	4.15^{b}	5.75^{bc}	4.63^{a}	4.92^{c}	6.49^{c}	5.41^{c}	5.43^{c}	6.46^{bc}	5.31^{c}
T_9	4.12^{b}	5.95^{ab}	4.66^{a}	5.41^{b}	6.89^{b}	5.71^{b}	5.84^{b}	6.95^{a}	5.99^{a}
T_{10}	5.31^{a}	6.04^{a}	4.72^{a}	5.90^{a}	7.89^{a}	6.42^{a}	6.10^{a}	7.11^{a}	6.20^{a}
SE.M±	0.08	0.07	0.06	0.07	0.08	0.06	0.06	0.08	0.07
CD (0.05)	0.29	0.22	0.19	0.19	0.26	0.20	0.20	0.26	0.22

Note: RDF for rice is 150 : 75 : 37.50 kg NPK ha^{-1}, Values are mean of three replications; Mean values followed by the same letter are not significantly different based on Duncan's multiple range test ($p<0.05$), a> b > c.

Table 2. Effect of inoculation of *Azospirillum*, PSB and AM fungus on MPN counts of *Azospirillum* and phosphate solubilising bacterial population in rhizosphere soil of direct seeded rice

Treatment	MPN of *Azospirillum* (CFU x 10^2 g^{-1} of roots)			Viable PSB (CFU x 10^3 g^{-1} of dry soil)		
	45 DAS	90 DAS	At harvest	45 DAS	90 DAS	At harvest
T_1	14.70^{d}	24.70^{b}	13.66^{d}	3.22^{f}	4.03^{g}	3.69^{h}
T_2	15.00^{cd}	26.70^{b}	17.00^{cd}	4.15^{d}	5.41^{f}	4.08^{g}
T_3	14.70^{d}	25.70^{b}	16.70^{cd}	3.68^{e}	5.29^{f}	4.99^{f}
T_4	20.00^{b}	36.70^{a}	21.66^{abc}	5.47^{c}	6.25^{de}	5.73^{d}
T_5	18.70^{bcd}	27.70^{b}	18.66^{bcd}	5.36^{c}	6.18^{e}	5.77^{d}
T_6	15.33^{cd}	27.30^{b}	17.66^{bcd}	5.28^{c}	6.48^{d}	5.45^{e}
T_7	22.70^{ab}	38.00^{a}	22.66^{ab}	5.64^{c}	6.38^{de}	5.77^{d}
T_8	21.70^{ab}	37.00^{a}	22.00^{abc}	6.14^{b}	7.07^{c}	6.35 ^{c}
T_9	19.00^{bc}	35.00^{a}	20.00^{bc}	6.94^{a}	7.67^{b}	6.97^{b}
T_{10}	24.33^{a}	39.30^{a}	25.33^{a}	7.10^{a}	8.01^{a}	7.77^{a}
SE.M±	0.26	0.44	0.26	0.12	0.09	0.07
CD (0.05)	077	1.31	0.77	0.38	0.26	0.22

Note: RDF for rice is 150:75:37.50 kg NPK ha^{-1}; Values are mean of three replications; Mean values followed by the same letter are not significantly different based on Duncan's multiple range test ($p<0.05$), a> b > c.

Table 3. Effect of inoculation of *Azospirillum,* PSB and AM fungus on per cent root colonization and spore count of AM fungi in direct seeded rice

Treatment	Per cent root colonization			Spore count of AM fungi 50^{-1} g soil		
	45 DAS	90 DAS	At harvest	45 DAS	90 DAS	At harvest
T_1	12.40^{f}	11.60^{ef}	9.10^{h}	9.40^{h}	11.30^{g}	11.40^{f}
T_2	13.10^{ef}	12.80^{ef}	10.70^{g}	11.30^{f}	13.51^{f}	11.90^{ef}
T_3	12.60^{f}	12.30^{ef}	11.90^{f}	10.30^{g}	13.50^{f}	11.60^{f}
T_4	13.50^{ef}	12.90^{ef}	10.80^{g}	11.50^{f}	13.70^{ef}	12.10^{ef}
T_5	13.90^{de}	13.10^{ef}	13.20^{e}	12.50^{e}	14.90^{ef}	13.20d^{e}
T_6	36.40^{d}	49.70^{d}	30.30^{c}	16.20^{d}	55.59^{d}	38.30^{c}
T_7	15.00^{e}	13.60^{e}	13.20^{e}	12.59^{e}	16.30^{e}	14.40^{d}
T_8	41.30^{b}	53.40^{c}	31.50^{c}	22.80^{c}	63.30^{c}	39.20^{c}
T_9	44.90^{a}	55.60^{b}	27.40^{a}	30.00^{b}	74.50^{b}	43.30^{b}
T_{10}	45.30^{a}	59.40^{a}	35.00^{a}	34.00^{a}	82.90^{a}	44.67^{a}
SE.M±	0.41	0.53	0.30	0.27	0.69	0.40
CD (0.05)	1.22	1.59	0.90	0.82	2.05	1.19

Note: RDF for rice is 150:75:37.50 kg NPK ha^{-1}; Values are mean of three replications; Mean values followed by the same letter are not significantly different based on Duncan's multiple range test ($p<0.05$), a> b > c.

Soil biomass carbon

The soil biomass carbon increased with increase in the age of host upto 90 DAS and decreased at harvest. However, soil biomass carbon was more in 90 than 45 DAS (Table 4). The soil biomass carbon in the rhizosphere soil of DSR were significantly highest due to combined inoculation of *Azospirillum,* PSB and AM fungus (305.00, 333.16 and 256.93 mg kg^{-1} of soil at 45, 90 DAS and at harvest respectively) as compared to rest of the treatment combinations and individual inoculation with either of *Azospirillum,* PSB, AM or uninoculated control.

It's a well know fact that soil microbial activity and nutrient concentration are found in the rhizosphere area (Brown, 1975). Any change in this region might affect the rhizosphere microflora and in turn plant growth (Oswald and Ferchau, 1968). It is likely that amount of root and its nutrient status might have decreased with the advancement in the age of the host especially during harvest which, in turn caused a decrease in the rhizosphere microbial population (Gajda and Martyniuk, 2004). The results of present study are strongly supported by the fact drawn by Kundu and Gaur (1980). They observed a synergetic interaction between *Azotobacter* and phosphate solubilising bacteria when the two organisms were inoculated together in cotton. In the combined inoculation treatments, the population of both the organism was enhanced in addition to increase in yield of cotton.

Chemical properties of soil

The initial organic carbon, nitrogen, phosphorus and potassium before sowing of DSR in the experimental site were non-significant with each other in different treatment plots.

The available OC (Table 4), N, P_2O_5 and K_2O content of soil (Table 5) showed the declining trend throughout the experiment from 45 DAS to harvest. The availability of OC, nutrients in the rhizosphere soil of DSR were significantly higher due to combined inoculation of efficient strains of *Azospirillum,* PSB and AM fungus as compared to other treatments and uninoculated control. The available organic carbon in the soil in T_{10} recorded 0.463, 0.406 and 0.396 per cent at 45, 90 DAS and at harvest, respectively. The available nitrogen in the soil in T_{10} recorded 279.00, 274.50 and 268.90 kg ha^{-1} at 45, 90 DAS and at harvest, respectively. Similarly, available phosphorus in the rhizosphere soil was 38.33, 34.60 and 30.90 kg ha^{-1} at 45, 90 DAS and at harvest, respectively. Likewise, available potassium in the rhizosphere soil of DSR in T_{10} was found to be 367.50, 352.50 and 352.06 kg ha^{-1} at 45, 90 DAS and at harvest, respectively. The results were in agreement with Masciandaro *et al.* (2000) as they observed that, *Azospirillum* 2 kg ha^{-1} increased the available nitrogen through fixation. Singh *et al.* (1990) studied the interaction effects of *Glomus fasciculatum* and *Azospirillum brasilense* on yields of various genotypes of wheat. They observed the differences in VAM root colonization and increased grain yield with high P_2O_5 content in the dual inoculated wheat plants grown under pot culture and field conditions. Similar results were obtained by Veeraswamy *et al.* (1992) where they studied the effect of *Glomus intraradices* and *Azospirillum lipoferum* on growth of sorghum. Dual inoculation treatment resulted in significant increase in plant growth, root acid and alkaline phosphatases, uptake of P, N, Zn, Cu and Fe.

Table 4. Effect of inoculation of *Azospirillum*, PSB and AM fungus on soil biomass carbon and organic carbon in direct seeded rice

Treatment	Soil biomass carbon (mg kg^{-1})			Organic carbon (%)		
	45 DAS	90 DAS	At harvest	45 DAS	90 DAS	At harvest
T_1	158.03^{g}	301.06^{f}	180.81^{f}	0.316^{d}	0.313^{c}	0.303^{c}
T_2	192.85^{f}	303.20^{ef}	191.13^{f}	0.376^{bc}	0.376^{bcd}	0.363^{cd}
T_3	189.20^{f}	306.06^{def}	187.76^{f}	0.383^{bc}	0.366^{d}	0.356^{d}
T_4	265.07^{e}	308.10^{def}	221.30^{e}	0.383^{bc}	0.376^{bcd}	0.363^{d}
T_5	273.20^{e}	311.13^{cdef}	224.42^{e}	0.390^{bc}	0.380^{bcd}	0.366^{bcd}
T_6	280.63^{d}	315.06^{cde}	230.60^{d}	0.393^{b}	0.373^{cd}	0.366^{bcd}
T_7	285.90^{c}	319.00^{bcd}	235.53^{c}	0.373^{c}	0.373^{cd}	0.373^{bc}
T_8	290.39^{b}	324.13^{abc}	241.20^{c}	0.393^{b}	0.383^{bc}	0.380^{b}
T_9	301.85^{b}	329.13^{ab}	249.74^{b}	0.393^{b}	0.390^{b}	0.376^{bc}
T_{10}	305.00^{a}	333.16^{a}	256.93^{a}	0.463^{a}	0.406^{a}	0.396^{a}
SE.M±	3.24	4.69	3.08	0.006	0.006	0.005
CD (0.05)	9.64	13.94	9.17	0.017	0.014	0.014

Note: RDF for rice is 150:75:37.50 kg NPK ha^{-1}; Values are mean of three replications; Mean values followed by the same letter are not significantly different based on Duncan's multiple range test ($p<0.05$), a> b > c.

Table 5. Effect of inoculation of *Azospirillum*, PSB and AM fungus on available nitrogen, phosphorus and potassium content in the rhizosphere soil of direct seeded rice

Treatment	Available Nitrogen (kg ha^{-1})			Available P_2O_5 (kg ha^{-1})			Available K_2O (kg ha^{-1})		
	45 DAS	90 DAS	At harvest	45 DAS	90 DAS	At harvest	45 DAS	90 DAS	At harvest
T_1	220.33^{g}	215.80^{g}	210.30^{g}	28.00^{d}	26.70^{d}	23.50^{c}	331.20^{d}	311.30^{c}	299.50^{b}
T_2	256.30^{ef}	248.20^{f}	246.30^{ef}	30.90^{c}	27.70^{cd}	24.00^{bc}	360.60^{c}	349.40^{ab}	338.90^{a}
T_3	252.70^{f}	244.10^{f}	242.60^{f}	28.70^{d}	27.20^{cd}	24.00^{bc}	355.00^{cd}	345.70^{b}	335.20^{a}
T_4	260.70^{de}	251.80^{ef}	250.60^{ef}	31.40^{c}	27.60^{cd}	24.90^{bc}	360.90^{bc}	349.80^{ab}	339.20^{a}
T_5	260.70^{de}	256.20^{de}	250.60^{de}	32.30^{c}	28.60^{cd}	25.30^{bc}	364.30^{abc}	345.70^{b}	340.00^{a}
T_6	261.00^{d}	256.20^{de}	250.60^{de}	32.70^{c}	29.00^{c}	26.20^{b}	364.70^{ab}	345.90^{b}	341.10^{a}
T_7	262.30^{cd}	256.50^{d}	250.90^{d}	36.03^{ab}	32.30^{b}	28.60^{a}	365.50^{a}	350.50^{a}	341.30^{a}
T_8	265.70^{bc}	257.80^{cd}	252.30^{cd}	36.60^{ab}	32.90^{ab}	29.20^{a}	366.50^{a}	351.60^{a}	341.40^{a}
T_9	267.00^{b}	262.50^{b}	255.60^{bc}	37.60^{ab}	33.80^{a}	30.20^{a}	366.80^{a}	351.99^{a}	342.00^{a}
T_{10}	279.00^{a}	274.50^{a}	268.90^{a}	38.33a	34.60^{a}	30.90^{a}	367.50^{a}	352.50^{a}	352.06^{a}
SE.M±	1.50	1.50	1.50	0.70	0.70	0.80	1.40	1.50	1.30
CD (0.05)	4.41	4.41	4.40	2.17	2.19	2.36	4.01	4.50	17.18

Note: RDF for rice is 150:75:37.50 kg NPK/ ha^{-1}; Values are mean of three replications; Mean values followed by the same letter are not significantly different based on Duncan's multiple range test ($p<0.05$), a> b > c.

Thus, it can be concluded that the initial soil microbial population before sowing of DSR *viz*., bacteria, fungi, actinobacteria, *Azospirillum*, PSB and AM fungi were non-significant in soils collected from experimental plots. However, the populations of the said soil microflora increased in the rhizosphere with increase in the age upto 90 DAS. At 90 DAS, treatment T_{10} which is combined inoculation of efficient *Azospirillum*, PSB and AM fungus with reduced doses of N and P (*Azospirillum* + PSB + AMF + 75% N + 75% P + 100% K) recorded 6.04 x10^6, 7.89 x10^3, 7.11 x10^4, 39.30 x10^2, 8.01 x10^3 cfu g^{-1} of dry soil population of bacteria, fungi, actinobacteria, *Azospirillum* and PSB respectively, while AM fungi recorded per cent root colonization of 59.40 per cent and spore count of 82.90 50^{-1} g soil. Meanwhile, soil biomass carbon of 333.16 mg kg^{-1} of dry soil was recorded at 90 DAS in T_{10}. Though, the biological parameters showed an increasing trend upto 90 DAS but it showed a decline at harvest stage. The present study indicated that the growth and yield of DSR would be improved by the application of beneficial microorganisms along with the nutrient management by reducing at least 25 per cent of the recommended N and P fertilizer.

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Evaluation of dye decolourization ability of native lignin degrading fungi and bacteria from organic farm fields of Hyderabad-Karnataka region

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ABSTRACT

Dyedecolourization was used as substrate to evaluate ligninolytic activity in lignin degrading fungi and bacterial strains isolated from organic farming fields of Hyderabad-Karnataka region. Eleven fungal and eight bacterial strains were selected and tested for their capacity to degrade lignin by subjecting them to the Bavendamm test using malt extract tannic acid medium (META), low nitrogen agar with methylene blue [LNAM (MB)] and low nitrogen agar with phenol red media [LNAM (PR)]. Eleven fungal strains *viz.*, KGST-1, KGST-2, KGSA, KKST, KKSU-1, KKSP, BKST, RLST-1, RLSP-1, RMSP and KKSU-2 and eight bacterial strains *viz.*, KGSB-1, KGSP, KGSB-2, KKSB-2, BKSB, RMSP-1, RMSP-2 and RMSB were positive on all the three media. The ability of the strains to decolourize different indicators Remazol Brilliant Blue R (RBBR), Methyl Green (MG), Methylene Blue (MB) and Congo Red (CR) of the respective indicator on NA and PDA agar plates was tested. The fungal isolates KGST-1, KGST-2 and KKSP could decolourize RBBR and Congo Red even better than the reference strains. KGSA and RMSP could efficiently decolourize methylene green along with RBBR. Among the bacterial isolates KGSB-1 and RMSP-2 could decolourize RBBR even better than native isolates and reference strains.

Key words: Dye dicolouriztion, ligninolytic activity, remazol brilliant blue R, methyl green, methyl blue and congo red

There is evidence that the capacity of white rot fungi (WRF) in decolourizing dyes results in part from the activity of enzymes that take partin lignin depolymerization process. Low molar weight compounds, as metals and H_2O_2 can also be involved in pesticides degradation (Sun *et al*., 1993). Peroxidative activity does not seem to be the only extracellular enzyme available to WRF for dyes degradation and decolourization rate could not be related to one particular enzyme, but to the result of the ligninolytic mechanism action. To understand the basidiomycetes complex enzymatic system and the use of different dyes such as Remazol Brilliant Blue R (RBBR), Methyl Green (MG), Methyl Blue (MB) and Congo Red (CR) indicative for the degradative potential system. The white rot fungi (WRF) seem to be the unique microorganisms, which show capacities of degrading and mineralizing lignin and a series of organic pollutant compounds, highly toxic and recalcitrant. This capacity is, at least in some extent, caused by non-specific enzymatic system produced by these fungi during the lignin degradation and includes several enzymes of Lignin Peroxidase (LiP), Manganese Peroxidase (MnP) and Laccases. The multi-enzymatic system involved in the lignin degradation and mineralization is constituted of different ligninolytic enzymes combinations, being the occurrence of MnP and Laccase higher than LiP (Wesenberg *et al.*, 2003). *Phanerochaete chrysosporium* has been widely used as a model system to understand the process of lignin and some environmental pollutants biodegradation. The use of dyes offers a series of advantages in relation to conventional substrate because they are stable, soluble and cheap substrates with high rates of molar extinction and low toxicity.

MATERIALS AND METHODS

Identification of production of extracellular oxidases and lignolyase

The microbes were spotted on indicator media like Malt Extract Tannic Acid Agar Medium (META) and low nitrogen agar medium amended with phenol red (0.02%) and methylene blue (0.02%) separately (Karthikeyan and Siva Kumar, 2000). The colonies showing halo zones surrounding them on META medium and low nitrogen with methylene blue medium were considered positive for lignin degradation. The colonies showing red color surrounding them on low nitrogen agar medium amended with phenol red were considered as positive. The isolates positive on all these media were purified and maintained on nutrient agar and potato dextrose agar plates and refrigerated till further use.

Dye decolourization activity on NA and PD plates

In dye decolorization tests (Katia *et al.*, 2005), PDA medium containing 0.04% w/v Ramazol Brilliant Blue R, (RBBR) (R8001, Sigma), 0.04% w/v methyl green (M5015, Sigma) or 0.01% w/v guaiacol (G5502, Sigma) were used. Guaiacol was added into PDA before sterilization while RBBR and methyl green were added as a sterilized filtered solution. Each fungal strain was inoculated onto all media, incubated at 30^0C and the growth was followed for a period of two weeks. The cultivation was done in duplicate.

RESULTS AND DISCUSSIONS

Screening of microorganisms for lignin degradation on indicator media

Three indicator media were employed as suggested by Karthikeyan and Sivakumar (2000). They were: a) malt extract tannic acid medium (b) low nitrogen agar medium with methylene blue and (c) low nitrogen agar medium with phenol red. All the native isolates along with two reference bacteria and fungi were spotted on these media and incubated.

Malt extract tannic acid medium

On this medium, 8 bacterial and 11 fungal isolates showed positive reaction by producing halo zones around the colonies. The bacterial isolates were KGSB-1, KGSP, KGSB-2, KKSB-2, BKSB, RMSP-1, RMSP-2 and RMSB and fungal isolates were KGST-1, KGST-2, KGSA, KKST, KKSU-1, KKSP, BKST, RLST-1, RLSP-2, RMSP and KKSU-2 (Table 1). Two bacterial and two fungal reference strains tested were positive on the META medium *viz.*, *Pseudomonas putida, Bacillus subtilis, Phanerochaete chrysosporium* and *Pleurotus ostreatus.*

Low nitrogen agar medium with methylene blue

On this medium, 8 bacterial and 11 fungal isolates showed positive reaction by producing halo zones around the colonies. The bacterial isolates were KGSB-1, KGSP, KGSB-2, KKSB-2, BKSB, RMSP-1, RMSP-2 and RMSB and fungal isolates were KGST-1, KGST-2, KGSA, KKST, KKSU-1, KKSP, BKST, RLST-1, RLSP-2, RMSP and KKSU-2 (Table 1). Two bacterial and two fungal reference strains tested were positive on the LNAM + MB medium *viz.*, *Pseudomonas putida, Bacillus subtilis, Phanerochaete chrysosporium* and *Pleurotus ostreatus.*

Low nitrogen medium with phenol red

On this medium, 8 bacterial and 11 fungal isolates showed positive reaction by producing halo zones around the colonies. The bacterial isolates were KGSB-1, KGSP, KGSB-2, KKSB-2, BKSB, RMSP-1, RMSP-2 and RMSB and fungal isolates were KGST-1, KGST-2, KGSA, KKST, KKSU-1, KKSP, BKST, RLST-1, RLSP-2, RMSP and KKSU-2 (Table 1). Two bacterial and two fungal reference strains tested were positive on the LNAM + PR medium *viz.*, *Pseudomonas putida, Bacillus subtilis, Phanerochaete chrysosporium* and *Pleurotus ostreatus.*

Table1. Screening of microorganisms for lignin degradation using different indicator media

Isolate	META	LNAM (MB)	LNAM (PR)
*Phanerochaete chrysosporium** (FR1)	+	+	+
*Pleurotus ostreatus** (FR2)	+	+	+
Native Fungal Isolates			
KGST-1	+	+	+
KGST-2	+	+	+
KGSA	+	+	+
KKST	+	+	+
KKSU-1	+	+	+
KKSP	+	+	+
BKST	+	+	+
RLST-1	+	+	+
RLSP-2	+	+	+
RMSP	+	+	+
KKSU-2	+	+	+
*Pseudomonas putida** (BR1)	+	+	+
*Bacillus subtilis** (BR2)	+	+	+
Native Bacterial Isolates			
KGSB-1	+	+	+
KGSP	+	+	+
KGSB-2	+	+	+
KKSB-2	+	+	+
BKSB	+	+	+
RMSP-1	+	+	+
RMSP-2	+	+	+
RMSB	+	+	+

Note: * FR1 = Fungi Reference 1; * FR2 = Fungi Reference 2; *BR1 = Bacteria Reference 1; *BR2 = Bacteria Reference 2; META - Malt Extract Tannic Acid; LNAM + MB - Low Nitrogen Agar Medium with Methylene Blue; LNAM + PR - Low Nitrogen Agar Medium with Phenol Red; (+) = Present; (") = Absent.

Dye decolourization ability of the isolates on PDA and NA plates

The ability of the strains to decolourize different indicators Remazol Brilliant Blue R (RBBR), Methyl Green (MG), Methylene Blue (MB) and Congo Red (CR) of the respective indicator on NA and PDA agar plates. The results are shown in table 2. The results show that fungal isolates KGST-1, KGST-2 and KKSP could decolourize RBBR and congo red even better than the reference strains FR1 and FR2. KGSA and RMSP could efficiently decolourize methylene green along with RBBR. Among the bacterial

isolates KGSB-1 and RMSP-2 could decolourize RBBR even better than reference strains (Table 2).

Screening of microorganisms for lignin degradation using different indicator media

Eleven fungal and eight bacterial strains were selected and tested for their capacity to degrade lignin by subjecting them to the Bavendamm test using malt extract tannic acid medium (META), low nitrogen agar with methylene blue (LNAM (MB)) and low nitrogen agar with phenol red media [LNAM (PR)]. Eleven fungal strains *viz.*, KGST-1, KGST-2, KGSA, KKST, KKSU-1, KKSP, BKST, RLST-1, RLSP-1, RMSP and KKSU-2 and eight bacterial strains *viz.*, KGSB-1, KGSP, KGSB-2, KKSB-2, BKSB, RMSP-1, RMSP-2 and RMSB were positive on all the three media. All the reference strains showed positive on the three media that were tested. Positive reaction indicated can be correlated for production of Lignin Peroxidase (LiP), Manganese Peroxidase (MnP) and Laccase activity.

Table 2. Dye decolourization ability of the isolates on PDA and NA Plates

Isolate	PDA + RBBR	PDA + MG	PDA + CR	PDA + MB
*Phanerochaete chrysosporium** (FR1)	+++	–	+	–
*Pleurotus ostreatus** (FR2)	++	+	–	–
Native Fungal Isolates				–
KGST-1	+++	–	++	–
KGST-2	+++	–	+++	–
KGSA	++	++	–	–
KKST	+	+	–	–
KKSU-1	+	+	–	–
KKSP	+	–	++	–
BKST	+	–	+	–
RLST-1	–	–	–	–
RLSP-2	–	–	–	–
RMSP	++	++	–	–
KKSU-2	–	+	–	–
Isolates	NA + RBBR	NA + MG	NA + CR	NA + MB
*Pseudomonas putida** (BR1)	+	+	–	+
*Bacillus subtilis** (BR2)	+	+	+	–
Native Bacterial Isolates				
KGSB-1	++	–	–	–
KGSP	+	–	–	–
KGSB-2	+	–	–	–
KKSB-2	–	–	–	–
BKSB	+	–	–	–
RMSP-1	+	–	–	–
RMSP-2	++	–	–	–
RMSB	+	–	–	–

Note: * FR1 = Fungi Reference 1; * FR2 = Fungi Reference 2; *BR1 = Bacteria Reference 1; *BR2 = Bacteria Reference 2; + = Positive for decolorization of dye; " = Negative for decolorization of dye

Similar studies were conducted by (Chamuris *et al.*, 2000). They used indicator media as an approach for rapid screening of fungal isolates for lignin degrading ability and Turpeinen *et al*. (2003) isolated fungi from decomposting organic materials and tested their ability to secrete radical generating oxido-reductases using a special ABTS-agar medium. Two isolates of *Paecilomyces inflatus* formed characteristic dark green rings around the fungal mycelium. This indicated the production of extracellular oxidoreductases *viz.*, laccases or peroxidases. Surprisingly, none of the bacterial and actinomycete strains showed positive reaction on any indicator media tested.

Dye decolourization ability of the isolates on PDA and NA plates

The ability of the strains to decolourize different indicators Remazol Brilliant Blue R (RBBR), Methyl Green (MG), Methylene Blue (MB) and Congo Red (CR) of the respective indicator on NA and PDA agar plates was tested. The fungal isolates KGST-1, KGST-2 and KKSP could decolourize RBBR and Congo red even better than the reference strains. KGSA and RMSP could efficiently decolourize methylene green along with RBBR. Among the bacterial isolates KGSB-1 and RMSP-2 could decolourize RBBR even better than native isolates and reference strains. The ability of more numbers of dye decolourization indicates the production of more numbers of extracellular Lignolytic enzymes.

Similar studies were conducted by (Katia *et al.*, 2005). They studied on enzymatic system involved in RBBR decolorization was produced by basidiomycete fungi in solid medium. This system was produced even in the absence of the RBBR and the decolorization was performed in the absence of added H_2O_2 showing that there are different combinations of extracellular lignolytic enzymes produced in significant quantities under simple conditions of cultivation. Which enzymes are being stimulated or activated and their relation to organochlorine compounds degradation is not well known. RBBR decolorizing activity is a simple indicative method for a multi-enzymatic system and can be a valuable approach to be used as a tool for xenobiotic biodegradation studies as well as an indication of the physiological conditions of basidiomycetes during bioremediation process.

This method is popular because the use of colour indicators is generally simpler because there is no requirement of complex and costly equipments and chemicals

for sample handling and measurement (Kalmiº *et al*., 2008). Similar kind of study was conducted by using several different compounds by using them as indicators for ligninolytic enzymes production. RBBR and guaiacol are used frequently and their results correlate well to each other (Krogh *et al*., 2004 and Ang *et al*., 2011). Some fungal strains tested gave positive test results in RBBR containing medium but failed to decolorize guaiacol might be due to several reasons. One of them could be because the fungi require a longer adaptation time to produce lignolytic enzymes for the decolorization of guaiacol dye, as it has been reported for other colour indicators, such as RBBR (Ang *et al*., 2011). Methyl green was also used for lignolytic enzymes screening. The lack of detection of growth for *T. hirsute* and *P. pinophilum* on methyl green containing PDA media could be attributed to the high concentration of the indicator, which might have an inhibitory effect (Eichlerova *et al*., 2006).

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Response of okra (*Abelmoschus esculentus* L.) cv. Parbhani Kranti to pre-sowing seed treatment with gibberellic acid

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ABSTRACT

The field experiment was carried out at Agricultural Research Station, Awashi Tal-Khed Dist-Ratnagiri during summer 20013-14 to study the effect of pre-sowing seed treatment of GA_3 on growth and yield of okra. The treatments involved five concentrations of GA_3 (0, 25, 50, 75 and 100 ppm). The results indicated that the maximum plant height, number of branches per plant, number of fruits per plant, yield per plant (g) and yield per (q ha^{-1}) were recorded in the treatment T_4 (75 ppm GA_3). The growth parameters and yield increased with increasing GA_3 concentration up to 75 ppm and reduced in the treatment T_5 (100 ppm GA_3), which was at par with treatment T_2 (25 ppm GA_3).

Key words: Gibberellic acid, seed treatment, okra, plant height and yield

Okra (*Abelmoschus esculantus* L.) is an important vegetable crop of India, as it has great medicinal and nutritional value. It has export potential and it is commercially grown throughout the country. It can adopt wide rage of soil and climatic conditions and grow better with optimum management practices. As it has commercial importance, it is necessary to study the influence of different growth regulators and nutrients on its performance. The GA_3 promotes the germination and influence growth of the crop. The present investigation was carried out to study the effect of pre-sowing seed treatment of GA_3 on growth and yield of okra cv. Parbhani Kranti.

MATERIALS AND METHODS

The field experiment was conducted at Agricultural Research Station, Awashi Tal-Khed Dist-Ratnagiri during summer 2013-2014. The experiment was laid out in the randomized block design, which was replicated four times. The experimental area is in South Konkan coastal agro-climatic conditions having red lateritic soils. The treatments consisted of five levels of GA_3 *viz.* 0 (Distilled water), 25 ppm, 50 ppm, 75 ppm and 100 ppm. The experimental land was prepared by ploughing and harrowing. The plots having net size of 3.0 X 2.4 m were prepared. The basal dose of FYM at the rate of 20 t ha^{-1} was incorporated in the soil at the time of field preparation. The recommended dose of fertilizer (100 kg N, 50 kg P_2O_5 and 50 kg K_2O) was applied as basal dose where the 1/3 N was applied as basal dose and remaining dose of N was applied in two split doses at three and six weeks after sowing. Before sowing, the okra seeds were soaked in the respective concentrations of GA_3 for 24 hours and then air dried. The seed of okra *cv.* Parbhani Kranti was sown on 20th February, 2013 at 45 X 30 cm spacing. The seed germination percentage was calculated by counting the germinated seeds in respective treatment plots. The plant height, number of branches, number of fruits per plant, fruit yield per plant in gram and yield per hectare in quintals were recorded from ten randomly selected plants from each plot. The data were statistically analyzed by the method suggested by Panse and Sukhatme (1985).

RESULTS AND DISCUSSION

The data presented in table 1 clearly revealed that pre-sowing seed treatments with different concentration of GA_3 had significant effect on vegetative growth and yield of okra.

Germination per cent

The highest germination percentage (89.63 per cent) was observed in the treatment T_4, where the seeds were treated with 75 ppm GA_3. The germination percentage was increased with increasing concentration of GA_3 up to 75 ppm and it was reduced in 100 ppm GA_3 treatment.

Plant height (m) and number of branches per plant

There was significant increase in height of plant and number of branches per plant with increased level of GA_3 up to 75 ppm and it was reduced in 100 ppm GA_3. The maximum plant height and number of branches per plant were recorded in treatment T_4 (169.88 cm and 2.37 respectively).

Yield characters

The same trend was observed among the yield attributing parameters. The number of fruits per plants, fruit yield per plant (g) and yield quintal per hectare were

Table 1. Effect of pre-sowing seed treatment of GA_3 on growth and yield of okra *cv*. Parbhani Kranti

Treatment	Germination (%)	Plant height (m)	No. of branches plant^{-1}	No. of fruits plant^{-1}	Fruit yield plant^{-1} (g)	Yield. (Q ha^{-1})
T_1 - Control (Distilled water)	77.80	149.20	1.82	11.33	183.43	12.53
T_2 - 25 ppm GA_3	85.18	153.73	1.96	12.22	201.80	13.25
T_3 - 50 ppm GA_3	88.15	161.20	2.32	13.23	208.10	14.03
T_4 - 75 ppm GA_3	89.63	169.88	2.37	13.38	211.18	14.13
T5 - 100 ppm GA_3	84.95	154.05	2.00	11.97	201.58	13.11
SE m±	0.69	0.66	0.02	0.04	0.96	0.03
C.D. at 5%	2.14	2.02	0.06	0.12	2.95	0.10

recorded maximum in the treatment of seed treatment T_4 (75 ppm GA_3). The treatment T_5 (100 ppm GA_3) was at par with the treatment T_2 (25 ppm GA_3). The increase in the germination percentage up to certain concentration might be due to stimulating activities of gibberellic acid on embryo of seed. Singh and Kumar (1998) also reported the promotion of seed germination by using GA_3. The increase in the vegetative growth might be due to cell elongation. The Gibberellic acid treated plants were more physiologically active, which may result in production of more flowers and fruits. Kumar *et al.* (1997) and Vijayraghavan (1999), Unamba *et al.* (2009) have found the similar trend in okra, which helps to confirm the present findings.

Thus, it can be concluded that the treatment T_4 (75 ppm GA_3) gave the best result for seed germination as well as plant height, number of branches plant^{-1} and yield.

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Evaluation of socio-economic status of farmers of an organic farm of Himachal Pradesh, India

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ABSTRACT

In a country like India, farming is a culture rather than a profession. The interest in farming depends upon the tradition and family background of a worker. Keeping this in view, the socio-economic status of worker engaged in organic farming was studied. Various practices under organic farming in the mountain regions like compost production, use of bio-pesticides, crop rotations, *etc.* not only increases soil health and quality but promotes the activity of beneficial macro-and-micro-flora in the soil. The organic practices including Homa bhasm, Agnihotra and Biodynamic farming besides promoting healthy agriculture is expected to have seeming effects on behavioral aspects of the farm workers. However, apparently systemic information on social attributes of organic farm is still limited. As such present study was undertaken to evaluate the socio-economic efficiency of an organic farm. The socio- economic aspects of an organic farm CSKHPKV, Palampur were studied. The data was analyzed by the use of indices, ratios, percentages *etc.* and the results were interpreted. The various aspects as family structure and size, age distribution, caste structure, literacy level, occupational pattern, economic status behavioral aspects and livelihood security were evaluated.

Key words: Literacy level, occupational pattern, economic status, behavioral aspects, livelihood security

Organic agriculture is one among the broad spectrum of production methods that are supportive of healthy environment on this earth. More specifically, organic agriculture is a holistic food production system, which promotes and enhances agro ecosystem health, including biodiversity, biological cycles and healthy production system. It is a sustainable production system in which various eco-friendly and healing activities of Homa-farming, Agnihotra, Biodynamic farming are involved. These activities are known to show positive effect on the environment and do have healing effects. This also attributes to have a positive effect on the workers working in such an environment. Thus, a socio-economic survey was undertaken at CSKHPKV, organic farm to study certain perspectives in case of workers working in the organic farm.

MATERIALS AND METHODS

Both primary and secondary data were used in this study. The primary data was generated through questionnaire from the records maintained on the organic farm, while secondary data was taken from published/ unpublished reports of different agencies. Both tabular and statistical tools were employed for ananlysis and interpretation of data. The primary data collected on organic farm was tabulated to work out averages, ratios, percentages and indices. Tabular techinque was employed to study the socio-economic status of field staff. The existing level of income, literacy rate, nutritional status of workers on the farm were analyzed.

To meet out the objectives of the present study, both tabular and statistical tools were employed for analysis and interpretation of data. The primary data collected on organic farm was tabulated to work out averages, ratios, percentages and indices. Tabular technique was employed to study the socio-economic status of field staff. The existing level of income, literacy rate and nutritional status of workers on the farm was also analyzed. The following types of indices and ratios were worked out for present study :

$$\text{Sex-ratio } (1000^{-1} \text{ males}) = \frac{\text{Total population of females}}{\text{Total population of males}} \times 1000$$

$$\text{Literacy rate (\%)} = \frac{\text{Total number of literate persons}}{\text{Total population}} \times 100$$

$$\text{Literacy index} = \frac{\text{Cumulative total literacy score of workers}}{\text{Total number of workers}}$$

Where, Literacy score (as per score used) : 0 for illiterate; 1 for primary; 2 for high school; 3 for matriculation; 4 for senior secondary; 5 for graduate and 6 for post graduate

$$\text{Income equivalent} = \frac{\text{Value of output of ith system}}{\text{Value of output of cereal based system}}$$

$$\text{Livelihood security} = \frac{\text{Actual working days - 183}}{\text{Maximum no. of working days - 183}}$$

RESULTS AND DISCUSSION

Socio-economic features of farm workers

Family structure and size

The family size of the farm workers is shown in table 1 and 2. The average size of family of the farm workers was found to be 4.86. The average size of family was more for unskilled workers, whereas it was low for skilled workers. It was observed that proportion of children was higher in male category. The sex ratio was found alarmingly low. Further, it was seen that large percentage of unskilled workers had joint family structure, whereas large percentage compared to unskilled workers had nuclear family structure in case of skilled workers. This was attributed to the tendency of the married couple to lead an independent life. However, this tendency may not be compatible with development of agriculture as it was resulting into fragmentation and subdivision of holdings into marginal farms. Sharma *et al.* (1999) also confirmed the regressive fragmentation and subdivision of holdings caused by inheritance laws and tenancy reforms leading to marginalization of holdings in Himachal Pradesh.

Table 1. Family size of farm workers on different categories (Per cent)

Particular family size (No.)	Skilled	Unskilled	Total
Male			
a) Adult	39.02	40.95	39.86
b) Children	19.51	14.28	15.75
Sub total	58.53	55.23	55.61
	(2.67)	(2.77)	(2.73)
Female			
a) Adult	34.14	34.28	33.78
b) Children	7.31	10.47	9.58
Subtotal	41.45	44.75	43.36
	(1.89)	(2.23)	(2.13)
Overall			
a) Adult	73.17	75.24	73.65
b) Children	26.83	24.76	25.00
Average family size (No.)	4.56	5.00	4.86
Sex-ratio (Females / 1000 males)	708	810	762

Figures in parentheses show number of persons in each category

Table 2. Family structure of farm workers on different categories (No. of persons)

Particular	Skilled	Unskilled	Total
Family structure			
Nuclear	4 (44.44)	3 (14.29)	7 (23.33)
Joint	5 (55.56)	18 (85.71)	23 (76.67)
Total	9 (100)	21 (100)	30 (100)
Average size			
Nuclear	3.8	5.0	4.3
Joint	5.2	4.9	5.0

Figures in parentheses show percentages to total in each column.

Age distribution

Age distribution of farm workers employed on the organic farm has been shown in table 3. About 30 per cent of the workers belonged to the young age category of 25-30 years and 23 per cent in the age group of 20-25 years. There were just 13 per cent of the workers in the higher age group above 45 years of age. The mean age was estimated 31.66 years for skilled and 30.33 for unskilled workers on the farm. This clearly shows that majority of the farm workers employed on the organic farm belonged to younger generation. Similar results were shown by Fernandez *et al.* (1998) who considered organic farming to be fundamentally different from conventional farming in that their control focuses on ecologically based pest and nutrient management practices. According to them, organic farmers also tend to have a different socio- economic profile. The empirical studies showed that organic vegetable growers tend to be younger and more educated than conventional farmers.

Caste structure

The caste-wise distribution of workers showed that maximum workers belonged to scheduled caste category (Table 4). Accordingly, large percentage in case of unskilled

Table 3. Age-wise distribution of farm workers on different categories (No. of persons)

Age class (years)	Skilled	Unskilled	Total
20-25	3 (33.34)	4 (19.05)	7 (23.34)
25-30	1 (11.11)	8 (38.10)	9 (30.00)
30-35	2 (22.22)	4 (19.05)	6 (20.00)
35-40	2 (22.22)	2 (9.52)	4 (13.33)
40-45	0	0	0
Above 45	1 (11.11)	3 (14.28)	4 (13.33)
Total	9 (100)	21 (100)	30 (100)
Mean age (years)	31.66	30.33	30.73

Figures in parentheses show percentages to total in each column

Table 4. Caste – wise distribution of farm workers (No. of persons)

Caste	Skilled	Unskilled	Total
General	6 (66.67)	3 (14.29)	9 (30.00)
Scheduled caste	1 (11.11)	11 (52.38)	12 (40.00)
Other backward classes	2 (22.22)	7 (33.33)	9 (30.00)
Total	9 (100)	21 (100)	30 (100)

Figures in parentheses show percentages to total in each column.

workers belonged to this category. The workers from general category were low in percentage in case of unskilled workers. However, majority of the skilled workers belonged to general category. This clearly showed the low socio-economic status of scheduled caste forming the low paid labour class in the society.

Literacy level

Literacy level plays catalytic role in enhancing human capital. The educated worker can easily comprehend the scientific recommended technologies besides improving his work efficiency. It can be visualized from table 5 that large proportions in case of skilled workers were graduates or post graduates. In case of unskilled workers large proportions of workers were matriculates. Literacy index was high in case of skilled workers and low for unskilled workers.

Occupational pattern

The occupational pattern of working persons is presented in table 6. It was seen that about large percentage of the total working persons were engaged in agriculture and animal husbandry, of which unskilled worker family members dominated. Thus, agriculture was found to be the main source of livelihood for majority of the unskilled workers. However, services formed the major avocation for families of skilled workers.

Economic status and contribution of organic farm in livelihood

The income status of the workers is presented in table 7. It was observed that service was the main source of income for the family income of skilled workers, whereas agriculture and dairy contributed mainly for the unskilled workers. Contribution of wage income from organic farm in case of skilled workers was comparatively low as the family members of skilled workers had other sources of income that predominated organic farm income. Whereas, in case of unskilled workers, organic farm income was the main source

Table 5. Literacy status of farm workers on organic farm (No. of persons)

Educational level	Skilled	Unskilled	Total
Illiterate	0	1 (4.76)	1 (3.33)
Primary	0	2 (9.52)	2 (6.67)
Middle	1 (11.11)	3 (14.29)	4 (13.33)
Matriculation	2 (22.22)	10 (47.62)	12 (40.00)
10+2	1 (11.11)	5 (23.81)	6 (20.00)
Graduate	3 (33.34)	0	3 (10.00)
Postgraduate	2 (22.22)	0	2 (6.67)
Total	9 (100)	21 (100)	30 (100)
Literacy Index	4.32	2.76	3.23

Figures in parentheses show percentages to total in each column.

Table 6. Occupational pattern of families of farm workers (No. of persons)

Occupation	Skilled	Unskilled	Total
Agri / Dairy	3 (15.79)	34 (64.16)	37 (51.39)
Business[1]	2 (10.52)	5 (9.43)	7 (9.72)
Service [2]	11 (57.90)	5 (9.43)	16 (22.22)
Others[3]	3 (15.79)	9 (16.98)	12 (16.67)
Total	19 (100)	53 (100)	72 (100)

[1]Business including cottage industries; [2]Service including DPL (daily paid labour) and pensioners; [3]Others including rural handicrafts and artisans; Figures in parentheses show percentages to total in each column.

Table 7. Economic status and contribution of organic farming towards livelihood of workers Rs / household

Occupation	Skilled	Unskilled	Total
Agri / Dairy	6,111(4.29)	20,666 (28.95)	16,300 (17.59)
Service	78333 (55.06)	10,571 (14.81)	30,900 (33.35)
Others	7222 (5.08)	9380 (13.14)	8,733 (9.42)
Earning from organic farm	50,600 (35.57)	30,780 (43.10)	36,726 (39.64)
Total family income	1, 42,266 (100)	71,397 (100)	92,659 (100)
Contribution of wage income from organic farm in total family income (%)	35.57	43.11	39.63

Figures in parentheses show percentages to total in each column

Table 8. Impact of organic farming on behavioral aspects of farm workers (No.of workers)

Particular	Skilled (9)	Unskilled (21)	Total (30)
Improved lifestyle	7 (77.78)	12 (57.14)	19 (63.33)
Farming knowledge	8 (88.89)	8 (38.10)	16 (53.33)
Better wage rate	9 (100)	9 (42.86)	18 (60.00)
Improved ethical and spiritual thinking	8 (88.89)	11 (52.38)	19 (63.33)
Work punctuality	8 (88.89)	12 (57.14)	20 (66.66)
Improved social cohesion and interaction	6 (66.67)	10 (47.62)	16 (53.33)

Figures in parentheses show percentages to total in each column.

Table 9. Extent of livelihood security of skilled farm workers (No. of days)

Actual working days	No. of skilled workers	Livelihood security
Working days less than 183	0	-
183 - 200	0	-
200 - 225	0	-
225-250	0	-
250 - 275	0	-
More than 275	9	7.99
Average 313.89		0.89

as many of the members of unskilled workers did not had service as occupation.

Impact of organic farming on behavioral aspects of farm workers

Organic farming not only paves the way for healthy agricultural practices but also affects the overall personality of the individual through participation in various activities of homa-farming, use of bio-pesticides, chanting of mantras, *etc*. Table 8 depicts the perceived changes in the behaviour of workers after joining organic farming. It was observed that in case of skilled workers majority of individuals were positively affected through increased farming knowledge, better wage rate, improved ethical and spiritual thinking. In case of unskilled workers no obvious positive reaction was observed for farming knowledge and wage rate though there was perceptible change in punctuality and spiritual thinking.

Livelihood security of farm worker

The livelihood security of farm workers was evaluated and it was found that the average livelihood security of skilled was high, whereas for the unskilled workers it was low. Many of the unskilled workers had no livelihood security because of lesser working days and uncertainty of getting work in the farm (table 9, 10).

Table 10. Extent of livelihood security of unskilled farm workers (No. of days)

Actual working days	No. of skilled workers	Livelihood security
Working days less than 183	7	-
183 - 200	2	0.204
200 - 225	1	0.25
225 - 250	4	1.576
250 - 275	6	3.454
More than 275	1	0.65
Average 313.89		0.29

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Effect of nutrient management and cropping system on productivity under different rice based cropping systems

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ABSTRACT

A field experiment was conducted during 2010-11 to 2012-13 at Jabalpur (Madhya Pradesh) to study the effect of nutrient management and cropping system on productivity under different rice based cropping systems. The 4 different cropping systems (CS1- Green manuring Sunhemp – Rice – Wheat; CS2 – Rice – Chickpea –Sesame; CS3 – Rice – Berseem; CS4 – Rice - Vegetable pea - Sorghum) and three nutrient managements M1- 100% Organic (1/3 N through each of FYM, Vermicompost and Neem oil cake), M2 - 100% Inorganic (100% NPK through fertilizers), M3 - INM (50% NPK through fertilizer + 50% N through organic sources) with 3 replications were conducted in Strip Plot Design. The soil of the experimental field was sandy clay loam in texture, neutral in reaction (7.3), normal EC (0.52), low in OC (0.72%), medium in available N (264.05 kg ha^{-1}) and P (12.8 kg ha^{-1}) and high in K (285.2 kg ha^{-1}). The highest weed intensity of (352.6 m^{-2}), weed biomass (1183 kg ha^{-1}), rice equivalent yield (69.35 q ha-1) and production efficiency (22.07 kg ha^{-1} day^{-1}) during *kharif*, *rabi* and summer season in 100 per cent inorganic nutrient management and Rice-Berseem cropping system.

Key words : Cropping system, integrated nutrient management, rice and wheat

Madhya Pradesh is relatively underdeveloped with regards to agricultural productivity rural employment and economic status as compared to most of the Indian states. Rice and wheat are grown in sequence in an area about 2.7 million hectares in Punjab and contribute 80 per cent in the total food pool of the state of Punjab (DAGP, 2011). It is observed that maximum infestation of weeds is observed in organic nutrient management plots in rice based cropping system (Vishwakarma *et al*., 2012). Thus, the severe incidence of weeds results in yield reduction as high as 40-60 per cent depending upon the intensity and the type of weeds (Singh *et al*., 2003). Thus, if changes in nutrient management and cropping systems are made, it affects the density of weeds. As weeds cause major loss to yield of rice, an experiment was conducted to evaluate weed intensity, weed biomass and rice equivalent yield as affected by organic, inorganic and integrated nutrient management in different rice based cropping systems including oilseeds, pulses, vegetable and fodder crops.

MATERIALS AND METHODS

The present study was conducted during 2011-12 to 2012-13 at the Research Farm of Jawaharlal Nehru Krishi VishwaVidhyalaya, Jabalpur (Madhya Pradesh), India on a sandy clay loam soil. The soil of the experimental site had a pH 7.4, EC 0.51dS/m and organic carbon 0.7 per cent. The available soil nitrogen, phosphorus and potash were 264, 12.6 and 282 kg ha^{-1}, respectively. The bulk density of the soil was 1.35 mg/m^3. The factors studied included 3 nutrient management practices *viz.*, organic manure (ONM), chemical fertilizers and integrated nutrient (50:50) (INM) and 4 cropping systems *viz.*, rice-durum wheat - green manuring, rice chickpea-sesame, rice berseem (fodder + seed), rice - vegetable pea – sorghum (fodder) in Strip Plot Design with 3 replication. The crop varieties grown were Pusa Sugandha Basmati-5 in rice, MPO-1106 in durum wheat, JG-24 for gram, JB-1 for berseem, Arkel for vegetable pea during winter season and TKG-55 in sesame and MP Chari in sorghum during summer season. These crops were raised with recommended agronomic practices. In organic manure treatment, nutrients were applied through farmyard manure. The manure was applied on the nitrogen equivalent basis for each crop. The nutrient composition of FYM was 0.5, 0.25, 0.5 per cent N, P_2O_5 and K_2O respectively. For the weed management, mechanical measures were adopted and for insect pest management, neem oil (Azadiractin 0.03%) was applied as and when required under organic nutrient management. In chemical fertilizer treatment, nutrient was applied through chemical fertilizers *viz.*, urea, single super phosphate, muriate of potash, while plant protection was done through recommended pesticides, when required. The recommended dose of fertilizers for rice, wheat, chick pea, sesame, vegetable pea, sorghum and berseem were 120 : 26.4 : 33.3, 120 : 26.4 : 33.3, 20 : 60 : 30, 30 : 60 : 30, 20 : 26.4 : 16.6, 100 : 22 : 25 and 20 : 26.4 : 16.6 kg N : P : K ha^{-1}.

RESULTS AND DISCUSSION

The presence of predominant weed flora was almost similar in all plots, when rice was grown under different rice based cropping system but under 100 per cent organic nutrient management recorded the maximum infestation of weeds. The predominant weed in transplanted rice was *Echinoclova crusgalli*, which contributed 39 per cent of the total weed intensity at most critical period 30 DAT. The next predominant weed was *Cyperusiria* with relative density of 33 per cent at 30 DAT. The infestation of *Echinochlova colona* and *Cyperusiria* declined at maturity, while density of other weeds increased. During *rabi* season *Medicago denticulate* was found to be more dominant in almost all crops. Its relative density was 42.3, 38.9, 22.8 and 46.9 per cent in wheat, chickpea, berseem and vegetable pea. During summer season *Portulacao leracea* was most dominating weed in all crops with relative density of 42.7 and 43.6 per cent in sesame and sorghum.

Effect on weed intensity

During the experiment 100 per cent organic nutrient management recorded significantly higher total weed intensity of 352.6 plants m^{-2} which was significantly higher than INM (316.6 plants m^{-2}) and 100 per cent inorganic nutrient management (273.2 plants m^{-2}). The rice-vegetable pea sorghum cropping system recorded the higher weed intensity of (327.5 m^{-2}) and lower in rice-berseem cropping system which recorded the weed intensity of (231.9 plants m^{-2}).

Effect on weed biomass

Similarly, higher total weed biomass was recorded in100 per cent organic nutrient management (1183 kg ha^{-1}) which was significantly higher than integrated nutrient management (1150 kg ha^{-1}) and lowest in 100 per cent inorganic nutrient management (1085 kg ha^{-1}). The rice-berseem cropping system recorded the lower weed biomass of (906 kg ha^{-1}) as compared to rice-chickpea-sesame and rice-vegetable-sorghum. Green manure-rice-wheat had weed mass (803 kg ha^{-1}). As berseem is fodder crop and its growth habit is such that it completely cover the ground and kasni is the only dominant weed which infest berseem crop.

Effect on rice equivalent yield

The weed intensity and weed biomass influenced the rice equivalent yield (q ha^{-1}) and production efficiency (kg ha^{-1} day^{-1}) in different rice based cropping system. 100 per cent inorganic nutrient management recorded (69.35 q ha^{-1}) rice equivalent yield which was at par with INM (66.07 q ha^{-1}) and 100 per cent organic nutrient management (60.00 q ha^{-1}). The rice berseem cropping system recorded the higher rice equivalent yield of (74.74 q ha^{-1}) followed by rice-vegetable pea-sorghum (70.19 q ha^{-1}), green manuring-rice-wheat (63.90 q ha^{-1}) and rice chickpea-sesame (51.74 q ha^{-1}).

Effect on production efficiency

The production efficiency of 100 per cent inorganic nutrient management was the maximum (22.07 kg ha^{-1} day^{-1}) which was at par with INM (21.07 kg ha^{-1} day^{-1}) and

Table 1. Effect of different nutrient management and cropping system on weed intensity m^{-2}, weed biomass (kg ha^{-1}), rice equivalent yield (q ha^{-1}) and production efficiency (kg ha^{-1} day^{-1}) (mean of 2011-12 and 2012-13)

Treatment	Weed intensity/m^{-2}				Weed biomass (kg ha^{-1})				Rice equivalent yield (q ha^{-1})	Production efficiency (kg ha^{-1} day^{-1})
	Kharif	*Rabi*	Summer	Total	*Kharif*	*Rabi*	Summer	Total		
100% organic (1/3 N through each of FYM, Vermicompost and Neem oil cake)	120.2	134.2	98.2	352.6	470	390	323	1183	60.00	19.08
100% Inorganic (100% NPK through fertilizers)	102.3	103.4	67.5	273.2	443	340	302	1085	69.35	22.07
Integrated Nutrient Management (50% NPK through fertilizer + 50% N through organic sources)	118.2	120.3	78.1	316.6	459	373	318	1150	66.07	21.03
SEm±	0.12	0.14	0.12	–	0.10	0.12	0.15		1.08	0.35
CD (P=0.05)	0.30	0.40	0.30	–	0.30	0.30	0.42		2.70	0.87
Green manuring (sunhemp) - rice (Pusa Sugandha 5) - wheat (MPO 1106)	104.5	134.0	–	238.5	462	341	–	803	63.90	20.16
Rice (Pusa Sugandha 5) - chickpea (JG 322) - sesame (TKG 55)	99.2	120.5	97.9	317.6	440	398	321	1159	51.74	16.02
Rice (Pusa Sugandha 5) - berseem (JB 5) (fodder + seed)	129.2	102.7	–-	231.9	568	338	–-	906	74.74	26.04
Rice (Pusa Sugandha 5) - vegetable pea (Arkel) -sorghum (MP Chari) (fodder)	118.3	106.9	102.3	327.5	523	352	331	1206	70.19	20.70
SEm±	0.13	0.11	0.15	–	1.5	1.4	1.6		6.48	2.15
CD (P=0.05)	0.39	0.27	0.42	–	4.6	4.1	4.6		16.20	5.37

100 per cent organic nutrient management (19.08 kg ha^{-1} day^{-1}). The rice-berseem cropping system recorded the higher production efficiency of (26.04 kg ha^{-1} day^{-1}) followed by rice-vegetable pea-sorghum (20.70 kg ha^{-1} day^{-1}), green manuring-rice-wheat (20.16 kg ha^{-1} day^{-1}) and rice-chickpea-sesame (16.02 kg ha^{-1} day^{-1}) which was in conformity as also reported by Upadhyay *et al*., 2011.

Therefore, it was conclude that the 100 per cent inorganic nutrient management in rice-berseem cropping system recorded the maximum rice equivalent yield and production efficiency and lower weed intensity and weed biomass during both the years.

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Effect of planting dates and varieties on growth and yield of potato (*Solanum tuberosum* L.) in Hamelmalo area

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ABSTRACT

Potato is one of the most important vegetable crop of Eritrea grown throughout the country. The appropriate planting time and the best promising potato varieties for growing under Hamelmalo environmental condition are not identified. Considering this point, a study has been carried out to find the suitable date of planting as well as varieties to grow in Hamelmalo area. The experiment consisted of four planting dates (Sept-20, Oct-2, Oct-14 and Oct-26) and three potato varieties Ajiba, Cosmos and Zafira were tested in Randomized Complete Block Design with three replications under irrigated condition at the experimental farm of Hamelmalo Agricultural College during the growing season of 2013-14 with objective to evaluate and find suitable potato variety and planting time. Observations on growth parameters were recorded at 15 days interval during growth period of the crop. The data on yield and yield attributing parameters were recorded at harvesting. Data recorded on various parameters were analyzed statistically and analysis of variance showed that stem diameter and the yield of potato varieties significantly differed with different planting dates. Whereas, the tuber numbers and average tuber weight were found to be significantly affected by planting dates and varieties. On the other hand, stem number of potato varieties significantly differed within the varieties. It has been observed that delay in planting decreased the potato tuber yield. The potato variety Cosmos was the most adaptive variety with average yield of 57.5 t ha^{-1} in Hamemalo area. Based on the findings of this research work it can be recommended that varieties Ajiba and Zafira varieties are preferable for planting early in the first week of October and Cosmos variety is preferable planting in the bit late in third week of October for cultivation in Hamelmalo region.

Key words: Potato, variety, planting date and tuber

Potato (*Solanum tuberosum* L.) is one of the most important vegetable crops grown worldwide for its edible underground tubers used as vegetable. As a food crop, potato is the third most important food crop in the world after rice and wheat and more than a billion people worldwide consume potato (Molina Av La and La Molina, 1895; Bagheripour *et al*., 2012; Dehdar *et al*., 2012). Potato is considered as a very important crop in feeding the developing countries of the world due to its high nutritional value (Felenji and Ahmadizadeh, 2011). It is a unique crop, which can supplement the food needs of the country in a sustainable manner. Growth and quality of potato is influenced by environmental factors. Other factors that influence growth of the crop but that can be controlled by the grower are variety, planting time and pest management (Khan *et al*., 2011). The potato yield varies with variety, planting time, quality of seed, soil type, crop duration, growing region and cultural operations followed for growing crop (Rana, 2008; Malic, 2010). Delay in cultivation of potato causes reduction in yield. Yield reduction is dependent on the reaction of maternal tubers to soil temperature at the time of planting (Tomar, 1995). Altering the planting time can influence the potato yield. Research on various dates of planting indicate that yield of tubers is reduced to 0.75 t ha^{-1} for $week^{-1}$ when the planting date is delayed (Radley, 1993).

Potato is an important cash crop in Eritrea, especially in the high and mid lands. It is mainly grown in the highland of Eritrea, particularly in Zoba Makel and Debub but rarely in Zoba Anseba, Gash Barka, Southern and Northern Red Sea (Anonymous, 2012) but it is not grown in Hamelmalo area, which is somewhat semi-arid zone. Thus, selection of potato variety and determining an appropriate planting time is an important step forward in potato production in this area

MATERIALS AND METHODS

The experiment was conducted at the experimental farm of Hamelmalo Agricultural College (HAC) during the year 2013-14 in the winter season. Hamelmalo is located about 12 km North of Keren in Sub Zoba Hamelmalo at altitude of 1278 meter above sea level and at $15^0 52' 16''$, North latitude and $38^0 27' 44''$ East longitude. It receives annual mean rainfall of 468 mm. The experimental area has mean annual minimum and maximum temperature of 15 and 22°C, with sandy loam soil and is one of the major fruit producing area.

Experimental design and treatments

The experiment consisting three potato varieties (Ajiba, Cosmos and Zafira) and four dates of sowing (Sept. - 20, Oct. - 02, Oct. - 14 and Oct. - 26) was carried out in Randomized Complete Block Design with three replications to determine the best varieties and suitable planting time in this area to expand the production area of potato outside the central high lands of Eritrea, since it is one of the most important crop to Eritrea**.**

Experimental procedures

The experimental field was ploughed, harrowed and leveled before three weeks of planting. Well decomposed Farm Yard Manure (FYM) at the rate of 50 t ha^{-1} was applied to enrich the soil. The experimental field was divided into treatment plot having size 2.8 x 2.4 m. A distance of 1 m between the plots and 1.5 m was maintained between blocks. Row spacing of 0.7 m was uniformly used. Each treatment plot was consisted of four rows, with 32 plants spaced at 0.3 m within the row. The treatments were assigned randomly. Well sprouted uniform seed tubers of potato varieties were planted at 10 cm depth at the base of the ridge in the furrow. After sprouts emergence, the treatment plots were irrigated uniformly whenever needed through furrow irrigation system at an interval of 5 days. Hoeing and weeding were done manually and uniformly to all experimental plots as and when necessary. Nitrogen (5.24 g $plot^{-1}$) and Phosphorus (3.5 g $plot^{-1}$) were applied at planting 5 cm away from the seed tuber on either side through urea (46% nitrogen) and DAP (46% P) respectively. Observations on growth yield and yield attributing parameters were recorded and compiled.

Data analysis: The data recorded on each parameter were subjected to statistical analysis through GENSTAT Software at 5% level of significance (95% confidence limit) for analysis of variance.

RESULTS AND DISCUSSIONS

Number of stems ($plant^{-1}$) at 15 days after planting

A significant difference was observed in number of stems $plant^{-1}$ among all four planting dates and varieties at 15 days afer planting. The interaction of varieties with planting date was found significant. The highest number of stems $plant^{-1}$ (4.4) was recorded by Ajiba planted on Oct.-26 followed by Cosmos (4) planted on Oct.-2. Whereas, minimum stem numbers $plant^{-1}$ (1.4) was found iin the case of Zafira planted on Oct.-14 (Fig.1). Some of the stems of the Zafira variety were damaged by collar rot when the crop was planted on Oct.-14. Because of this, the stem number of the varieties decreased markedly during this planting date. The seed tubers might be infected from the store by itself not from difference of planting dates because symptoms of the disease were observed in some tubers in the store.

Number of stems ($plant^{-1}$) at 30 days after planting

The results showed that the effect of planting dates on number of stems $plant^{-1}$ after 30 days of planting had highly significant difference statistically. The difference of stem number $plant^{-1}$ among the varieties was also found significant. The interaction between the planting dates and variety had also significant effects at 30 days after planting. The maximum numbers of stems $plant^{-1}$ (5.13) was recorded in case of Cosmos when the crop was planted on Oct.-14 followed by Ajiba (5.07) planted on Oct.-26 (Fig.1). On the other hand minimum number of stems $plant^{-1}$ (2) was recorded in case of Zafira planted on Oct.-14. The means values showed that planting from Sept.-20 to Oct.-26 increased the number of main stems $plant^{-1}$. With Cosmos and Ajiba the stem number appeared to decrease slightly this might be due to seed tuber size or number of eyes on seed tuber seed differences. This result is in agreement with the finding of Bagheripour *et al*. (2012). Regardless of the seed piece size physiologically old seeds generally produce more stems $seed^{-1}$ piece than non-aged seed pieces of the same size.

Number of stems ($plant^{-1}$) at 45 days after planting

The stems $plant^{-1}$ at 45 days of planting showed no significant response to treatments ($p<5\%$). However, the highest number of stems $plant^{-1}$ (5.07) was recorded from both varieties, Ajiba planted on Oct.-14 and Oct.-26. Whereas, in Cosmos (5.07) planted on Oct.-14^{th} (Fig. 1). While the minimum number of stems $plant^{-1}$ (2.13) was recorded with Zafira planted on Oct.-14, which was not significant. The stem number was found positively correlated with stem diameter, leaf number and tuber yield.

Number of stems ($plant^{-1}$) at 60 days after planting

Stem number was significantly influenced by varieties. The difference among all four planting dates and their interaction with the varieties was not found significant statistically. However, maximum stem numbers $plant^{-1}$ (5.33) was recorded in case of Cosmos, when the crops were planted on Oct.-14 followed by Ajiba (5.13) again when planted on Oct.-14 (Fig. 1). While, minimum number of stems $plant^{-1}$ (2.2) was found with Zafira, when the crop was planted on Oct.-14. Cosmos variety producing the higher number of stems may be its genetic potential or a better adaption with Hamelmalo environment conditions. The interaction between the planting dates and varieties was found non significant at 60 days after planting. It was

observed that delay in planting resulted in increased number of stem plant except in case of Zafira was low when the crops was planted on Oct.-14 then increased at fourth planting dates. These results are supported by the findings of Farooq *et al*. (1989) and Thornton *et al*. (2007). The reason for the increase number of stems with delay in planting date could be aging of seed tubers used for planting, which were stored in the college from Sept.-1 to Oct.-26, which got sufficient time and sprouted before they get planted. Early planted seed tubers produced lesser sprouts before planting, while tubers planted later had already sprouted and produced maximum number of sprouts before planting, which finally resulted in higher number of stems plant^{-1} at late planting. These results are supported by the findings of Farooq *et al*. (1989) and Darini *et al*. (2013).

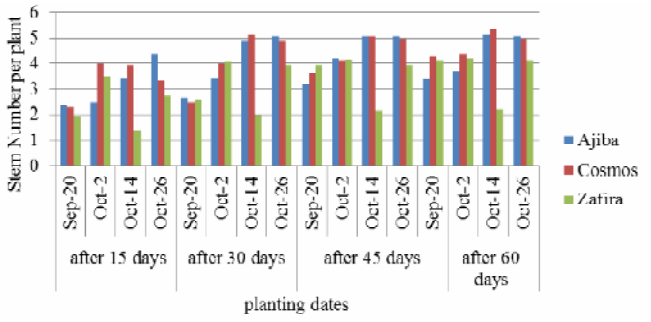

Fig. 1. Effect of planting dates and varieties on plant stem number

Stem diameter at 15 days after planting

Highly significant difference was observed in stem diameter among all four planting dates at 15 days after planting and the difference among the varieties was also found highly significant statistically. Interaction between the planting dates and varieties also gave significant results at 15days after planting. The maximum stem diameter (0.973) was observed in Ajiba followed by Cosmos (0.863cm), when both crops were planted on Oct.-2, whereas the minimum stem diameter (0.45cm) was found with Zafira when planted on Oct.-26th at 15 days after planting (Fig. 2).

Stem diameter at 30 days after planting

The stem diameter was significantly affected by both varieties and planting date but their interactions have not shown signicant affects. The maximum stem diameter (1.207 cm) was observed in Ajiba followed by Cosmos (1.113 cm), when both were planted on Oct.-2 at 30 days after planting whereas, the minimum stem diameter (0.873 cm) was found when planted on Sept.-20 by Zafira at 30 days after planting (Fig. 2).

Stem diameter at 45 days after planting

Highly significant difference was observed in stem diameter among all four planting dates. The difference of the stem diameter among varieties and interaction between the planting dates and varieties were not found significant statistically at 45 days after planting. However, the highest stem diameter (1.207 cm) was observed in Ajiba followed by Cosmos (1.18 cm), when the crop was planted on Oct.-2 and Sept.-20 respectively, whereas the minimum stem diameter (0.877 cm) was found when planted on Oct.-26th in Zafira at 45 days after planting.

Stem diameter at 60 days after planting

The stem diameter had highly significant variation with planting dates. However, the difference among the varieties and their interaction with planting dates was not found significant statistically. The Ajiba variety had maximum stem diameter (1.208 cm) followed by Cosmos and Zafira with 1.18 cm and 1.12 cm, when crop was planted on Oct.-2 and Sept.-20 respectively, while the minimum stem diameter (0.94cm) was obtained in Ajiba when planted on Oct.-26th (Fig. 2). Generally the stem diameter was appeared to reduce with planting delay to Oct.-26th. The decrease in temperature, physiological age and stem number resulted to decrease stem diameter. This result is in agreement with the finding of Farooq *et al*. (1989) who reported that normally when seed is getting very old it produces week stems. The maximum average stem diameter was found when crop was planted on Sept.-20 among the four planting dates. Among the three varieties maximum stem diameter was recorded in Cosmos. Negative correlation with stem diameter was found as stem number plant^{-1} increased the diameter of the stem decreased.

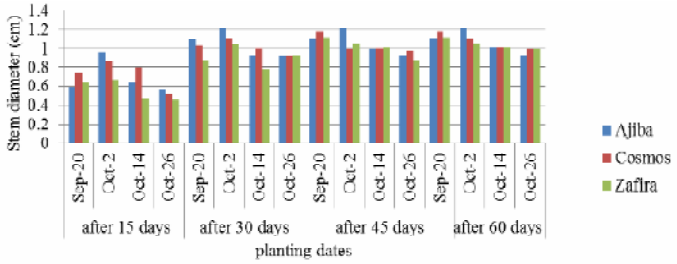

Fig. 2. Effect of planting dates and varieties on plant stem diameter

Yield attributing parameters

Average number of tubers (plant^{-1})

Data recorded on total number of tubers plant^{-1} harvested is presented in table 1 which revealed very high significant ($p<0.001$) response to the three varieties used and significant response to all four planting dates ($P<5\%$).

Interaction between the planting dates and varieties showed no significant difference. Average values from all four planting dates showed that maximum number of tubers obtained with planting on Oct.-2, whereas minimum tuber number was obtained from the last planting date. Among three varieties the maximum average number of tuber was recorded by Ajiba, followed by Cosmos. Old seed tubers produced less tuber number stem^{-1} than younger seeds. These results are in agreement with the findings of Struik and Wiersema (1999). With Zafira and Ajiba maximum numbers of tubers were produced when crop was planted on Oct.-2. Seed tubers planted on Oct.-26 resulted in least number of tubers count plant^{-1} except Zafira. Maximum number of tubers plant^{-1} (13.73) was recorded by Cosmos, when the crop was planted on Oct.-14 followed by Ajiba (13.33) from second planting date that was Oct.-2. The possible reason for their highest tuber numbers of the two varieties could be due to more numbers of stems. These results are in agreement with the findings of Thornton *et al*. (2007) in which it was reported that the number of stems produced by a potato plant is directly related to the number of tubers that plant set. Because each stem tends to produce a certain number of tubers, the higher the number of stems, the more tubers that will be produced by each plant. The Cosmos variety held the most tubers with 13.73 and Ajiba with 13.33 tubers plant^{-1}, when the crop was planted on Oct.-14 and Oct.-2 respectively. Whereas, the number of tubers plant^{-1} was recorded minimum for the three varieties when was planted on Oct.-26th except for Zafira that had the least tubers at the third (Oct.-14) planting dates due to the less number of stems produced by disease problems (collar rot). With Ajiba stem number produced when the crop was planted on Oct.-2 and Oct.-14 was equal but number of tubers produced and the average tuber weight was less when planted on Oct.-14 than tubers harvested from Oct.-2. The main stem number increased from the first (Sept.-20) planting date to the last planting date (Oct.-26th) because it was observed delay planting gave enough opportunity for producing active sprouts on seed tubers and the number of main stems were increased, which led to the production of more tuber. The results obtained from this study were in agreement with those obtained by Darini *et al.* (2013). These variations could be due to the result of differences in the time of planting and temperature, *etc.* Temperature probably has one of the greatest effects. At high temperatures increased respiration rates means that solids accumulated through photosynthesis are burnt up more quickly than they were formed, resulting in a decreased yield. The result obtained from this study was in accordance with those results obtained by Khan *et al*. (2011).

Tuber number was found positively correlated with stem number, stem diameter, plant height, leaf number, leaf area and tuber yield but it was found significant negative correlation between tuber number and tuber weight. The correlation of tuber number with plant height was significant positive correlation.

Average tuber weight (g)

Data presented in table 1, the variance of analysis showed the significant influence of varieties and planting date on the average weight of the tubers of the three varieties. But the difference between their interaction (planting date and varieties) was not statistically meaningful. Maximum average tuber weight was observed with planting on Oct.-2 among all the four planting dates, whereas among the varieties the maximum average tuber weight was recorded with Zafira followed by Cosmos. Even though the difference of varieties with their interaction was not found significant but among three varieties Zafira with 187.3 g had the highest tuber weight plant^{-1} followed by Cosmos with 152.3 g when crops were planted on Oct.-14 and Oct.-2 respectively. The impact of planting date on the average tuber weight with delay in planting from Oct.-14 planting date to the fourth planting date that was Oct.-26, which decreased the average tuber weight significantly. These results are supported by the findings of (Dehdar *et al*., 2012, Bagheripour *et al*., 2012, and Van Der Zaag, 1992). The highest average tuber weight that obtained when crop was planted on Oct.-14 by Zafira was due to the less number of stems and tubers produced plant^{-1} which then increases average tuber weight plant^{-1}. Generally, it was observed the increase stem number from Sept.-20 to Oct.-2 planting date and the tuber number and average tuber weight was observed to increase but planting delay from planting on Oct.-14 increased stem number but decreased tuber number and average tuber weight except with Zafira. This report is also supported fairly by the finding of Taheri and Shamabadi (2013), in that it was reported increasing number of tubers, decreased tuber weight. This finding also in agreement with the work of Lemaga and Caesar, 1990, who reported that there was a significant decrease in average tuber weight with increase number of stems plant^{-1}. There was a significant negative correlation between tuber number and tuber weight, and increasing the number of tubers, decreased tuber weight. The results obtained from this study were in accordance with those obtained by Taheri and Shamabadi (2013) and Laei *et al*. (2012).

Tuber yield plant^{-1} (kg)

The effect of planting date on tuber yield plant^{-1} was highly meaningful statistically (Table 1), but the difference

among the varieties and the interaction between planting date and varieties was not significant statistically. Among the three varieties highest average tuber yield plant^{-1} was recorded by Cosmos and among all four planting dates average tuber yield plant^{-1} was highest with planting on Oct.-2. The delay in planting from Sept.-20 to Oct.-26 resulted to decrease the yield significantly. These results are supported by the findings of Bagheripour *et al.* (2012) and Darini *et al.* (2013). Delay in planting may be along with the reduction of aerial plant organ caused due to the fluctuation in environmental conditions. The highest tuber yield plant^{-1} (1.543 kg) was obtained when crops was planted on Oct.-2 by Ajiba, whereas, the lowest (0.567 kg) was obtained when planted on Oct.-26. The reason for highest tuber yield for Ajiba variety could be due to more numbers of tubers produced. These results are in agreement with the findings of Thornton *et al*. (2007). There was a highly significant positive correlation between tuber yield plant^{-1} and tuber weight. Increasing the weight of tubers, increased tuber yield plant^{-1}. Increasing physiological age, increases the yield plant^{-1} to all the varieties up to the third planting dates but except Cosmos. The yield plant^{-1} for the remaing two cultivars was higher at second planting date (Oct.-2) and again yields plant^{-1} was observed to decrease with increasing age at the end of harvest period with crop planted on Oct.-26 (fourth planting date). Similar results were also reported by the findings of Harries (1992). In that it was reported that total yield increases with increasing age at early harvests, might be unaffected by the age at harvest at the middle of the season and decrease with increasing age at the end of harvest period.

The coefficient of variance of some treatment results was very large this might be due to the soil type difference of the field experiment.

Total tuber yield

The analysis of variance showed that the effect of planting date on tuber yield hectare^{-1} was highly significant. The difference among various dates of planting on the total tuber yield hectare^{-1} of the three varieties was significant. But difference among the varieties and the interaction between planting date and varieties was not found significant statistically (Table 1). Among the planting dates highest average yield hectare^{-1} was obtained with crop planted on Oct.-2 while among the varieties highest average yield hectare^{-1} was produced by Cosmos. Planting delay from Sept.-20 to Oct.-26 resulted to decreases the yield significantly. These results are supported by the findings of Bagheripour *et al*. (2012), Khan *et al*. (2011). They also reported that yield declined with delay in planting. Delay in planting may be along with the reduction of aerial plant organ due to the change in environmental conditions, which reduced to transform sufficient energy to the tubers. Old seed tubers also produced less yield, as compared to young seed tubers (Struik and Wiersema, 1999), therefore this is additional reason to yield reduction with delay in planting. The highest tuber yield hectare^{-1} (73.5 t ha^{-1}) was obtained by potato variety Ajiba, when crop was planted on Oct.-2 followed by Cosmos (71.7 t ha^{-1}) when planted on Oct.-14 (third planting date). Whereas, the lowest yield (27 t ha^{-1}) was obtained by Ajiba, when planted on Oct.-26. These results are supported by the findings of Hassanpanah *et al*. (2009). From all the four planting dates it was found that on Sept.-20th, Oct.-14th and Oct.-26th planting dates in Cosmos variety gave higher yield and also produced more number of tubers, when the crop was planted on Oct.-14 and Oct.-26. These results are supported by the findings of Farooq *et al*. (1989) and Hassanpanah *et al*. (2009). The possible reason for the highest yield for Ajiba at second planting date (Oct.-2) and for Cosmos at third planting date (Oct.-14) could be also due to more number of stems that leads to more tubers

Table 1. Effect of planting dates and varieties on tuber number, tuber weight, tuber yield plant^{-1} and total tuber yield (t ha^{-1}) at different dates of planting

Treatment		Tuber no. (plant^{-1})	Tuber wt. plant^{-1} (g)	Tuber yield plant^{-1} (kg)	Total yield (t ha^{-1})
Variety	Ajiba	12.12^{a}	89.2^{b}	0.989	47.1
	Cosmos	11.03^{a}	118.6^{ab}	1.207	57.5
	Zafira	8.63^{b}	143.6^{a}	1.087	51.8
	LSD	1.581	32.72	NSNS	
Planting date	Sept.-20	10.82^{a}	101.3^{bc}	0.940^{bc}	44.8^{bc}
	Oct.-2	11.40^{a}	147.2^{a}	1.472^{a}	70.1^{a}
	Oct.-14	11.31^{a}	133.1^{ab}	1.257^{a}	59.8^{a}
	Oct.-26	8.84^{b}	86.9^{c}	0.710^{c}	33.8^{c}
	LSD	1.826	37.78	0.2814	13.4
Interaction	Ajiba Sept.-20	13.07	63.8	0.787	37.5
	Ajiba Oct.-2	13.33	138.7	1.543	73.5
	Ajiba Oct.-14	12.87	91.0	1.060	50.4
	Ajiba Oct.-26	9.20	63.5	0.567	27.0
	Cosmos Sept.-20	10.4	120.5	1.133	54.0
	Cosmos Oct.-2	10.47	152.3	1.447	68.9
	Cosmos Oct.-14	13.73	120.9	1.507	71.7
	Cosmos Oct.-26	9.53	80.5	0.743	35.4
	Zafira Sept.-20	9.00	119.7	0.900	42.9
	Zafira Oct.-2	10.40	150.6	1.427	67.9
	Zafira Oct.-14	7.73	187.3	1.203	57.3
	Zafira Oct.-26	7.80	116.7	0.820	39.1
	LSD	NS	NS	NS	NS
	CV%	11.3	33.0	26.3	23.2

Means having same letter(s) in a column are not significantly different according to) LSD) at P = 0.05.

plant^{-1}. This is also in agreement with the findings of Harries (1992) in which it was reported that yield increases with increasing stem number density and then either remains unchanged with further increase in density or eventually begins to decline. Present finding is also in agreement with the work of Lemaga and Caesar (1990), who concluded that tuber number and total tuber yield plant^{-1} increased significantly with increasing number of stems plant^{-1}. There was also a consistent and significant decrease in average tuber weight with increase stem, but this is not agreed by findings of Abubaker *et al*. (2011). They studied impact of variety and growing season on potato and the result was indicated that the increase in number of stems per plant didn't increase total yield. There was a significant positive correlation between tuber number and total tuber yield, and increasing the number of tubers, increased total tuber yield. Negative correlation with yield was found as the date of planting delayed to Oct.-26. These results are supported by the findings of Farooq *et al*. (1989).

The performance of Cosmos variety was better in all planting dates except at the second planting date (Oct.-2nd) as compared to the other two varieties. The superiority could be due to the production of more aerial organ like increase better plant height more transferring the photosynthetic materials to underground stems or due to the fact that more vegetative growth parts including more leaves that produces more photosynthates and may due to better adaption of the investigation area could be mentioned as possible reason for its highest yield stand of the Cosmos variety. These results are in conformity with findings of Bagheripour *et al*. (2012) and Abubaker *et al*. (2011).

Thus, it can be concluded that planting date is the main factor that has significant role in potato cultivation. Because of this reason and based on the results obtained by determining the most appropriate time for planting and selection an appropriate variety for planting in Hamelmalo area is so vital for producing high yield. Further, it can be concluded that Cosmos variety is suitable with planting date of 14th October for cultivation in Hamelmalo region and for Ajiba and Zafira varieties planting date of 2nd October is recommended. Thus, cultivation in Hamelmalo refers more to the judicious choice of planting date. Delay in planting to Oct.-26 decreased the vegetative growth of potato. Because of this, tuber yield of the three varieties decreased markedly and could not produce as much as their potential.

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Management of gall midge (*Orseolia oryzae* Wood Mason) through plant nutrients supplied through organic and inorganic sources with emphasis on *neem* and *karanj* cakes

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ABSTRACT

Rice (*Oryza sativa* L.) is the most important food crop of India including the state of Jharkhand. Among the half of a dozen of major insect pests of rice prevailing in the state, gall midge (*Orseolia oryzae* W.M.) is one of them. Due to feeding by the maggot of the insect on the growing point (of central leaf sheath) of rice plant, silver shoot (SS) is produced, resulting failure of emergence of panicle in the affected tiller, which causes direct yield loss. The present experiment was conducted for two consecutive years, 2013 and 2014 in wet season with the sole objectives for management of gall midge through use of organic and inorganic form of plant nutrients and their appropriate and balanced combination in right quantity. The overall results of the two years experimentation revealed that *neem* cake @ 2.5 t ha^{-1} proved to be the most effective in minimizing the incidence of gall midge (incidence 0.75% SS). Result of *karanj* cake @ 2.5 t ha^{-1} remained at par (incidence 1.42% SS). It was interesting to note that *neem* and *karanj* cake applied @ 0.5 to 2.5 t ha^{-1} were superior over the other treatments in reducing the pest incidence. The highest incidence of gall midge (10.78% SS) was recorded in case of rice plants receiving sole application of nitrogen @ 80 kg ha^{-1} supplied through urea. It was almost at par with rice plants receiving all the three N, P, K @ 80, 40 and 20 kg ha^{-1} respectively (RDF) (silver shoot incidence of 9.78%). The highest yield (41.50 q ha^{-1}) was obtained in treatment *i.e.* sole use of *neem* cake @ 2.5 t ha^{-1}, which was at par with sole use of *karanj* cake applied @ 2.5 t ha^{-1} (38.60 q ha^{-1}). Even the minimum dose of *neem* and *karanj* cake (@ 0.5 t ha^{-1}), each separately applied, with supplementary application of N, P, K) could be able to reduce the silver shoot incidence upto appreciably lower level and higher grain yields of 35.90 and 34.20 q ha^{-1}, respectively. The, rice plant receiving sole nitrogen @ 80 kg ha^{-1} through urea had lowest grain yield of rice (22.40 q ha^{-1}).

Key words: Rice, gall midge, INM, NPK, *neem* cake, *karanj* cake, IPM, pest management, yield

Rice (*Oryzae sativa* L.) is the major staple food crop of Jharkhand. It is grown in the state in around 18 lakh hectares. Among the half of a dozen of major insect pests of rice prevailing in the state of Jharkhand, gall midge (*Orseolia oryzae* W.M.) is one of them. It causes loss in yield of the crop to the tune of 10 to 25 per cent (Prasad and Prasad, 2006). Plant nutrients applied through *neem* cake, *karanj* cake, FYM, green manure or their combined use to meet the requirement of appropriate proportion of NPK from inorganic source could be instrumental in suppressing the pest intensity upto the desirable level. Moreover, plant nutrients supplied through organic source (s), with more emphasis on use of *neem* and *karanj* cake and also in combination with those of inorganic source (s) may be of immense value in improving the soil health and fertility without harming the agro-ecological conditions as well as useful for pest management. Information on all these aspects of pest management is lacking. Hence, the present field investigation has been undertaken to meet this noble objectives for sustainable cultivation of rice through integrating organic combinations.

MATERIALS AND METHODS

A field experiment was conducted in the Rice Research Farm of Birsa Agricultural University, Ranchi in wet season for two consecutive years, 2013 and 2014 with rice variety, Pusa Basmati-1 in plot size of 5 x 4 square meters. The experiment was executed in the randomized block design with 13 treatments and 4 replications (Table 1). Date of seed sowing was 2nd July and date of transplanting was 26th July during both the years (2013 and 2014). Harvesting was done on 28th and 30th of November during 2013 and 2014, respectively. The treatments comprised of certain organic manures *viz.* FYM (Farm yard manure), green manure (GM) in the form of *dhaincha* (*Sesbania rostrata* L.), vermi-compost (VC), *neem* (NC) and *karanj* cake (KC) and recommended dose of chemical fertilizers (RDF) in the form of their sole and separate use and also in the form of their balanced combinations (Table 1). In addition, one treatment, without manures and fertilizers was kept for comparison as control. No additional plant protection measures were provided for raising the crop.

Observations on the incidence of gall midge were recorded in the form of percentage of silver shoot. Total number silver shoot (SS) and number of tillers were counted on 10 randomly selected hills (plants) in each treatment and replication for calculating percentage of silver shoot (SS%) by applying following formula :

$$SS\ (\%) = \frac{\text{Total number of silver shoots (SS)}}{\text{Total number of tillers (SS + healthy tillers) in 10 hills}} \times 100$$

Observation on the incidence of gall midge (in term of per cent of silver shoot) were recorded at 20, 35, 50 and 60 days after transplanting (DAT) and mean of percentage of silver shoot was calculated. Yield was recorded at harvesting of the crop. Ultimately, the pooled data were subjected to the statictical analysis for their interpretation and documentation.

RESULTS AND DISCUSSION

Incidence of rice gall midge *Orseolia oryzae* (Wood Mason)

A perusal of data in table 1 revealed that the gall midge incidence in terms of percentage of Silver Shoot (SS%) was found to increase with the age advancement of rice plants right from 20 DAT to 50 DAT and thereafter it declined during the cropping season.

It was general observation that *neem* cake (NC) applied @ 2.5 t ha^{-1} proved to be the most effective, which was statistically at par with *karanj* cake (KC) @ 2.5 t ha^{-1}. It was also observed that highest incidence of SS was recorded, when nitrogen was applied through inorganic source (@ 80 kg ha^{-1}) followed by in the treatment of recommended dose of N : P : K @ 80 : 40 : 20 kg ha^{-1} (chemical fertilizer). It is interesting to note that almost all the organic sources of plant nutrients *viz*., *neem* cake, *karanj* cake, vermi-compost, green manure and FYM were superior in reducing the incidence of gall midge as compared to those of inorganic sources (chemical fertilizers) in the form of RDF or through sole use of urea. As far as the efficacy of *neem* and *karanj* cakes is concerned, even the lowest dose of these cakes (i.e., @ 0.5 t ha^{-1}) proved significantly superior over the other treatments.

Gall midge incidence recorded at 20 DAT

The silver shoot incidence ranged from 0.09 to 7.82 per cent in the different treatments. The lowest SS of 0.09 per cent was recorded in case of rice plants receiving *neem* cake (NC) applied @ 2.5 t ha^{-1} and *karanj* cake @ 2.5 t ha^{-1} (0.30% SS), which were at par. It was followed by the treatment *neem* cake @ 1.0 t ha^{-1} (0.85% SS), NC @ 0.5 t ha^{-1} (1.25 % SS). It was interesting to note that *neem* cake applied @ 0.5 t ha^{-1} remained statistically at par. Next to *neem* cake (@ 0.5 to 2.5 t ha^{-1}, *karanj* cake @ 0.5 to 2.5 t ha^{-1} proved significantly effective in suppressing the incidence of silver shoot having 0.30 to 1.76 per cent. *Neem* and *karanj* cake applied @ 0.5 to 2.5 t ha^{-1} provided statistically superior protection to rice plants against gall midge as compared to those of other organic manure *viz*. FYM (3.43% SS) and vermi-compost (2.05% SS), FYM (3.43% SS) and green manure (3.68% SS). FYM @10 t ha^{-1} and green manure (GM) @ 10 t ha^{-1} remained statistically at par (silver shoot incidence of 3.43 and 3.68 per cent, respectively).

It was quite encouraging to note that all the organic treatments had lower incidence of silver shoot (0.09 to 3.68%) as compared to those of inorganic treatments (6.53 to 7.85%). However, in treatment of N applied through urea had highest incidence of the silver shoot *i.e*. 7.82 per cent. Even, the combination of green manures, GM @ 10 t ha^{-1} + 50% RDF (*i.e*., NPK @ 40 : 20 : 10 kg ha^{-1}) had lower gall midge incidence (4.15%) as compared to full dose of RDF *i.e*., NPK @ 80 : 40 : 20 kg ha^{-1} (6.53%).

Gall midge incidence recorded at 35, 50 and 60 DAT

Gall midge incidence, in terms of silver shoot formation, was found to gradually increase from 20 to 50 DAT with advancement of growth and age of plant and after that it began to decrease at 60 DAT. However, efficacy of different treatments recorded at 35 and 50 DAT's in suppressing the formation of silver shoot (SS%), caused by gall midge, received almost the similar trends as that of the pest suppression noticed at 20 DAT, however, on 60 DAT were with lower value but the trend was as of 20 DAT (Table 1).

The data of overall mean of all the four observational dates, *viz*. 20, 35, 50 and 60 DAT's of the two year's (2013 & 2014) results also received almost the similar trends of gall midge incidence as influenced by various organic manures and inorganic fertilizers and their combinations in respect of the pest incidence of gall midge, recorded at 20 DAT in the present studies.

Earlier, Rani *et al*. (2007) advocated that *neem* cake treated rice plants had the lowest incidence of silver shoot. Recently, Prasad *et al*. (2008) found more or less similar results with *neem* and *karanj* cakes applied @ 2.5 t ha^{-1} in rice scented variety, Birsa Mati. Their findings also revealed that azadirachtin in *neem* cake and karanjin in *karanj* cake could be responsible for the pest suppression. Owing to the aforesaid insecticidal principles in *neem* and *karanj* cakes, their varying doses (0.5 to 2.5 t ha^{-1}) were effective in reducing the incidence of gall midge in the present studies.

Table 1. Effect of organic and inorganic sources of plant nutrients on the incidence of gall midge (*Orseolia oryzae* Wood Mason) infesting rice (Pusa Basmati - 1) (Based on pooled mean of experimental results of 2013 and 2014)

Treatment and dose	Percentage of Silver Shoot (SS) caused by gall midge at				Overall mean of SS %	Yield of rice grains (q ha^{-1})
	20 DAT	35 DAT	50 DAT	60 DAT		
100% RDF : NPK (80 : 40 : 20) kg ha^{-1}	6.53 (14.77)	9.98 (18.0)	15.35 (23.07)	7.25 (15.62)	9.78 (17.87)	35.65
FYM @10 t ha^{-1}	3.43 (10.63)	6.33 (14.54)	9.35 (17.81)	3.22 (10.31)	5.12 (12.76)	28.70
Green Manure (GM) @ 10 t ha^{-1}	3.68 (11.02)	5.78 (12.87)	6.75 (15.06)	2.95 (9.89)	5.51 (13.25)	26.56
GM + 50% RDF, N as top dressing in two splits	4.15 (11.76)	5.44 (13.44)	8.65 (17.11)	2.45 (9.01)	6.50 (14.50)	31.80
Vermicompost @ 2.5 t ha^{-1}	2.05 (8.23)	3.52 (10.78)	5.05 (12.99)	2.20 (8.53)	8.61 (10.20)	27.60
Karanj cake @ 2.5 t ha^{-1}	0.30 (3.14)	0.78 (5.07)	2.25 (8.63)	0.75 (4.97)	1.42 (6.13)	38.60
Neem cake @ 2.5 t ha^{-1}	0.09 (1.72)	0.12 (1.99)	1.88 (7.82)	0.13 (2.07)	0.75 (3.79)	41.50
KC @ 0.5 t ha^{-1} + NPK (40 : 40 : 20 kg ha^{-1}) rest quantity of N comes through inorganic sources *	1.76 (7.60)	2.45 (9.01)	3.86 (11.32)	2.06 (8.23)	3.3 (10.28)	34.20
KC @ 1.0 t ha^{-1} + NPK (40 : 40 : 20 kg ha^{-1}) rest quantity of N comes through inorganic sources *	1.36 (6.68)	1.74 (7.49)	3.15 (10.23)	1.16 (6.16)	1.89 (7.70)	36.60
NC @ 0.5 t ha^{-1} + NPK (53 : 40 : 20 kg ha^{-1}) rest quantity of N comes through inorganic sources *	1.25 (6.42)	2.14 (8.33)	3.86 (11.31)	1.88 (7.82)	2.55 (9.06)	35.90
NC @.1.0 t ha^{-1} + NPK (26 : 40 : 20 kg ha^{-1}) rest quantity of N comes through inorganic sources *	0.85 (5.29)	1.35 (6.68)	2.67 (9.37)	0.99 (5.71)	1.76 (7.55)	37.40
$N_{80}P_0K_0$, Nitrogen = 80 kg, P = 0, K=0 kg ha^{-1}) through inorganic sources*(*i.e.* through urea) in 3 plots	7.82 (16.21)	10.31 (18.72)	16.82 (24.25)	8.16 (16.55)	10.78 (18.93)	22.40
No manures and fertilizers (untreated control)	2.75 (9.55)	5.44 (13.44)	10.78(19.14)	4.76 (12.59)	1.36 (6.58)	8.95
CD (P=0.05)	(1.16)	(0.48)	(1.07)	(0.76)	(0.87)	3.82

Figures under the parentheses are angular transformed values; VC = Vermicompost; KC = *Karanj* cake; NC = *Neem* cake; SS – Silver shoot; DAT - Days after transplanting; RDF - Recommended doses of fertilizer through inorganic sources (i.e., N : P : K : : 80 : 40 : 20 kg ha^{-1}) in the form of chemical fertilizers. *Inorganic sources refer to chemical fertilizers

Grain yield

The results (Table 1) revealed that sole use of *neem* cake @ 2.5 t ha^{-1} resulted the highest yield of 41.5 q ha^{-1}, which was at par with that of the sole use of *karanj* cake @ 2.5 t ha^{-1} (38.60 q ha^{-1}) and followed by *neem* cake @1.0 t ha^{-1} + N, P, K @ 26, 40, 20 kg ha^{-1} (37.40 q ha^{-1}), *karanj* cake @ 1.0 t ha^{-1} + N, P, K @ 40, 20, 20 kg ha^{-1} (36.60 q ha^{-1}), *neem* cake @ 0.5 t ha^{-1} + N, P, K @ 53, 40, 20 kg ha^{-1} (35.90 q ha^{-1}) and 100% RDF : N, P, K @ 80, 40, 20 kg ha^{-1} (35.65 q ha^{-1}). The sole use of N @ 80 kg ha^{-1} through urea resulted to the substantially lower yield of 22.40 q ha^{-1}. The lowest yield of 8.95 q ha^{-1} was obtained from rice plant receiving zero N, P, K from outside (i.e. control).

Hence, it was found that *neem* and *karanj* cake used in the varying doses of 0.5, 1.0 and 2.5 t ha^{-1} proved to be significantly effective in reducing the incidence of rice gall midge (*O. oryzae*) and substantially higher yield. Hence, the present findings suggest that inclusion of *neem* and *karanj* cake as organic manure as a part of INM could be an important component of IPM for the sustainable cultivation of rice. As such, it may be concluded that *neem* and *karanj* cakes applied either @ 0.5, 1.0 or 2.5 t ha^{-1} based on their availability in the region, could be recommended as organic manure not only for supplying plant nutrients but also for protecting rice plants against gall midge for enhancing the grains yield of rice.

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Temporal dynamics of mango flower visitors and their contribution in pollination

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ABSTRACT

Insect pollination is essential for fruit set in mango (in cv. Dushehari). Insects visiting mango bloom were recorded during 2016 in three orchards located in major mango-growing areas in Lucknow. Thirty-four distinct species (identified upto genus level) were found. Most of them belonged to the orders Diptera (16), Hymenoptera (13) and Coleoptera (5). The effectiveness of pollinators was assessed in one orchard. Highest number of flowers were pollinated under open pollination condition (41%) followed by diurnal pollination (33%). There was no fruit (incipient) set on completely bagged panicles.

Key words: Insects, incipient fruit, pollination, flower visitors, mango

Pollination of flowers is an essential step in the sexual reproduction of plants. Major pollinator dependent crops are fruit, vegetable, spices, plantation crops, pulses and oilseeds. Most flowering species rely on insects for transfer of pollen. Lack of sufficient number of suitable pollinators causes decline in fruit and seed production (Partap, 2001). According to Klein *et al*. (2007), insect pollinators are responsible for 35 per cent of global crop-based food production and mango is one of the crop show 80-100 per cent (Hein, 2009) dependency on pollinators. Worldwide, the pollination by wild and managed pollinators has shown a gradual but steady decline. Causes for this decline may be credited to intensification of agriculture, mono-cropping, pesticide, urbanisation, changing climate and reductions in the availability of natural ecosystems for wild pollinators. The overall global warming and change in precipitation have affected phenology of many plant species and associated insect dynamics (Kelly and Goulden, 2008). Distinct changes in air temperature since the end of the 1980s led to clear responses in mango and guava phenology in many parts of India (Rajan, 2009). Reddy *et al*. (2012) in their study noticed a decline in activity of *Apis florae* in mango at temperatures above 32°C.

Mango (*Mangifera indica* L.) plants are andromonocious and its panicles bear two types of flowers i.e. staminate and hermaphrodite. The compatibility of pollen and stigmas cause failure in mango fruit set (Sharma and Singh 1970; Pandey *et al*., 1973; Dutta *et al*., 2013). Usman *et al*. (2001) and Bally *et al*. (2009) reported that cross pollination significantly contributed in mango fruit set. Some researchers have indicated that mango plants are air pollinated (Mallik, 1957; Free and Williams, 1976) but the small size, whitish colour of flowers, fleshy hemispherical disc, presence of nectar, fewer pollen grains and small stigma indicate entomophilous pollination. Observations have shown that mango flowers are visited by various groups of native insects namely Diptera, Hymenoptera Coleoptera *etc.* (Singh, 1997; Dag and Gazit, 2000; Fajardo *et al*., 2009; Kumari *et al*., 2014) and the pollen-grains have been observed adhering to their bodies (Huda *et al*., 2015).

Dushehari is a popular cultivar of mango in northern plains of India. Plants have short period of mass blooming and is strictly insect pollinated. It requires peak pollinator population for successful pollination at blooming period. But as is reported by Rajan (2009) that with the changing climate there is distinct changes in mango phenology, there is possibility of change in diversity and dynamics of pollinators in this region too. Changes in flower-visiting insect populations or communities can be documented by measuring spatial or temporal trends, or by comparing abundance or species composition before and after change. In this regard, a short-term study was planned to re-examine mango pollinators and their contribution in pollination of Dushehari cultivar of mango.

MATERIALS AND METHODS

Systematic notes were taken up in three mango orchards for pollinators in mango cv. Dushehari in the Research farms of Central Institute of Subtropical Horticulture situated at Rehmankhera (26°90' N - 80°76' E) and Telibagh (26°80' N - 80°93' E) during flowering season of year 2016. Two observations were taken at fifteen days interval during January -February months. Pollinators were recorded on 4 panicles per tree from 10 randomly selected trees. For diurnal variation, observations were taken at two-hour interval from 6.00 hrs to 18.00 hrs. To assess the pollinator visitation rate,

observations were taken at 10.00 - 11.00 hrs., period of maximum activity, based on our preliminary observations. Visitation frequency represents the number of single visit flower for each insect species during one min observation period.

To study the pollinator contribution in reproductive success, 48 panicles of similar length and width, were selected when the first flower opened from four trees (3 replication tree^{-1}). Muslin cloth bags (H"45cm × 25cm) were used to cover panicles as per treatment. Treatments were (i) self- pollination – panicles were bagged throughout the experiment period (ii) open pollination – panicles were not bagged (iii) diurnal pollination (panicles were bagged during 6 pm to 7 am and (iv) nocturnal pollination (panicles were bagged during 7 am to 6 pm). Treatments number three and fourth were designed on the basis of the flower type, colour and fragrance, which suggest some possibility of visit of nocturnal pollinators. The experiment was continued for twenty-five days and in the end, all the flowers with mustard size swollen green ovary (incipient fruit set) were counted to mark for the positive pollination.

RESULTS AND DISCUSSION

A total of 34 species of insects were recorded on the blossom of Dushehari mango during observation period. This insect community consist of 16 species of flies, 2 wasps, 6 bees and 9 other insects (Table 1). Thus, flies contributed around (47%) to the pollinator guild followed by bees (17%). Among dipterans *Episyrphus* sp., *Eristalis* sp. were the dominant and stingless bees among the hymenopterans were dominant species (Fig 1). Results reveal that flowers of this plant are predominantly visited by flies and wasps. The blossom visiting insects started their visits early in the morning (between 6.00 hrs to 7.00 hrs) and increase in their numbers by 10.00 hrs -11.00 hrs (Fig. 2). But, there was a sharp decline in the species number and their individuals

Table 1. Mango flower visitors

Order	Family	Species (#)
Hymenoptera	Apidae	5
	Formicidae	1
	Vespidae	2
	Xylocopidae	2
	Unidentified	3
Diptera	Syrphidae	4
	Muscidae	3
	Sarcophagidae	2
	Calliphoridae	3
	Unidentified	4
Coleoptera	Coccinalidae	5
Total		34

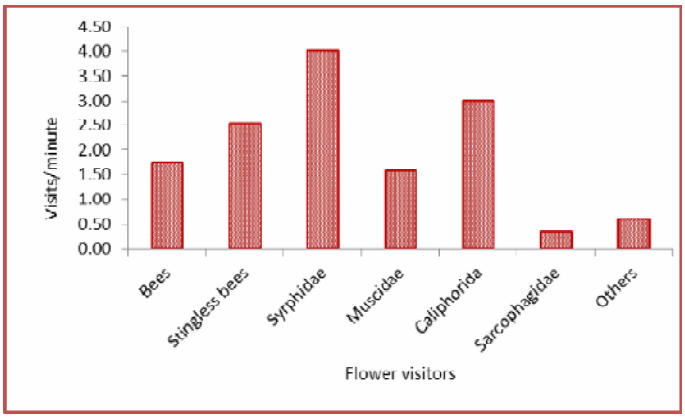

Fig. 1. Pollinators' guild in mango orchard

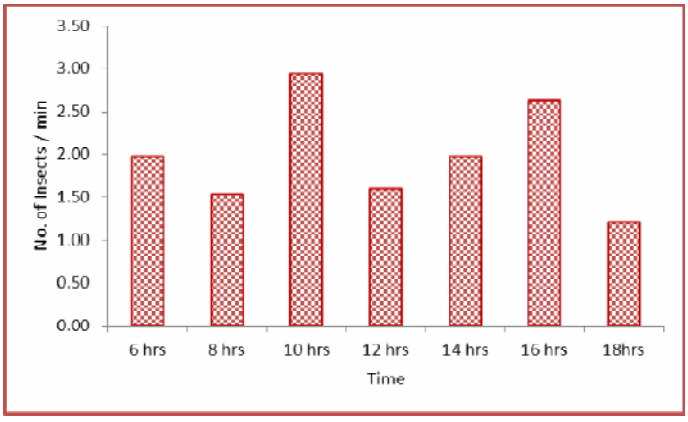

Fig. 2. Diurnal variation in pollinator's abundance

afterwards between 12.00 hrs to15.00 hrs. Again during 16.00 hrs an increase in activity was observed which tended to decline later.

In this study, we have encountered fewer (34) species in comparison to 39 in Southern Taiwan, 46 in Israel and 80 in Australia (Dag and Gazit, 2000). However, this may be due to the survey having been limited places and use of insecticides during the bloom period to control hoppers and other pests. After insecticides spray stingless bees need about 15 days to revisit the orchards. However, species of Syrpidae, Sarcophagidae and Calliphoridae were observed within five days after spray. Bhatia *et al*., (1995), Sharma *et al*., (1998), Dag and Gazit, (2000) and various others have also reported that dipterans and stingless bees are chief pollinators of mango plants. While, Kumari *et al.* (2014) observed that honey bees (*Apisdorsata, A. florae, A. ceranaindica*) are the major pollinators (35%) in mango orchards at Hyderabad.

There is significant temporal variation in pollinator visits in the observed orchards. More number of insects visited the flower during late morning hours followed by a

sharp decline in the noon and resumed activity during early evening hours. Abrol, (1988) indicated that diurnal temperature variation may have substantial influence on pollinator systems by affecting the activity of insects or by altering the volatilization of attractants and nectar flow.

In this study, authors observed significant role of flower visitors in pollination (Table 2). The covered panicles bear few smaller, pale and fragile incipient fruits. While, in open pollination conditions, pollinated flowers had mustard size ovaries and appeared bright green in colour. The proportion of hermaphrodite/perfect flowers to the male/staminate flowers (sex ratio) within the panicles, trees, was less than fifty per cent. In the context of the above information, authors recorded quite high number of flowers were pollinated in open pollination condition (41.3%) and under diurnal pollination (32.8%), where pollination was allowed only during day light period. Tangible fruit yield in mango depend on various plant and climate factors (Chad, 1964; Pandey *et al*., 1973) and the study was planned to have an account of insect's role in pollination hence, mature fruit counts (yield) was not recorded.

Table 2. Flowers pollinated under self and open pollination in Dushehari mango

Treatment	Incipient fruit set panicle[1]
Open pollination	41.3
Nocturnal pollination	15.3
Diurnal pollination	32.8
Self-pollination	9.5
CD (0.05)	9.7

The mango flowers are unspecialized, allowing pollination by most visiting insects (Jiron and Hedstrom, 1986). The removal and deposition of pollen depend on the type of insect, frequency of visit and stay at flower (Sung *et al*., 2006; Huda *et al*., 2015). The yield in mango is significantly enhanced by the hymenopteran and dipteran pollinators (Rafique *et al*., 2016; Saeed *et al*., 2016).

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Status of prevailing insect pest fauna associated with transplanted rice in Jharkhand

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ABSTRACT

Rice (*Oryza sativa* L.) is one of the most important staple food crop of India including the state of Jharkhand. The crop is damaged by several insect pests almost throughout the cropping season in its all the growth stages. The occurrence of the pest species takes place in succession and also in overlapping manner, in varying intensities. Pest spectrum of the crop changes in time and space. Information on the current status of the real pest problem of rice in the state of Jharkhand is lacking in literature. In view of visualizing and exploring information and reckoning the real pest situation in the rice ecosystem in the state, pest survey and surveillance work was conducted periodically throughout the cropping season from 2010-1015 in different regions of Jharkhand. The results revealed that as many as 17 insect pests belonging to 7 orders and 8 families were observed to prevail in the rice agro- ecosystem of the state. Termites and white grub were the root feeders for rice crop in general and in case of upland rice ecologies in particular. Green leaf hoppers, brown plant hopper, white backed plant hopper, thrips and mealy bug behaved as sap suckers. Hispa, grass hoppers, case worm, leaf folders and black hairy caterpillar were the leaf eaters and defoliators. Grub of hispa acted as leaf minor. Larvae of yellow stem borer were found to bore the stem both in vegetative and reproductive stages of the crop. Gall midge was able to form shoot gall from central leaf sheath of the crop plant. Ear bug was able to suck the milk from the developing grains of the crop. Based on the extent and level of crop damage, gall midge, yellow stem borer, rice leaf folder, green leaf hopper, ear bug and caseworm could be categorized as mild to major pest. Hispa could be treated as minor to mild pest. Termite and thrips remained as negligible and minor pest. Black hairy caterpillar, mealy bug, white grub, white backed plant hopper and brown plant hopper, attained the status of negligible pest.

Key words: Rice, insect pest fauna, status, economic significance.

Rice (*Oryza sativa* L.) is one of the most important staple food crop of Jharkhand. It is grown in an area of about 18-19 lakh hectares in the state in rain fed situation. Right from sowing and transplanting to harvesting stages of the rice crop, various types of insect pests are found to attack rice crop both in succession and overlapping manner. They could be able to cause damage in yield of the crop from 21 to 51 per cent in India, varying from area to area as per variation in the agro-ecological conditions (Kalode *et. al.,* 1995). Pest scenario of a crop varies in time and space. More than 100 insect pest species have been reported in India in rice agro-ecosystem (Krishnaiah *et. al*., 2008). Information of real insect pest situation prevailing in rice-ecologies in the state of Jharkhand is lacking. In view of this, pest survey and surveillance was undertaken for consecutive five years (2010-2015) in wet season in the state and the information was generated, which is presented in the present communication.

MATERIALS AND METHODS

Roving and fixed plot survey and surveillance was conducted at fortnightly intervals in different regions of whole state of Jharkhand during last five years (*i.e.* 2010-2015) in wet season in order to explore information on the insect pest scenario and their status associated with rice.

Observations were recorded right from 1st fortnight of June to 2nd fortnight of November during five years (2010-2015). Incidence and intensities of attack of insect pest species and their population across the cropping season were recorded.

A pocket lens (10X) was used for the detection of minute insect species. The pest affected plant materials *e.g*. leaves, stems, panicles and roots *etc*. (whichever necessary) as well as the insect pests were plucked/collected in the polythene bags and glass vials and brought to the laboratory for their close examination and for their estimation in terms of qualitative and quantitative composition in transplanted rice ecosystem in wet season. Population of the insect pest species were recorded by employing the measuring units as mentioned in table 2. Lastly, all the data obtained from different locations (pest wise) according to the incidence, status and succession of the species were pooled together (data of 2010 to 2015).

According to the quantum of abundance or incidence, the pests were classified into: minor, mild, major, severe or key pest.

As some pest species (*i.e.*, stem borer, gall midge, thrip, rice hispa, green leaf hopper, leaf folder, ear head bug *etc*.) in their adult stages required an insect catching (collecting) net, these were collected by sweeping and standard number of sweep were made to collect the data on their population, incidence and intensity. Ultimately the pooled data were analysed on the basis of mean value and interpreted to draw the valid conclusion.

RESULTS AND DISCUSSION

Insect pests' incidence present in rice ecosystem in Jharkhand obtained from the field observations have been presented in table 1 along with the taxonomic position and damaging stages. Table 2 contains the information on fortnightly abundance of incidence of pest species recorded during June-Nov. (pooled mean of 2010-2015).

Incidence, abundance and status of insect pest complex of transplanted rice

It was observed that 17 insect pest species infest rice crop at its different growth stages, almost in overlapping manner (Table 1&2) in the region.

Gall midge (*Orseolia oryzae* Wood Mason)

This insect pest appeared as pest of minor to mild economic status. It's incidence started from 2nd fortnight of July with very low level of incidence in terms of percentage of silver shoot, SS (0.06%). The pest incidence attained its peak (13.6% SS) with hill (plant) infestation of 34.40 per cent in the 2nd fortnight of September and then the pest incidence began to decline from 6.80 per cent SS in the 1st fortnight of October to the minimum level of 4.1 per cent SS with hill damage of 20.40 per cent in 2nd fortnight of October in the present experimentation, which was found to be in agreement with the results of earlier workers (Reissing *et al*., 1985, Rizwi and Singh,1980, Prasad and Prasad, 2011).

Yellow stem borer (*Scirpophaga incertulas* Walker)

This insect pest was noticed as pest of major economic status in rice ecosystem almost right from transplanting to maturity stage of the crop. The pest incidence, in terms of percentage of dead heart (DH), was found to start from the 1st fortnight of July with very low level of 5.30 per cent DH in the early vegetative stage of the crop. The maximum dead heart incidence (15.90% DH) was noticed in the 2nd fortnight of September and declined to the minimum of 2.80 per cent DH in 1st fortnight of October. The pest incidence, in the form of white ear (WE) was, first of all noticed in the 1st fortnight of October, with low level of 4.30 per cent WE. Gradually, the pest incidence increased upto the maximum level of 8.20 per cent WE in 1st fortnight of November. As such, WE (%) ranged from 0.0 to 8.20 per cent. Experimental findings of Saha *et al*. (2005) and Prasad and Prasad (2006) were almost in consonance with the present findings.

Rice hispa (*Dicladispa armigera* Olivier)

This insect was registered as minor to mild pest right from the beginning of the vegetative stage of the crop (*i.e.*, 1st week of July) with lowest level of 1.20 per cent leaf damage due to hispa (HDL%), which gradually increased upto 9.70% HDL in the 1st fortnight of September, 2010. Earlier workers (Kumar *et al*., 2003, Prasad and Prasad (2006) also found similar results.

Leaf folder (*Cnaphalocrosis medinalis* Guenee)

The larval stage of insect was found to cause damage

Table 1. Insect pest incidence in the state of Jharkhand (based on pooled observations of 2010-2015).

Common Name	Scientific Name	Order	Family	Damage stage of insect pest (s)
Yellow rice stem borer	*Scirpophaga incertulas* Walker	Lepidoptera	Pyralidae	Larvae
Rice hispa	*Dicladispa armigera* Ol.	Coleoptera	Chrysomellidae	Adult & grub
Rice case worm	*Nymphula depunctalis* Gn.	Lepidoptera	Pyralidae	Larvae
Rice grass hopper	*Hieroglyphus banian* Fab.	Orthoptera	Acrididae	Adult & nymph
Rice gundhi bug	*Leptocprisa acuta* Th.	Hemiptera	Coredidae	Adult & nymph
Rice gall fly	*Orseolia oryzae* Wm.	Diptera	Cecidomyiidae	Maggot
Rice green leaf hopper	*Nephotettix nigro* pincuts	Homoptera	Cicadillidae	Adult & nymph
Brown plant hopper	*Nilaparvata lugens* Stal	Homoptera	Delphacidae	Adult & nymph
Rice thrips	*Stenchaetothrips biformis* (Bogn.)	Thysanoptera	Thripidae	Adult and nymph
Rice mealy bug	*Heterococous rehi* (Lind)	Homoptera	Pseudococcidae	Nymph and adult
Army worm (Ear cutting caterpillar)	*Mythimna unipunctata* Haw.	Lepidoptera	Noctuidae	Caterpillar
Swarming caterpillar	*Spodoptera mauritia* Boisd	Lepidoptera	Noctuidae	Larvae
Whorl maggot	*Hydrellia griseola* (Fall)	Diptera	Ephydridae	Maggot
Termite	*Odontotermes Obesus* Ramb.	Isoptera	Termiidae	Adult and young

Table 2. Insect pest fauna, their incidence, succession and status infesting transplanted rice in the agro climatic conditions of Jharkhand recorded during, wet season (based on pooled mean of 2010-2015)

Insect pest (s)	Measuring units (s) for the estimation of abundance of the insect pests	Fortnightly abundance of incidence of pest species recorded during June-Nov. (pooled mean 2010-2015)												Range	Overall pest status
		June		July		August		September		October		November			
		1stF	2ndF	1stF	2ndF	1stF	2ndF	1stF	2ndF	1stF	2ndF	1stF	2ndF		
Gall midge *Orseolia oryzae* (Wood Mason)	(i) Percentage of Silver Shoot (SS) due to Gall midge (SS%)	0.00	0.00	0.00	0.06	4.80	7.80	9.60	13.60	6.80	4.70	0.00	0.00	0.00-15.70	Minor-mild
	(ii) Hill (plant) infestation due to gall midge (HI%)	0.00	0.00	0.00	1.98	12.60	18.90	23.40	34.40	20.40	10.00	0.00	0.00	0.00-26.70	
Yellow rice stem borer *Scirpophaga incertulas* Walker	(i) Dead heart DH (%)	0.00	0.00	5.30	9.50	12.20	10.20	13.80	15.90	2.80	0.00	0.00	0.00	0.00-15.80	Major-mild
	(ii) White ear head WEH (%)	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	4.30	7.60	8.20	5.20	0.00-8.20	
Rice hispa *Dicladispa armigera* Olivier	(i) No. of damaged Leaves (HDL) /10hills (plants)	0.00	0.00	2.80	6.20	16.70	20.50	24.30	11.50	0.00	0.00	0.00	0.00	0.00-24.30	Minor-mild
	(ii) Leaf damage (%)	0.00	0.00	1.20	2.70	4.80	8.60	9.70	5.30	0.00	0.00	0.00	0.00	0.00-9.70	
Leaf folder *Cnaphalocrocis medinalis* (Guenee) *N. nigropictus*	(i) No. of damaged leaves (LFDL) /10 hills (plants)	0.00	0.00	0.00	4.90	5.80	12.70	17.30	22.60	28.50	32.60	0.00	0.00	0.00-	Mild –major
	(ii) Leaf damage (%)	0.00	0.00	0.00	0.75	1.80	3.75	6.12	9.30	11.50	10.00	0.00	0.00	0.00-11.50	
Green leaf hopper, (GLH) *Nephotettix virescens* (Distant) *N. nigropictus*	(i) No. of GLH /10 hills (plants) or, No. of GLH /5 Net sweeping	0.00	0.00	0.00	8.86	28.60	55.70	70.60	93.80	125.60	130.60	10.50	0.00	0.00-130.60	Mild –major
Gundhi bug /Ear bug (EB) *Liptocorisa acuta* Thunberg *L. oratorius*	(i) No. of the bugs /10 Hills (plants) Or, No. of bugs /5 Net sweepings	0.00	0.00	0.00	0.00	0.00	0.00	8.70	13.50	24.60	28.30	0.00	0.00	0.00-28.30	Mild –major
	(ii) Grain damage (%)	0.00	0.00	0.00	0.00	0.00	0.00	1.60	3.60	7.90	10.50	0.00	0.00		
Grass hopper *Hieroglyphus banian* Fab. *Oxya chinensis* Thumb.	(i) No. of the insects /10 hills Or, No. of the insects /5 Net sweepings	0.00	0.00	0.70	1.45	2.06	2.15	2.66	3.30	0.96	0.08	0.50	0.90	0.00-3.30	Minor
Black hairy caterpillar *Nisaga simplex* (Wlk).	(i) Black hairy caterpillar *Nisaga simplex* (Wlk) /MRL	0.00	0.00	0.80	1.20	0.86	0.56	0.00	1.30	0.66	0.00	0.00	0.00	0.00-1.30	Negligible
Caseworm *Nymphula depunctalis* (Guenee)	(i) Percentage of leaf damage	0.00	0.00	0.00	0.00	7.90	8.80	6.80	18.60	0.00	0.00	0.00	0.00	0.00-18.60	Mild –major
Termite (ant) *Odontoterme obesus* Ramb.	(i) Percentage of plant (hill) damaged in lowland	0.00	0.00	0.00	2.50	2.60	1.80	0.00	0.00	0.00	0.00	0.00	0.00	0.00-2.60	Negligible to minor
	(ii) percentage of plant damage in medium land	3.80	0.00	1.40	1.90	0.00	0.00	0.00	1.80	0.00	4.00	4.70	2.80	0.00-4.70	
Rice mealy bug *(Brevennia rehi (Lindinger)*	(I) Percentage of leaf damage bearing cottony growth in leaves	0.00	0.00	0.00	0.08	0.20	0.80	1.30	1.60	2,08	0.00	0.00	0.00	0.00-2.08	Negligible
Thrips *Stenchaetothrips biformis* (Bagnall)	(I) Percentage of plant (hill) damage Showing narrow rolling or pinning leaves due to thrips attack.	0.00	0.00	0.00	0.04	0.18	0.34	0.60	0.90	1.70	0.00	0.00	0.00	0.00-1.70	Negligible to minor
White grub *Holotrichia* spp.	(I) Percentage of plant (hill) damage showing weathering	0.00	0.00	0.00	0.00	0.09	0.25	1.30	1.80	0.00	0.00	0.00	0.00	0.00-1.80	Negligible
Brown plant hopper (BPH) *Nilaparvata lugens* (Stal.)	(i) No. of the BPH/10 hills	0.00	0.00	0.00	0.00	0.06	0.08	1.20	1.50	0.00	0.00	0.00	0.00	0.00-1.50	Negligible

to the crop from the period of 2nd fortnight of July with minimum level of leaf damage (0.75%). The pest incidence ended in the 2nd fortnight of October with its maximum incidence (10.00% leaf damage). The experimental findings of Kumar *et al.* (2003) and Prasad and Prasad (2006) are almost in agreement with the results of present findings.

Green leaf hopper (*Nephotettix nigropictus* and *N. viresence*)

Both adult and nymphal stages of the pests were found to suck cell sap from leaf of rice plants. The occurrence of the pest was observed with very low level of GLH/10 hills from 2nd fortnight of July, with lower population level of 8.66 GLH/ 10 hills. However, maximum incidence of the pest was found to the tune of 130.60 GLH/10 hills in the 2nd fortnight of October. As such, the pest was rated as mild pest of rice in Jharkhand. Kumar *et al.* (2003) and Prasad and Prasad (2006) expressed more or less similar views.

Gundhi bug (*Leptocorisa oratorius* Fab., *L. acuta* Thunberg)

The adults and nymphal stages of the insect were found to suck milky juice of developing grains of rice. The pest incidence, in terms of number of the bug/10 hills, was found to vary from 8.70 to 28.30 bugs /10 hills, during the milking stage of rice during 1st fortnight of September to 2nd fortnight of October. As such, the pest was rated as mild pest of rice. Prasad and Prasad (2006) and Krishhaiah *et al.* (2008) obtained more or less similar observations.

Grass hopper (*Hieroglyphus banian* Fab., *Oxya chinensis* Thumb.)

Both adults and nymphs were found to cause defoliation to rice plants. The pest rated as minor pest of rice, because the pest incidence started with very low incidence (0.70 insect /10hills) in 1st fortnight of July and a little higher incidence (3.30 insect /10hills) in 2nd fortnight of September. Although, negligible incidence of the pest (0.90 insect /10 hills) was found to occur in 2nd fortnight of November. Prasad and Prasad (2006) expressed similar views. Jhala and Sisodiya (2003) reported that the pest (*H. banian*) is severe form in the agro-climatic conditions of Gujarat.

Black hairy caterpillar (*Nisaga simplex* Walker)

The insect pest appeared as a negligible pest of rice in the present study. As low as 0.80 larvae / MRL (metre row length) was noticed in the 1st fortnight of July, 2010, whereas the highest incidence of 1.30 larvae/ metre row length (MRL) of the pest was registered in 2nd fortnight of September. Prasad and Prasad (2006) reported that the pest is occasional one and sometimes it causes severe defoliation in upland direct sown rice and lesser or negligible damage to mid and low land rice in the agro-climatic conditions of Jharkhand.

Case worm (*Nymphula depunctalis* Gueen)

This insect appeared as a pest of mild status and in low land late transplanted rice. Crop was found to suffer heavy defoliation between 1st fortnight of August (7.90% leaf damage) to 2nd fortnight of September, 2010 (18.60% leaf damage). Prasad and Prasad (2006) expressed almost similar views.

Termite (*Odontotermes obesus* Ramb.)

Termite appeared as a negligible pest for rice grown in transplanted ecologies. Very low pest incidence, ranging from 1.40 to 4.7 per cent of dead heart (DH), was noticed in the medium land. In the low land 1.8 to 2.60 per cent plant damaged due to termite was recorded during July to August. Prasad and Prasad (2006) opined almost similar views.

Rice mealy bug (*Brevinnia rehi* Lindinger)

Mealy bug appeared during the period of middle of July to 1st week of October (0.08 to 2.08 per cent leaf damage), with maximum of 2.08 per cent leaf damage in 1st fortnight of October, 2010. Negligible level of incidence (0.0 to 2.08% leaf damage), bearing cottony growth on leaves were noticed in the present studies. Prasad and Prasad (2006) found almost similar results in rice ecologies of Jharkhand in wet season.

Rice thrips (*Stenchaetothrips biformis* Bagnall)

This sucking pest also appeared as a pest of negligible economic significance with its low level of incidence, ranging from 0.04 to 1.70 per cent of leaf damage, during the period of 1st fortnight of August to 1st fortnight of October. Prasad and Prasad (2006) found almost similar results.

White grub (*Holothrichia* spp.)

The pest appeared as a pest of very negligible form ranging from 0.09 per cent plant damage (1st fortnight of August) to 1.80 per cent plant damage in 2nd fortnight of September. Prasad and Prasad (2006) also observed the pest almost in similar fashion.

Brown plant hopper (*Nilparvata lugens* Stal.)

The insect pest, BPH also appeared as a negligible pest, as the incidence in terms of no. of BPH /10 hills ranged from 0.06 to 1.50 BPH/10 hills in rice ecologies of Jharkhand. Prasad and Prasad (2006) also found that BPH remained the pest of negligible economic significance in state of Jharkhand in general.

Based on the overall mean results of five years' observations, it may be concluded that out of 17 insect pest species, gall midge, yellow stem borer, leaf folder, green leaf hopper and case worm appeared as mild to severe pest

species, hispa and gundhi bug as moderate (mild) pest, whereas other insect pest species *viz.* grass hopper (*H. Banian, Oxya chinesis*), black hairy caterpillar (*N. simplex*), termite (*O. obesus*), mealy bug (*B. rehi*), thrips (*S. biformis*), white grub (*H.* spp.) BPH (*N. lugens*) remained the pest of negligible and minor economic importance in medium and low land transplanted rice ecologies in the Jharkhand region of India.

The finding of the present investigation may be of immense economic significance in planning formulating, devising integrated management of major insect pests of rice for the state of Jharkhand.

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Management of rice leaf folder (*Cnaphalocrosis medinalis* Guen.) through INM with emphasis on use of *Neem* and *Karanj* cake

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ABSTRACT

An experiment was conducted for two consecutive years, 2012 and 2013 in wet season in the agro-ecological conditions of Jharkhand with the sole objective for management of leaf folder (*Cnaphalocrosis medinalis* Guen.) through use of organic and inorganic form of plant nutrients and their appropriate and balanced combinations in the right quantity with more emphasis on application of *neem* and *karanj* cake. The overall results revealed that *neem* cake @ 2.5 t ha^{-1} proved to the most effective in minimizing the incidence of rice leaf folder and *karanj* cake remained at par. As such, the minimum incidence of the pest interval of leaf damage due to leaf folder (LDLF) 10^{-1} hills (or plants) was obtained through sole use of *neem* cake @ 2.5 t ha^{-1} and the efficacy of *karanj* cake @ 2.5 t ha^{-1} remained at almost at par, resulting in as low as 6.36 LDLF 10^{-1} hills (*i.e.* plants) of rice followed by *neem* and *karanj* cake kept as separate treatment @ 0.5 and 1.0 t ha^{-1}, which in turn remained at par in themselves, in reducing the incidence of the pest. The maximum incidence of leaf folder (24.18 LDLF 10^{-1} hills) on rice plants receiving the sole use of N @ 80 kg ha^{-1} (through urea) was registered as against significantly of recommended dose of fertilizer, RDF (N, P, K @ 80, 40, 20 kg ha^{-1}) in inorganic form, which was further lowered down up to the lower level of the pest incidence of 15.66 LDLF 10^{-1} plants through use of reduction in RDF upto 50 per cent, but supplemented with green manure, @ 10 t ha^{-1} in the form of *Sesbania rostrata* L. (dhaincha). The results indicated that sole use of *neem* cake @ 2.5 t ha^{-1} realized the maximum grains yield of rice (41.50 q ha^{-1}) which, in turn remained at par with sole use of *karanj* cake @ 2.5 t ha^{-1} (38.60 q ha^{-1}) followed by *neem* cake @ 1.0 t ha^{-1} plus N, P, K @ 26, 40, 20 kg ha^{-1} in inorganic form (37.40 q ha^{-1}), *karanj* cake @ 1.0 t ha^{-1} plus N, P, K @ 40, 40, 20 kg ha^{-1} in the form of inorganic source (36.60 q ha^{-1}); *neem* cake @ 0.5 t ha^{-1} + NPK @ 53, 40, 20 kg ha^{-1} (through chemical fertilizers (35.90 q ha^{-1}). The sole use of nitrogen @ 80 kg ha^{-1} applied to the plants through urea received the maximum incidence of the pest (*Cnaphalocrosis medinalis* Guen.) resulting in substantially lower grains yield of rice (22.40 q ha^{-1}).

Key works : Rice, leaf folder, *neem* cake, *karanj* cake, NPK, RDF, INM, IPM, yield.

Rice (*Oryza sativa* L.) is one of most important food crop of India including the state of Jharkhand. Out of half of a dozen major prevailing insect pests in the state of Jharkhand, leaf folder (*Cnaphalocrosis medinalis* Guen.) is one of the important pests. Larvae of the pest usually fold the leaves of rice plants and live within the leaf folds. The larvae feed on the chlorophyll content of upper surface of leaf, while living within the leaf folds. As such, photosynthetic area is considerably reduced, which ultimately results in reduction in yield of the crop. The pest is responsible for loss in yield ranging from 10 to 20 per cent. The loss in yield is more pronounced when the larvae feed on the boot leaf compared to the other lower leaves of the plants (Prasad and Prasad, 2006; Krishnaiah *et al*., 2008). Although, use of chemical insecticides is highly effective against the pest, but it has several ill and after effects such as eradication of natural enemies of pest, secondary pest outbreak, pest resurgence, environmental hazards and residue in crop produce. *Neem* and *karanj* cake and other organic manures as their sole use or in combination with the supplementary use may play significant role in suppressing the pest species without harming the agro-ecosystem. Information on these aspects are lacking in literature in general and with particular reference to rice. Hence, the present field investigation was undertaken to generate the required information for ecofriendly management of rice leaf fold.

MATERIALS AND METHODS

A field experiment was conducted in the rice research farm of Birsa Agricultural University, Ranchi in wet season for two consecutive years, 2012 and 2013 with rice variety, Pusa Basmati-1 in plot size of 5 x 4 square metre. The experiment was executed in the randomized block design with 13 treatments and 4 replications (Table 1). Date of seed sowing of rice (var. Pusa Basmati-1) was on 2nd July and date of transplanting was 26th July during both the years. Harvesting was made on 28 and 30th of November during 2012 and 2013, respectively. The treatment details are given in table 1. The treatments comprised of certain organic manures *viz*. Farm yard manure (FYM), green manure (GM)

in the form of dhaincha (*Sesbania rostrata* L.), vermicompost, *neem* and *karanj* cake and recommended dose of chemical fertilizers (RDF) in the form of their sole and separate use and also in the form of their balanced combinations (Table 1). The observations on the incidence of leaf folder (*C. medinalis*) were recorded by counting the total number the leaf damaged due to leaf folder (LDLF) on 10 randomly selected plants (*i.e.* hills) in each treatment and replications at 45, 65 and 85 dates after transplanting (DAT) during the both years of experimentation. Grain yield was recorded after harvesting during both of the years. Data of both the years were pooled together. The pooled data of the two years experiments were subjected to the appropriate statistical analysis after suitable transformation for their interpretation, documentation and drawing the conclusion.

RESULTS AND DISCUSSION

Leaf folder (*Cnaphalocrocis medinalis* Guenee)

Incidence of leaf folder, in terms of no. of leaf damage due to leaf folder (LDLF) per 10 hills (plants) were recorded at 20 days intervals starting from first observation at 45 days after transplanting (DAT). The results of are presented in table 1.

Incidence of leaf folder at 45 DAT

The incidence of leaf folder was ranging from minimum of 5.14 LDLF 10^{-1} hills in treatment *neem* cake @ 2.5 t ha^{-1} to the maximum of 21.94 LDLF 10^{-1} hills in N @ 80 kg ha^{-1} through urea. Rice plants treated with *karanj* cake (5.46 LDLF 10^{-1} hills) remained at par, followed by *neem* cake @ 1.0 t ha^{-1} (7.52 LDLF 10^{-1} hills) and *karanj* cake @ 1.0 t ha^{-1} (7.53 LDLF 10^{-1} hills), which in turn, remained at par with each other in terms of reducing the incidence of the pest. The rice plants receiving N, P, K through *neem* cake (@ 0.5 t ha^{-1}) proved superior to the same dose of *karanj* cake (*i.e.*, 0.5 t ha^{-1}) and both NC and KC, separately, applied @ 0.5 t ha^{-1} proved significantly more effective in reducing the incidence of leaf folder as compared to those of vermicompost @ 2.5 t ha^{-1},

Table 1. Effect of organic and inorganic sources of plant nutrients on the incidence of leaf folder (*Cnaphalocrocis medinalis)* infesting rice (var. Pusa Basmati-1). (Based on pooled mean of experimental results of 2012 and 2013)

Treatment combination	No. of damaged leaf LDLF 10^{-1} hills			Overall mean	Yield of grains
Treatment and dose	45 DAT	65 DAT	85 DAT		(q ha^{-1})
T_1 - Use of 100% RDF: NPK (80 : 40 : 20) kg ha^{-1}	20.35	24.65	24.52	22.62	35.65
	(26.82)	(29.76)	(29.67)	(28.37)	
T_2 - Use of FYM @10 t ha^{-1}	18.13	21.33	23.15	20.47	28.70
	(25.18)	(27.49)	(28.76)	(26.87)	
T_3 - Use of green manure (GM) @ 10 t ha^{-1}	15.48	17.65	20.52	17.88	26.56
	(23.15)	(24.84)	(26.92)	(24.97)	
T_4 – GM + 50% RDF, N as top dressing in two splits	14.68	15.45	16.84	15.66	31.80
	(22.51)	(23.15)	(24.20)	(23.28)	
T_5 - Vermicompost, VC @ 2.5 t ha^{-1}	12.14	13.48	14.36	13.32	27.60
	(20.36)	(21.52)	(22.26)	(21.38)	
T_6 - *Karanj* cake, KC @ 2.5 t ha^{-1}	5.46	6.29	8.97	6.36	38.60
	(13.5)	(14.60)	(17.41)	(14,57)	
T_7 - *Neem* cake, NC @ 2.5 t ha^{-1}	5.14	5.13	5.84	5.48	41.50
	(13.05)	(13.05)	(13.94)	(13.49)	
T_8 – KC @ 0.5 t ha^{-1} + NPK (40 : 40 : 20 kg ha^{-1}) *i.e.*, rest	10.79	11.36	12.14	11.46	34.20
quantity of N comes through inorganic sources*	(19.14)	(19.69)	(20.36)	(19.73)	
T_9 – KC @ 1.0 t ha^{-1} + NPK (40 : 40 : 20 kg ha^{-1}) *i.e.*, rest	7.53	8.48	9.36	8.35	36.60
quantity of N comes through inorganic sources*	(15.89)	(16.9)	(17.80)	(16.76)	
T_{10} – NC @ 0.5 t ha^{-1} + NPK (53 : 40 : 20 kg ha^{-1}) *i.e.*, rest	9.73	10.24	10.68	10.21	35.90
quantity of N comes through inorganic sources *	(18.15)	(18.63)	(19.05)	(18.61)	
T_{11} – NC @ 1.0 t ha^{-1} + NPK (26 : 40 : 20 kg ha^{-1}) *i.e.*, rest	7.52	8.16	7.66	8.33	37.40
quantity of N comes through inorganic sources *	(15.89)	(16.59)	(16.06)	(16.73)	
T_{12} - $N_{80}P_0K_O$ Nitrogen = 80kg P_0K_0 kg ha^{-1})	21.94	24.65	25.96	24.18	22.40
through inorganic sources *i.e.*, through urea	(27.92)	(14.60)	(30.48)	(29.39)	
T_{13} - No use of manures and fertilizers	17.82	20.15	21.96	20.37	8.95
(*i.e.*, untreated control)	(24.95)	(26.68)	(27.94)	(26.79)	
CD (P=0.05)	(0.96)	(1.05)	(0.71)	(0.91)	3.82

Figures under the parentheses are square root transformed values; DAT - Days after transplanting; GM - Green manure; FYM - Farm yard manure; KC - *Karanj* cake; NC - *Neem* cake; RDF – Recommended dose of fertilizer through inorganic sources (*i.e.*, N : P : K : 80 : 40 : 20 kg ha^{-1}). *Inorganic sources refer to chemical fertilize

GM (10 t ha^{-1}) + 50% RDF (N, P, K @ 40, 20 & 10 kg ha^{-1}), green manure, GM (10 t ha^{-1}) as well as FYM (10 t ha^{-1}), used separately and independently. In broader sense, it was noticed that all the organic manures provided to the rice plants were found to be instrumental in causing significantly more suppression of leaf folder as compared to those of rice plants receiving RDF (N, P, K @ 80, 40 & 20 kg ha^{-1}) through sources inorganic and inorganic nitrogen @ 80 kg ha^{-1} through urea each applied as separate treatments.

The highest incidence of the pest (21.94 LDLF 10^{-1} hills) was found when N, P, K was supplied through sole use of inorganic nitrogen, N @ 80 kg ha^{-1} in the form of urea even applied in 3 splits. The highest level of the pest incidence (21.94 LFDL 10^{-1} hills) in case of sole use of N @ 80 kg ha^{-1} through urea could be significantly lowered down to the level upto 20.35 LDLF 10^{-1} hills, through use of full RDF (N, P, K @ 80, 40 & 20 kg ha^{-1}), which was further lowered down to the level of 14.68 LDLF 10^{-1} hills by the combined application GM @10 t 10^{-1} hills and 50% RDF (N, P, K @ 40, 20, 10 kg ha^{-1}). All the three doses of *neem* and *karanj* cake (@ 0.5,1.0 to 2.5 t ha^{-1}) proved superior over the use of inorganic fertilizer independently applied in the form of RDF (N, P, K @ 80, 40, 20 kg ha^{-1}) and sole N @ 80 kg ha^{-1} supplied through urea in suppressing the incidence of incidence of leaf folder. Even, the lowest dose of *neem* and *karanj* cake each separately applied @ 0.5 t ha^{-1}, provided superior protection to the crop 9.73 & 10.79 LDLF 10^{-1} hill) over the other forms of organic manures *viz.* vermi compost @ 2.5 t ha^{-1} (12.14 LFDL 10^{-1} hills), GM + 50% RDF (14.68 LDLF 10^{-1} hills), GM (15.48 LDLF $hills^{-1}$) and FYM (18.13 LDLF 10^{-1} hills) in reducing the incidence of leaf folder.

Incidence of leaf folder at 65 and 85 DAT

The incidence of the pest recorded at 65 and 85 DAT as influenced by the use of different organic manures and chemical fertilizer and their combination followed more or less similar trends to that of the observations recovered at 40 DAT (Table-1).

Overall incidence of leaf folder

Based on the overall mean of three observations recorded at 45, 60 and 85 DAT (days after transplanting). It may be interpreted that the pest suppressing capabilities of the test treatments could be more or less similar in fashion to that of the observations registered at 40 DAT (Table 1). As such, *neem* cake @ 2.5t ha^{-1} proved to be the most efficacious and *karanj* cake @ 2.5 t ha^{-1} remained at par followed by *neem* and *karanj* cake followed by *neem* and *karanj* cake @ 1.0 t ha^{-1} and *neem* and *karanj* cake @ 0.5 t ha^{-1} which, in turn remained at par, in reducing down the incidence of leaf folder.

The highest incidence of leaf folder (24.18 LDLF 10^{-1} plants) on rice plants receiving sole use of N @ 80 kg ha^{-1} (through urea) was observed as against significantly lower level of the pest occurrence (22.62 LDLF 10^{-1} hills) under the influence of RDF (N, P, K @ 80, 40, 20 kg ha^{-1}) in inorganic form which was further lowered down upto the statistically lower level of 15.66 LDLF 10^{-1} plants through use of reduction in use of RDF upto 50 per cent, but supplemented with GM @ 10 t ha^{-1}. The present findings suggested that nutrients (N, P, K) supplied to rice plants by following the principles of integrated nutrient management (INM) through combined use of organic and inorganic sources, could significantly be instrumental in not only suppressing the incidence of leaf folder but also in realizing higher grains yield of rice (Table 1).

Dhaliwal *et al*. (1983) revealed that increased dose of nitrogenous fertilizer could be responsible for enhancement in incidence leaf folder resulting in reduction in yield of grain yield of rice in the agro-climatic conditions of Indian Punjab state almost in accordance with findings of the present studies..

Prasad *et al*. (2004) opined that balanced quality of N, P and K in the form of chemical fertilizer applied to upland rice gave not only desirable vigour to plants but also substantial protection to the crop against major insect pests including leaf folder in the agro-climatic conditions of plateau region of Chotanagpur. Their findings also indicated that plants receiving zero (Nil) N, P, K from outside source remained too weak to be infested with almost nil incidence of major insect pests like findings of the present studies.

Findings of Prasad *et al*. (2008) indicated that organic manures *viz*. green manure, FYM, vermicompost, *neem* and *karanj* cakes treated plants received lower incidence of leaf folder of rice. Moreover, *neem* and *karanj* cakes proved to be the most effective against leaf folder (*C. medinalis*).

As such, it is concluded that *neem* and *karanj* caks applied @ 0.5, 1.0 and 2.5 t ha^{-1}, as organic manure to the rice plants could find a good place in not only in the form of nutrient supplication but also in the form of proctectant against leaf folder too .

Grains yield

The overall mean of two years experimental results (Table 1) revealed that sole use of *neem* cake @ 2.5 t ha^{-1} revealed that the highest yield of rice grains of 41.5 q ha^{-1} which remained at par with that of the sole use of *karanj* cake @ 2.5 t ha^{-1} (38.60 q ha^{-1}) followed by *neem* cake @ 1.0 t ha^{-1} plus rest of N, P, K @ 26, 40, 20 kg ha^{-1} from the chemical fertilizers (37.40 q ha^{-1}), *karanj* cake @ 1.0 t ha^{-1} plus N, P, K @

40, 20, 20 kg ha^{-1} from the inorganic sources *i.e* through fertilizers (36.60 q ha^{-1}), *neem* cake @ 0.5 t ha^{-1} + N, P, K @ 53, 40, 20 kg ha^{-1} from inorganic sources (35.90 q ha^{-1}) and 100% RDF : N, P, K @ 80, 40, 20 kg ha^{-1} (35.60 q ha^{-1}). The sole use of N @ 80 kg ha^{-1} through urea resulted to the substantially lower yield of 22.40 q ha^{-1} .

Based on the overall results of the experiment, it may be concluded that *neem* and *karanj* cake used in the varying doses of 0.5, 1.0 and 2.5 t ha^{-1} proved to be significantly effective in reducing the incidence of rice leaf folder (*C. medinalis*) resulting in the substantially higher yields of grains of rice. Hence, the present findings suggested that inclusion of *neem* and *karanj* cake as organic manure as a part of INM could be an important component of IPM for the sustainable cultivation of rice.

As such, it may be concluded that *neem* and *karanj* cakes applied either @ 0.5, 1.0 or 2.5 t ha^{-1} could be recommended as organic manure not only for supplying plant nutrients but also for protecting rice plants against the leaf folder (*C. medinalis*) for enhancing the grains yield of rice .

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Survey on insect-pests of guava and management practices around Hamelmalo sub-zoba

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ABSTRACT

Guava (*Psidium guajava*) is ever green fruit, belongs to Myrtacae family. It grows in tropical and sub-tropical area of the world and it is important source of vitamin C. The survey was conducted to identify the insect pest of guava in four villages of sub-zoba Hamelmalo *viz*. Fredarb, Genfelom, Wazntet, and Hamelmalo. The main objective of the study was to identify the major insect-pests of guava. The surveys were carried out by providing questioner to the farmers and observations on fruits. Guava fruit fly (*Bactrocera* sp.) is found to be the major insect-pest. Fruit fly infestation was observed in 89 per cent number of trees. Out of the four villages, the infestation of fruit fly was highest in Hamelmalo and Wazntet (94%) and the lowest was in Fredard (78%). Estimation of the fruit dropping due to fruit fly was made and it was found that average fruit dropping in different villages by fruit fly was maximum in Wazntet 100 per cent during summer and 22 per cent during winter season. Minimum average fruit dropping during summer season was 85 per cent in Genfelom and during winter season it was 14 per cent in Fredard.

Key words: Guava, *Bactrocera* sp., survey, management

Guava (*Psidium guajava*), the apple of the tropics, is one of the most common fruits in the tropics and sub-tropics area of the world. It is claimed to be the fourth important fruits in area and production after mango, banana and citrus. It belongs to family *Myrtaceae*. It is large shrubs or a small spreading tree with a fairly thin trunk and scaly multi-coloured bark (Cobley and Steele, 1976). Flowers are white epigenothus and develop on current growth cymes or solitary in leaf axial (Chadha and Pandey, 1986). Fruit are round to vermiform varies drastically in size. Among the tropical and subtropical fruits, guava is one of the most tolerant fruit crops to environmental stresses (Adsule and Kadam, 1995).

In Eritrea, it is grown mainly in Anseba, Gash-Barka, Northern Red Sea and Maekel. It is mainly cultivated in zoba Anseba, particularly in sub-zoba of Elabered and Hamelmalo for local market and domestic consumption. Sometimes vegetables are also grown along with the guava.

Several insect-pests cause heavy loss, such as guava fruit fly (*Bactrocera* sp.), mealy bug (*Planococcous cirri*), scale insect (*Chlorpulvinaria psidii*), thrips (*Rhipiphorothrips cruentatus*), fruit borer (*Congethespunctiferalis*), and bark eating caterpillar (*Inderbella* sp.). Hence, it is important to identify insect pests of guava and manage them. This will enable the farmers to increase their total production and per capita income. The objective of the present study are to assess the general insect pest problems of guava and management practice at sub-zoba Hamelmalo.

MATERIALS AND METHODS

Site description

Sub-zoba Hamelmalo of zoba Anseba is located about 12 Km North of Keren at an altitude of 1286 m.a.s.l. It has an annual temperature ranging from 16-38^{0}C and annual rainfall of 350–670 mm.

Sites, which were selected for survey in the sub-zoba were 4 villages, which are found along the bank of Anseba river, where guava and other fruits are commonly grown. From each village guava producing eight farmers were randomly selected with total number of trees 185, 465, 268 and 157 in village Fredarb, Hamelmalo, Genfelom, Wazntet respectively (Table 1).

Table 1. Size of the sample taken for the survey

Village	Number of farmers	Total number trees
Fredarb	8	185
Hamelmalo	8	465
Genfelom	8	268
Wazntet	8	157

Land site and the area of the orchards

The total area cultivated with guava in the surveyed orchards was 2.35 ha. Out of this Hamelmalo has the highest average area (0.13 ha) with maximum and minimum land area of orchard being 0.37 ha and 0.02 ha respectively, while Wazntet has lowest average area (0.03 ha) with maximum and minimum size of the orchards 0.12 ha 0.01 ha respectively.

Based on the information collected from the farmers, the total number of trees on the surveyed area was 1075 and the average number of trees per orchard was 35. Among the four villages Hamelmalo had the highest number of tree which is 465 with maximum and minimum being 230 and 12, respectively. The average number of trees per orchard was 58. Wazntet has the lowest total number of trees *viz.*157 with an average number of trees per orchard as 19, the maximum and minimum number of trees per orchard being 77 and 8, respectively.

Data collection and analysis

Data was collected from a sample of 32 farmers who are active in guava cultivation using a questionnaire and visual observation. Finally the collected data was organized and analysed.

RESULTS AND DISCUSSION

Chemical control

Insecticides, such as Malathion was frequently used by farmers in this sub zoba to protect the crop from insect pests for guava orchard. According to the farmers interviewed the chemicals are not easily available in the market and are expensive too. Only 16 per cent of the farmers use insecticides against guava pests. In Fredard, only 37.5 per cent of them apply chemicals, where as in Wazntet and Genfelom each 13 per cent of the farmers use chemicals pesticide. In Hamelmalo 25 per cent farmers use chemical pesticide for guava trees (Table 2).

Table 2. Percentage of farmers applying chemical

Village	Apply chemicals		Not apply chemicals	
	No. of farmers	%	No. of farmers	%
Wazntet	1	13	7	87
Fredard	3	37.5	5	62.5
Genfelom	1	13	7	87
Hamelmalo	2	25	6	75

Guava insect pest

During survey, various guava pests were found attacking guava trees which were categorised as major and minor pest (Table 3). According to the farmers in the study area, guava trees are infested and damaged by several pests in the field. Thrips, weevils, scale insects, bark eating caterpillar, aphids, mealy bug and termite were recorded as minor insect-pests in guava orchards in the sub-zoba of Hamelmalo. Guava fruit fly is found to be the major key pest in guava production (Hussain *et al.*, 2015). The infestation of this tree by the fruit fly starts under field condition, as the fruit matures and causes heavy damage (Table 3).

Fruit fly infestation

The infestation of fruit fly was severe during rainy/ summer season. This may be because of the soil moisture and the environmental temperature, which enhance the exclusion of the pupae and large number of adults emerge. Species of fruit flies are low during the dry season. In conformity with the present study Vayssieres *et al.* (2006) reported that the population increases when the rainy season starts and persist until the end of the rainy season.

The occurrence of fruit fly in this region started 4-5 years back and is increasing every year. The farmers are therefore removing their guava trees and replacing them with citrus, mango and other vegetables. Due to the infected fruits, farmers are not able to get proper income from their guava trees.

The adult and maggot of fruit fly attack the semi-ripe fruits. The female of the fruit fly punctures the fruit by its ovipositor and lay eggs inside the fruit. Maggots destroy and convert pulp into a bad smelling and discoloured semi-liquid mass. Small holes on the fruits are visible when the maggot leaves the fruit.

Fruit fly infestation was observed in 89 per cent number of trees. Out of the four villages the infestation of fruit fly was highest in Hamelmalo and Wazntet (94%) and the lowest

Table 3. Insect-pest observed in Hamelmalo area

Common name	Scientific name	Order	Family	Damaging	Nature of damage	Status
Guava fruit fly	*Bactrocera* sp.	Diptera	Tephritidae	Maggots	Bore in fruit pulp	Major or key pest
Aphids	*Aphis gossypi*	Homoptera	Aphididae	Nymph & Adult	Suck cell sap	Minor
Thrips	*Selenothrips rubrocinctus*	Thysanoptera	Thripidae	Nymph & adult	Suck cell sap	Minor
Scale insect	*Coccus viridis chloro pulvinaria psidii*	Hemiptera	Coccidae	Nymph & adult	Suck the cell sap of levels and twigs	Minor
Bark eating	*Indarbela tetraonis quadrinotata*	Lepidoptera	Meterbelidae	Caterpillar	Bore in trunk and feed on the bark	Minor
Weevil	*Myllocerus* weevil	Coleopteran	Curculionidae	Adult	Feed on leaf blade by eating blade	Minor
Striped mealy bug	*Ferresia virgate*	Hemiptera	Coccidae	Nymph & adult	Suck cell sap of tender	Minor
Termite	*Nasutitermes* spp.	Isoptera	Termitidae	Workers	Chewing & bitting	minor

was in Fredard (78%) (Fig. 1). The variation in percentage infestation was inversely proportional with the management practices and application of chemical pesticide. According to Morton (1987) infestation of fruit fly ranged from 20 to 46 per cent. In Indonesia guava fruit fly cause yield loss to guava fruit from 50-100 per cent (Broughton, 2004).

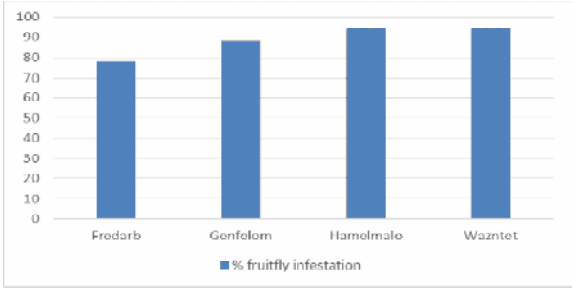

Fig. 1. Average percentage of fruit fly infested fruit

Estimation of average fruit dropping due to fruit fly in the surveyed area

Around the surveyed areas, fruit fly caused the sever dropping of matured fruits. The areas under infested trees were covered with the dropped fruits. The larva of fruit fly drops from the ripening fruits and pupate in the soil. When we cut the infected fruit, lot of white maggots were observed in the fruits.

Estimation of the fruit dropping due to fruit fly was made and it was found that average fruit dropping in different villages by fruit fly was maximum in Wazntet 100 per cent during summer and 22 per cent during winter season. Minimum average fruit dropping during summer season was 85 per cent in Genfelom and during winter season it was 14 per cent in Fredard (Fig. 2).

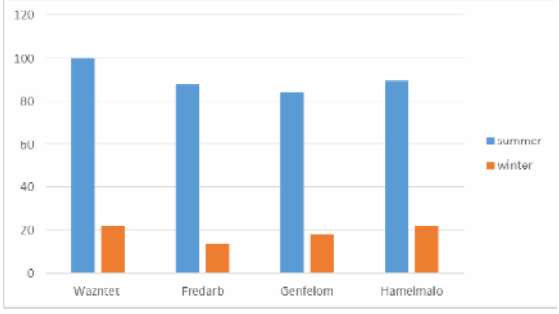

Fig. 2. Average percentage of fruit dropping in different villages in season

Fruit fly management

Farmers in the surveyed area were practicing different methods to control the fruit fly infestation, but the management practices followed by most of the farmers were not efficient in case of fruit fly control or reduction of the damage. The reasons could be the lack of knowledge about the pest, unavailability of pesticide, lack of application equipment, method of application, and the nature of the pest.

The management practice followed by the farmers included mainly the collection of the dropped and rotten fruits from the orchards and throwing them by the riverside. This could also be one of the factors that lead to higher infestation in the area. Chemical control is also practiced by few farmers, but the way of their application and lack of the sprayers increase the problem rather than solving.

Among the total farmers interviewed, 71 per cent used to leave the rotten and dropped fruits in the field and the remaining were collection and throwing them to the river side. In the surveyed villages, 87.5 per cent, 62.5 per cent farmers of Genfelom and Wazntet, respectively, did not care about the field sanitation and leave the dropped and rotten fruits in the field. Whereas, 42.8 per cent (highest) and 12.5 per cent (lowest) farmers of the Fredarb and Genfelom, respectively, collected and thrown the rotten fruits to the riverside (Table 4).

Table 4. Number of farmers and their percentage management practices on dropped fruits

Village	Leave in the field		Collect and thrown in riverside	
	No. of farmers	(%)	No. of farmers	(%)
Wazntet	5	62.5	3	37.5
Fredarb	4	57	3	42.8
Genfelom	7	87.5	1	12.5
Hamelmalo	6	75	2	25

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Severity of infestation by collar feeding insect pest (*Agrotis ipsilon*) on potato in northern districts of Kashmir

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ABSTRACT

Severity of infestation by collar feeding insect pest (*Agrotis ipsilon*) on was recorded at different locations ranging from plains to higher belts of districts Baramulla, Kupwara and Bandipora during the cropping season 2011 and 2012. Cutworm was noted as the collar feeding insect pest that cut the stem of potato crop near the base. Severity percentage of cutworm was recorded highest in May at Pattan, Kunzer (Baramulla), Yonus (Kupwara) and Sumbal (Bandipora) and highest in June at Yarikhah (Baramulla), Handwara, Budnambal (Kupwara) and Ajas (Bandipora). However, at Gurez (Bandipora), the pest severity was noted high in July during 2011 and 2012. During the year 2011, the mean severity of cutworm was recorded highest 5.00 at Sumbal (Bandipora), whereas 3.00 per cent as lowest at Yarikhah (Baramulla) and Budnambal (Kupwara). However, during 2012 the highest severity percentage of 6.25 was recorded at Pattan (Baramulla) and Sumbal (Bandipora), while lowest percentage of severity 4.00 was recorded at Budnambal (Kupwara). Pooled mean severity of cutworm was recorded highest (5.26%) at Sumbal (Bandipora) followed by Pattan (Baramulla) 5.00 per cent, while lowest percentage of severity 3.50 was recorded at Budnambal (Kupwara).

Key words : Severity, collar feeder, potato, Northern Kashmir

Potato crop is attacked and damaged by a number of insect pests including wireworms, white grub, aphids, cutworm and others. As a result, the yield of the crop is adversely affected. Cutworms are the most devastating polyphagous insect pest. Larvae of which can damage 30 cultivated and 20 wild species of plants but the greatest damage is observed on tobacco, potato, maize, beet and vegetable crop (Nicolova, 1971). It attack tobacco, potato, tomato, bottle gourd, lady's finger, cabbage, sugar beat, turnips, grams and many ornamental plants at different times of the year (Khan, 1976). The full grown larvae of the cutworm are dark or dark brown in colour with greasy body. The adult female cutworm lays their eggs on the grasses or on the weeds. The larva on emergence feeds on the epidermis of the leaves, biting the stems of seedling, eating the leaves and sometime the entire seedling and their habit changes according to their growth. Cutworm is nocturnal, as it attacks the young seedling of the plants at night. They feed on the plants by cutting their stem either below or just above the ground level. The larvae of cutworm hide and live inside the cracks and holes in the soil during the day. The per cent damage caused by it varies from 20-37 per cent, but in severe cases the damage occur as much as 80 per cent, depending on the severity of infestation (Atwal, 1976).

MATERIALS AND METHODS

Collar feeding insect pests

Severity of infestation of collar feeding insect pests was recorded at all the locations ranging from plains to higher belts of districts Baramulla, Kupwara, and Bandipora. The severity of collar feeder was determined by selecting 10 random plants from different rows during the cropping season 2011 and 2012, which were examined at fortnightly, based on partial and full cut. The per cent severity of collar feeding insect pests was calculated by the formula:

$$\text{Per cent severity} = \frac{\text{Number of plants with partial and full cut}}{\text{Total number of plants examined}} \times 100$$

RESULTS AND DISCUSSION

Cutworm (*A. ipsilon*) as a collar feeder that cuts the stem near the base was found associated with the potato crop (Plate 1). The data recorded (Table 1) for the year 2011 revealed that the severity percentage was high at Pattan and Kunzer during May whereas, at Yarikhah, it was noted to be high in June with mean severity of 3.75, 3.75 and 3.00 per cent respectively in Baramulla district. The severity percentage was noted high in May at Yonus, while in June at Handwara and Budnambal locations of Kupwara district with mean severity of 3.75, 3.75 and 3.00 per cent, respectively. However, in Bandipora district, Sumbal location indicated high severity percentage during May and June, whereas, at Ajas and Gurez locations, per cent severity was noted to be high during June and July, respectively with mean severity of 5.00, 3.75 and 3.75 per cent, respectively.

Severity of collar feeding cutworm was high in 2012 as

Larvae of cutworm

Partial cut

Full cut

Cutworm larvae

Adult cutworm

Plate 1. Collar feeding cutworm (*Agrotis ipsilon* Hufnagel) on potato crop

compared to 2011 (Table 2), where Pattan and Kunzer locations received high severity percentage during May. At Yarikhah location, peak per cent severity was observed in June with overall mean severity of 6.25, 4.57 and 5.00 per cent, respectively in Baramulla district. Almost similar trend was observed in Kupwara district, where Yonus location exhibited highest severity percentage in May, while Handwara and Budnambal locations received peak severity in June with mean severity of 5.00, 5.00 and 4.00 per cent, respectively. The pest showed high severity percentage in May and June at Sumbal and Ajas locations of Bandipora district, whereas at Gurez location, the crop received high per cent severity in July with mean severity of 6.25, 5.00 and 5.00 per cent at Sumbal, Ajas and Gurez, respectively.

Pooled mean severity of cutworm during 2011 and 2012 (Table 3 and Fig. 1) showed that Pattan location exhibited highest severity of 5.00 per cent and lowest 4.00 per cent severity at Yarikhah location of Baramulla district whereas, equal range of severity 4.37 per cent as highest was recorded at Yonus and Handwara locations and lowest 3.50 per cent severity at Budnambal location of Kupwara district. However, Bandipora, Sumbal locations received high severity 5.62 per cent. Ajas and Gurez locations recorded equal 4.37 per cent severity.

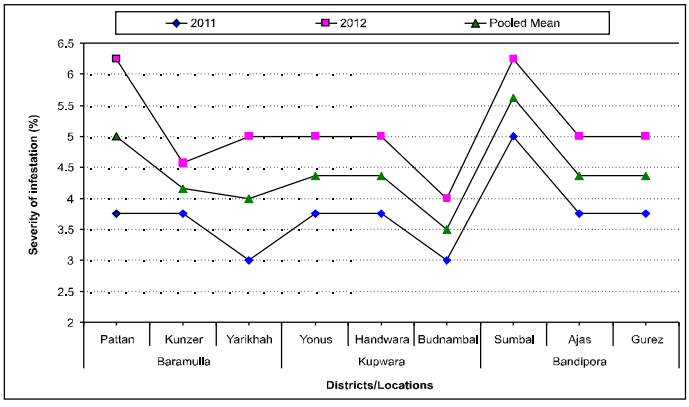

Fig. 1. Severity of infestation by collar feeding cutworm on potato crop in northern districts of Kashmir during 2011 and 2012

Table 1. Severity percentage of collar feeding insect pests of potato (*Solanum tuberosum* L.) at different locations of district Baramulla, Kupwara and Bandipora during 2011

District	Location	Monthly severity of infestation						
		April	May	June	July	August	September	Mean ±SE
Baramulla	Pattan	0.00	10.00	5.00	0.00	-	-	3.75 ±2.39
	Kunzer	0.00	10.00	5.00	0.00	-	-	3.75 ±2.39
	Yarikhah	-	0.00	10.00	5.00	0.00	0.00	3.00 ±2.00
Kupwara	Yonus	0.00	10.00	5.00	0.00	-	-	3.75 ±2.39
	Handwara	0.00	5.00	10.00	0.00	-	-	3.75 ±2.39
	Budnambal	-	0.00	10.00	5.00	0.00	0.00	3.00 ±2.00
Bandipora	Sumbal	0.00	10.00	10.00	0.00	-	-	5.00 ±2.88
	Ajas	0.00	5.00	10.00	0.00	-	-	3.75 ±2.39
	Gurez	-	-	5.00	10.00	0.00	0.00	3.75 ±2.39

*Data based on 10 random plants, Data expressed as Mean ± SE

Table 2. Severity percentage of collar feeding insect pests of potato (*Solanum tuberosum* L.) at different locations of district Baramulla, Kupwara and Bandipora during 2012

District	Location	Monthly severity of infestation						
		April	May	June	July	August	September	Mean ±SE
	Pattan	0.00	15.00	10.00	0.00	-	-	6.25 ±3.75
Baramulla	Kunzer	0.00	13.30	5.00	0.00	-	-	4.57 ±3.13
	Yarikhah	-	10.00	15.00	0.00	0.00	0.00	5.00 ±3.17
	Yonus	0.00	15.00	5.00	0.00	-	-	5.00 ±3.53
Kupwara	Handwara	0.00	5.00	15.00	0.00	-	-	5.00 ±3.53
	Budnambal	-	0.00	15.00	5.00	0.00	0.00	4.00 ±2.91
	Sumbal	0.00	10.00	15.00	0.00	-	-	6.25 ±3.75
Bandipora	Ajas	0.00	10.00	10.00	0.00	-	-	5.00 ±2.88
	Gurez	-	-	5.00	15.00	0.00	0.00	5.00 ±3.53

*Data based on 10 random plants, Data expressed as Mean ± SE

Table 3. Severity percentage of infestation by collar feeders on potato (*Solanum tuberosum* L.) at different locations of district Baramulla, Kupwara and Bandipora for 2011 and 2012

Year	Pooled mean severity of collar feeders								
	Baramulla			Kupwara			Bandipora		
	Pattan	Kunzer	Yarikhah	Yonus	Handwara	Budnambal	Sumbal	Ajas	Gurez
2011	3.75	3.75	3.00	3.75	3.75	3.00	5.00	3.75	3.75
2012	6.25	4.57	5.00	5.00	5.00	4.00	6.25	5.00	5.00
Pooled Mean ±SE	5.00±0.56	4.16±0.40	4.00±1.00	4.37±0.62	4.37±0.62	3.50±0.49	5.62±0.62	4.37±0.62	4.37±0.62

*Data based on 10 random plants, Data expressed as Mean ± SE

From the present findings, it was found that Pattan location having severity percentage of 5.00±0.56 as highest but 4.0±1.00 per cent as lowest at Yarikhah location in Baramulla district. Whereas, equal range of severity percentage 4.37±0.62 was recorded as highest at Yonus and Handwara locations and 3.50±0.49 as lowest at Budnambal location in Kupwara district. However, Bandipora, Sumbal locations received high severity percentage of 5.62±0.62 whereas, Ajas and Gurez locations had equal 4.37±0.62 per cent severity. These findings suggest that pest pressure was almost equal at all nine locations in north Kashmir, which is supported by number of workers. Das (1988), Das and Ram (1988), Singh (2002) and Khan *et al.* (2009) reported cutworm as a devastating pest that causes heavy losses in potato. Chandla and Chandel (2005) reported cutworm as major constraint in potato production in higher hills of Himachal Pradesh as it is polyphagous pests causing heavy damage and yield losses to many horticultural crops including potato.

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Control of *Spodoptera litura* (F.) by botanicals in FCV tobacco

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ABSTRACT

Acetone extracts of 34 plant species were evaluated for control of *Spodoptera litura* (F.), in FCV tobacco in Andhra Pradesh under laboratory and field conditions. The crude plant extracts of *Leucas aspera* and *Terminalia arjun* killed 95 per cent of third instar larvae of *S. litura* @ 10 µl larvae^{-1} dose after 72 hr of the treatment. Field evaluation of crude extracts of *L. aspera*, *T. arjun*, and *Tagitus erecta* var. *african* was carried out at five concentrations. The treatment, crude leaf extract of *L. aspera* (10% spray) was at par with Chlorpyriphos spray (0.05%) in controlling the larval damage to plants. It can find a place in IPM priogramme of *S. litura* and further purification/ refinement of plant extract (*L. aspera*) / mixing of crude extracts of two or three plant species are required for promoting its use in large scale field trials.

Key words: *S. litura,* Tobacco caterpillar, control, botanicals

Tobacco is grown as one of the most important commercial crops in Southern Light Soils (SLS) of Andhra Pradesh under rain fed conditions. During 2013-14, it was grown in about 58,000 ha, with a production of 55.00 million kg of cured leaf (Anonymous, 2015). Tobacco caterpillar, *Spodoptera litura* (F.) is an important pest of FCV tobacco and cause damage to both nursery and field crop. It was estimated that *S. litura* caused a yield loss of 15.0 per cent green leaf ha^{-1} annually in SLS region (Anonymous, 2002). Since, the crop is considered as one of the bread yielding crops among the farming community of this region, farmers invariably apply insecticides 4-5 rounds or more to save the crop. Hence, considerable amount of residues is left in the produce. These residues restrict the export of raw material as well as finished products of tobacco to different countries. To reduce the use of pesticides and to get good quality leaf with fewer residues, there is a need to investigate alternate sources of insect controlling substances of friendly nature to environment and other biotic agents. So, the current study was taken up to evaluate some plant extracts for control of *S. litura*.

MATERIALS AND METHODS

Fresh leaf samples (100 g each) of 34 selected plant species were collected separately and allowed to wither under shade for 12 hrs. Each withered leaf sample was cut into small pieces, filled into thimbles and plugged with cotton. The thimbles with leaf samples were subjected to the extraction process in a Soxhlet apparatus using acetone as a solvent. The process continued for 12 hrs. Later, each extract was subjected to solvent separation by using a vacuum evaporator. Crude leaf extracts thus obtained, were used in this study. Each crude extract was applied topically on the thoracic region of 3rd instar caterpillars of *S. litura* at 1, 2, 3, 5 and 10 µl larvae^{-1} concentrations using a micro pipette. There were two replications with 8 caterpillars in each replication. The caterpillars were allowed to feed on fresh tobacco leaf. The experiment was conducted under laboratory conditions at 25 ± 2°C. Mortality of caterpillar was recorded at 24 hrs interval after the treatment for three days. Earlier, the caterpillars were reared in the laboratory on cut tobacco leaf.

Crude leaf extract was obtained in large quantity form selected plants based on the results of the 1st experiment and on the basis of availability of leaf locally. Field evaluation of promising crude plant extracts was conducted at 1, 2, 3, 5 and 10 per cent spray. Detergent (1%) was added to the crude extract during the preparation of spray solution. There were 36 tobacco plants in each replication and planted at a spacing of 65 x 65 cm. Plants damaged by *S. litura* was recorded after fourth day of the treatment. The experiment was conducted for three years during 2003-2007 cropping season and the data thus recorded was analysed statistically (Gomez and Gomez, 1984).

RESULTS AND DISCUSSION

Control of *S. litura* by crude plant extracts.

Mortality of *S. litura* caused by different doses (1, 2, 3, 5 and 10 µl larvae^{-1}) of crude acetone plant extract at 24 hr after treatment is presented in table 1. All the treatments differed significantly in causing mortality of the larvae. Mortality increased with the increase in the dose of the crude extract. Some treatments did not cause mortality of larvae at lower doses but caused mortality at 3 or 5 µl dose. The mortality after 24 hr at 10 µl dose was lowest (15%) in *Annona reticulate* (seed) and *Dendrobium aphyllum*. The highest mortality (90%) of larvae was recorded in the extracts of *Leucas aspera* and *Terminalia arjun*. Further, the mortality increased

with the prolongation of exposure time indicating delayed action of some treatment *viz*., *Nishinda* sp.,*Cyda cordifolia, Acasia safeda, Calamus* sp., *Annona reticulate* (seed), *etc*. (Table 2).

Larval mortality after 72 hr of the treatment ranged between 40-95 per cent (at 10 µl larvae^{-1} dose). Highest mortality (95 %) of larvae was recorded by the crude extract of *L. aspera* and followed by *T. arjun*, *Nishinda* sp. and *Tagitus erecta* var. *african* (Table 3). Mortality of larvae increased with the increase of exposure time in some treatments. The lowest mortality (40%) was recorded in the extract of *D. aphyllum*. So, it is clear that the crude acetone extracts of *L. aspera*, *T. arjun*, *Nishinda* sp. and *T. erecta* var. *african* might contain some insecticidally active compounds that caused mortality to larvae of *S. litura*.

Several reports are available that many plant species contain potent molecules, which can cause mortality to insects (Koul, 2003). Some of them may act as anti-feedents or inhibitors of physiological processes or growth retardants *etc*. Phago-deterrent property of Neem seed extract against

Table 1. Mortality of *S. litura* caused by crude acetone plant extracts after 24hrs of treatment

Name of the plant	Per cent larval mortality after 24 hrs				
	1 µl	2 µl	3 µl	5 µl	10 µl
Leucas aspera	15 (22.5)	25 (29.9)	35 (36.2)	55 (47.9)	90 (76.7)
Metastomata malbathricum	00 (00.0)	05 (09.2)	15 (22.5)	35(36.2)	55 (47.9)
Cleodendron unfortunatum	10 (18.4)	20 (26.6)	15 (22.5)	35 (36.2)	60 (50.8)
Nictanthus sp.	10 (18.4)	15 (22.5)	35 (36.2)	45 (42.1)	75 (60.1)
Piper sp.	05 (09.2)	10 (18.4)	45 (42.1)	60(50.6)	75 (60.1)
Myconia sp.	05 (09.2)	15 (22.5)	50 (44.9)	50(44.9)	75 (60.1)
Adiantum sp.	05 (09.2)	15 (22.5)	35 (36.2)	50 (44.9)	70 (57.1)
Thivita nerifolia	10 (18.4)	10 (18.4)	20 (26.6)	20 (25.9)	35 (36.2)
Adhatoda vasica	00 (00.0)	00 (00.0)	05 (09.2)	15 (22.5)	35 (36.2)
Nishinda sp.	05 (09.2)	10 (18.4)	15 (22.5)	35 (36.2)	55 (47.9)
Cyda cordifolia	05 (09.2)	05 (09.2)	10 (18.4)	15 (22.5)	40 (39.2)
Acasia safeda	05 (09.2)	05 (09.2)	10 (18.4)	30 (33.2)	55 (47.9)
Acasia tora	05 (09.2)	05 (09.2)	15 (22.5)	30 (33.2)	60 (50.8)
Oxalis sp.	05 (09.2)	10 (18.4)	20 (26.6)	45 (42.1)	70 (57.1)
Datura stramonium	10 (18.4)	15 (22.5)	35 (36.2)	50 (44.9)	70 (57.1)
Calamus sp.	05 (09.2)	10 (18.4)	20 (26.6)	25 (29.9)	55 (47.9)
Vinca rosea var. *alba*	00 (00.0)	00 (00.0)	15 (22.5)	25 (29.9)	50 (44.9)
Vinca rosea var. *ruby*	00 (00.0)	00 (00.0)	10 (18.4)	15 (22.5)	55 (48.3)
Cleodendron irereme	05 (09.2)	05 (09.2)	15 (22.5)	25 (29.9)	50 (44.9)
Lucenae lucocephala	00 (00.0)	00 (00.0)	10 (18.4)	25 (29.9)	55 (47.9)
Anona eticulate (leaf)	00 (00.0)	00 (00.0)	00 (00.0)	10 (18.4)	35 (36.2)
Tagitus erecta var. *african*	00 (00.0)	00 (00.0)	05 (09.2)	25 (29.9)	55 (47.9)
Terminalia arjun	10 (18.4)	15 (22.5)	35 (36.2)	55 (47.9)	90 (76.7)
Ocimum sp.	05 (09.2)	05 (09.2)	05 (09.2)	15 (22.5)	40 (39.2)
Tagitus patula var. *french*	00 (00.0)	00 (00.0)	10 (18.4)	25 (29.9)	55 (47.9)
Acacia auriculiformis	00 (00.0)	00 (00.0)	15 (22.5)	45 (42.1)	70 (57.1)
Annona reticulate (seed)	00 (00.0)	00 (00.0)	00 (00.0)	10 (18.4)	15 (22.5)
Juniperus sp.	00 (00.0)	00 (00.0)	25 (29.9)	55 (47.9)	75 (60.1)
Juniperus sp. (thorny)	00 (00.0)	00 (00.0)	15 (22.5)	25 (29.9)	55 (47.9)
Lantena camera	00 (00.0)	00 (00.0)	10 (18.4)	20 (26.6)	50 (44.9)
Dendrobium aphyllum	00 (00.0)	00 (00.0)	00 (00.0)	00 (00.0)	15 (22.5)
Cestrum nocturnum	00 (00.0)	00 (00.0)	00 (00.0)	00 (00.0)	25 (29.9)
Cuscuda sp.	00 (00.0)	00 (00.0)	10 (18.4)	25 (29.9)	55 (47.9)
Cajanus cajan (seed coat)	00 (00.0)	05 (09.2)	05 (09.2)	30 (33.2)	55 (47.9)
Control (water)	00 (00.0)	00 (00.0)	00 (00.0)	00 (00.0)	00 (00.0)
SEm.	4.4	4.0	3.3	2.1	5.9
CD 5%	12.1	11.1	9.2	5.7	16.3
CV	96.8	59.2	22.1	9.2	16.7

* Figures in the parentheses indicate angular transformed values

Table 2. Mortality of *S. litura* caused by crude acetone plant extracts after 48hrs of treatment

Name of the plant	Per cent larval mortality with the treatment after 48 hr.				
	1 µl	2 µl	3 µl	5 µl	10 µl
Leucas aspera	15 (22.5)	25 (29.9)	40 (39.2)	65 (53.8)	95 (80.8)
Metastomata malbathricum	00 (00.0)	10 (18.4)	20 (26.6)	40 (39.2)	55 (44.9)
Cleodendron unfortunatum	10 (18.4)	15 (22.5)	20 (26.6)	40 (39.2)	75 (60.1)
Nictanthus sp.	15 (22.5)	15 (22.5)	40 (39.2)	50 (44.9)	85 (73.4)
Piper sp.	05 (09.2)	15 (22.5)	45 (42.1)	65 (53.8)	85 (73.4)
Myconia sp.	05 (09.2)	20 (26.6)	50 (44.9)	50 (44.9)	85 (73.4)
Adiantum sp.	10 (18.4)	20 (26.6)	40 (39.2)	55 (47.9)	75 (60.1)
Thivita nerifolia	10 (18.4)	15 (22.5)	25 (29.9)	25 (29.9)	50 (44.9)
Adhatoda vasica	00 (00.0)	00 (00.0)	10 (18.4)	20 (26.6)	55 (47.9)
Nishinda sp.	05 (09.2)	10 (18.4)	20 (26.6)	40 (39.2)	85 (73.4)
Cyda cordifolia	05 (09.2)	05 (09.2)	15 (22.5)	20 (26.6)	70 (57.1)
Acasia safeda	05 (09.2)	10 (18.4)	10 (18.4)	30 (33.2)	75 (60.1)
Acasia tora	05 (09.2)	05 (09.2)	20 (26.6)	30 (33.2)	70 (57.1)
Oxalis sp.	05 (09.2)	10 (18.4)	20 (26.6)	50 (44.9)	75 (60.1)
Datura stramonium	10 (18.4)	20 (26.6)	40 (39.2)	50 (44.9)	70 (57.1)
Calamus sp.	05 (09.2)	10 (18.4)	25 (29.9)	30 (33.2)	85 (73.4)
Vinca rosea var. *alba*	00 (00.0)	00 (00.0)	20 (26.6)	25 (29.9)	55 (47.9)
Vinca rosea var. *ruby*	00 (00.0)	00 (00.0)	15 (22.5)	20 (26.6)	70 (57.1)
Cleodendron irereme	10 (18.4)	10 (18.4)	15 (22.5)	30 (33.2)	55 (47.9)
Lucenae lucocephala	00 (00.0)	00 (00.0)	15 (22.5)	25 (29.9)	55 (47.9)
Anona eticulate (leaf)	00 (00.0)	00 (00.0)	00 (00.0)	15 (22.5)	50 (44.9)
Tagitus erecta var. *african*	00 (00.0)	00 (00.0)	10 (18.4)	30 (33.2)	85 (73.4)
Terminalia arjun	10 (18.4)	20 (26.6)	40 (39.2)	55 (47.9)	90 (76.7)
Ocimum sp.	05 (09.2)	10 (18.4)	10 (18.4)	20 (26.6)	50 (44.9)
Tagitus erecta var. *french*	00 (00.0)	00 (00.0)	15 (22.5)	25 (29.9)	70 (57.1)
Acacia auriculiformis	00 (00.0)	00 (00.0)	20 (26.6)	50 (44.9)	75 (60.1)
Annona reticulate (seed)	00 (00.0)	00 (00.0)	00 (00.0)	15 (22.5)	40 (39.2)
Juniperus sp.	00 (00.0)	00 (00.0)	25 (29.9)	60 (50.8)	85 (73.4)
Juniperus sp. (thorny)	00 (00.0)	00 (00.0)	15 (22.5)	25 (29.9)	80 (70.4)
Lantena camera	00 (00.0)	00 (00.0)	15 (22.5)	20 (26.6)	70 (57.1)
Dendrobium aphyllum	00 (00.0)	00 (00.0)	00 (00.0)	05 (09.2	35 (36.2)
Cestrum nocturnum	00 (00.0)	00 (00.0)	00 (00.0)	05 (09.2)	55 (47.9)
Cuscuda sp.	00 (00.0)	00 (00.0)	10 (18.5)	25 (29.9)	70 (57.1)
Cajanus cajan (seed coat)	00 (00.0)	05 (09.2)	05 (09.2)	30 (33.2)	70 (57.1)
Control (water)	00 (00.0)	00 (00.0)	00 (00.0)	00 (00.0)	00 (00.0)
SEm.	3.9	3.7	2.8	3.0	4. 5
CD 5%	10.8	10.3	7.7	8.4	12.4
CV	73.2	48.6	16.4	12.4	10.6

* Figures in the parentheses indicate angular transformed values.

coffee mealy bug was reported by Jacobson *et al.* (1978), growth inhibitory effect of NSKE on *S. litura* and *P. xylostella* was reported by Sombastri and Tignattannont (1984). The methanol extract of seeds of *Cleome viscose* totally deterred the egg laying in *S. litura* (Anonymous, 2004). Castor leaves treated with leaf extracts (2% dimethy sulfoxide) of *Calotropis* sp. caused 90 per cent mortality in *S. litura* larvae (Anonymous, 2007). Reports indicate that seed husk of pigeon pea contain phenolics and other chemicals that caused mortality to *S. litura* (Bhattacharya and Chenchaiah, 2007) and *C. ganges* (Chenchaiah and Bhattacharya, 2000) in semi synthetic diets. Similarly, Singh and Singh (2008) reported that crude leaf extracts of *Trichilia connaroides* caused mortality to *S. litura*, while crude ethanol extracts of *Acacia arabica*, *Annona squamosa* and *Datura stramonium* gave higher control of *S. litura* than the corresponding aqueous extrats (Rajguru *et al.*, 2011). In the current study, the crude acetone extracts of *L.* aspera, *T. arjun*, *Nishinda* sp. and *T. erecta* var. *african* might contain some insecticidal active compounds that caused mortality to larvae of *S. litura*.

Table 3. Mortality of *S. litura* caused by crude acetone plant extracts after 72hrs of treatment.

Name of the plant	Per cent larval mortality with the treatment after 72 hr.				
	1 µl	2 µl	3 µl	5 µl	10 µl
Leucas aspera	20 (26.6)	30 (33.2)	50 (44.9)	70 (57.1)	95 (80.8)
Metastomata malbathricum	00 (00.0)	10 (18.4)	20 (26.6)	40 (39.2)	70 (57.1)
Cleodendron unfortunatum	10 (18.4)	20 (26.6)	20 (26.6)	40 (39.2)	85 (73.4)
Nictanthus sp.	15 (22.5)	20 (26.6)	45 (42.1)	55 (47.9)	85 (73.4)
Piper sp.	05 (09.2)	20 (26.6)	50 (44.9)	70 (57.1)	85 (73.4)
Myconia sp.	10 (18.4)	25 (29.9)	50 (44.9)	55 (47.9)	85 (73.4)
Adiantum sp.	10 (18.4)	20 (26.6)	40 (39.2)	60 (50.8)	85 (73.4)
Thivita nerifolia	10 (18.4)	15 (22.5)	25 (29.9)	30 (33.2)	60 (50.8)
Adhatoda vasica	00 (00.0)	10 (18.4)	10 (18.4)	20 (26.6)	60 (50.8)
Nishinda sp.	05 (09.2)	15 (22.5)	25 (29.9)	45 (42.1)	90 (76.7)
Cyda cordifolia	05 (09.2)	05 (09.2)	15 (22.5)	20 (26.6)	85 (73.4)
Acasia safeda	10 (18.4)	10 (18.4)	15 (22.5)	35 (36.2)	85 (73.4)
Acasia tora	05 (09.2)	05 (09.2)	25 (29.9)	35 (36.2)	85 (73.4)
Oxalis sp.	05 (09.2)	15 (22.5)	30 (33.2)	50 (44.9)	85 (73.4)
Datura stramonium	10 (18.4)	25 (29.9)	40 (42.1)	55 (47.9)	70 (57.1)
Calamus sp.	05 (09.2)	10 (18.4)	25 (29.9)	35 (36.2)	85 (73.4)
Vinca rosea var. *alba*	00 (00.0)	00 (00.0)	20 (26.6)	30 (33.2)	70 (57.1)
Vinca rosea var. *ruby*	00 (00.0)	00 (00.0)	20 (26.6)	25 (29.9)	80 (70.4)
Cleodendron irereme	10 (18.4)	10 (18.4)	20 (26.6)	30 (33.2)	70 (57.1)
Lucenae lucocephala	00 (00.0)	00 (00.0)	20 (26.6)	25 (29.9)	60 (50.8)
Anona eticulate (leaf)	00 (00.0)	00 (00.0)	05 (09.2)	15 (22.5)	60 (50.8)
Tagitus erecta var. *african*	00 (00.0)	00 (00.0)	20 (26.6)	35 (36.2)	90 (76.7)
Terminalia arjun	15 (22.5)	25 (29.9)	45 (42.1)	60 (50.8)	95 (80.8)
Ocimum sp.	10 (18.4)	10 (18.4)	15 (22.5)	25 (29.9)	65 (54.2)
Tagitus erecta var. *french*	00 (00.0)	00 (00.0)	20 (26.6)	25 (29.9)	85 (73.4)
Acacia auriculiformis	00 (00.0)	00 (00.0)	30 (33.2)	55 (47.9)	85 (73.4)
Annona reticulate (seed)	00 (00.0)	00 (00.0)	10 (18.4)	20 (26.6)	50 (44.9)
Juniperus sp.	00 (00.0)	05 (09.2)	35 (36.2)	65 (53.8)	85 (73.4)
Juniperus sp. (thorny)	00 (00.0)	00 (00.0)	20 (26.6)	40 (39.2)	85 (73.4)
Lantena camera	00 (00.0)	00 (00.0)	15 (22.5)	20 (26.6)	80 (70.4)
Dendrobium aphyllum	00 (00.0)	00 (00.0)	00 (00.0)	05 (09.2)	40 (39.2)
Cestrum nocturnum	00 (00.0)	00 (00.0)	00 (00.0)	05 (09.2)	60 (51.3)
Cuscuda sp.	00 (00.0)	00 (00.0)	15 (22.5)	25 (29.9)	70 (57.1)
Cajanus cajan (seed coat)	00 (00.0)	05 (09.2)	10 (18.4)	35 (36.2)	70 (57.1)
Control (water)	00 (00.0)	00 (00.0)	00 (00.0)	00 (00.0)	00 (00.0)
SEm.	3.8	3.6	2.7	3.2	5.4
CD 5%	10.5	9.9	7. 5	8. 8	14.7
CV	66.7	38.6	13.8	12.3	10.8

* Figures in the parentheses indicate angular transformed values.

Field evaluation of promising crude plant extracts for control of *S. litura.*

From the above results, three plant species (*L. aspera, T. arjun, T. erecta* var. *african*) were selected for field evaluation. The results of field evaluation trial indicated that all the treatments differed significantly in controlling the insect attack to plants (Table 4). Plants damaged by the larvae were the lowest (8.17%) in Chlorpyriphos spray and followed by the crude plant extract of *Leucas aspera* (13.67%). No significant differences were found among these two treatments. The damage caused by larvae in the treatment, crude extract of *L. aspera* (10% spray) was similar to that of chlorpyriphos spray (0.05%). Damage to plants decreased with the increase in the concentration of crude extract.

From the above, it is evident that *Leucas aspera* contain some compounds which are equally potent as Chlorpyriphos. It can find a place in the IPM programme of *S. litura* as the use of botanicals reduces the ill effects of chemical pesticides. Further, the purification/refinement of plant extract (*L. aspera*)/mixing of crude extracts of two or three plant

Table 4. Plants damaged by *S. litura* as effected by the sprays of plant extracts.

Plant extract/Spray Concentration	1%	2%	3%	5%	10%	Mean *
Leucas aspera	18.07 (4.25)	14.90 (3.86)	14.23 (3.77)	11.80 (3.43)	9.37 (3.06)	13.67 (3.67)
Terminalia arjun	23.50 (4.85)	21.63 (4.65)	19.77 (4.44)	17.13 (4.14)	15.50 (3.92)	19.51 (4.40)
Tajetus erecta var. *african*	22.33 (4.73)	21.50 (4.64)	18.80 (4.33)	17.27 (4.15)	14.90 (3.85)	18.96 (4.34)
Control (Chlorpyriphos spray 0.05%)	8.17 (2.85)	8.17 (2.85)	8.17 (2.85)	8.17 (2.85)	8.17 (2.85)	8.17 (2.85)
Mean	18.02 (4.17)	16.55 (4.00)	15.24 (3.85)	13.59 (3.64)	11.98 (3.42)	
	SEm	CD @5%	CV %			
Treatments	0.08	0.27	8.41			
Concentrations	0.03	0.09	6.11			
Interaction	0.06	0.21	9.30			

* Figures in the parentheses indicate angular transformed values.

species are required for promoting its use in large scale field trials.

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Eco-friendly methods for management of downy mildew of grapevines

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ABSTRACT

Leaf disc sensitivity assay showed that EC_{50} value of *P. viticola* isolates against chitosan was 64.70 ± 12.76 µg l^{-1} and in field trial foliar applications of chitosan significantly provided 57 per cent control of downy mildew on grape leaves and 70 per cent control on bunch. Three integrated disease management schedules were prepared and implemented in field by substituting 6 fungicide applications with soil application of ISR strains of *Trichoderma* and *Bacillus* or foliar application of chitosan or by reduction of 7 fungicide applications by use of activated potassium salt of long chain phosphorus (APSP) with multisite low risk fungicides. The field trial showed that all the schedules were equally effective as farmers' practice for disease control even with 6 -7 less fungicide applications. In schedule 2 *Bacillus* DR-92 recorded more number of bunches and yield per vine, while in schedule 3 with chitosan had better shelf-life. This study brings out the possibility of integrating soil application of microorganisms, with foliar applications of APSP and chitosan along with need based application of low risk fungicides indicates potential for management of fungicide resistance and fungicide residues in vineyards.

Key words: *Plasmopara viticola*, downy mildew, grapes, chitosan, induced systemic resistance, integrated management

Downy mildew is one of the most destructive diseases of grapevines in grape-growing regions of India. The disease occurs on flower buds and young shoots, leaves and berries during wet and warm weather (Sawant *et al*., 2012).During fruiting season, crop losses can range from 30 per cent to cent per cent, if rainfall occurs during early growth to fruit set stages. Infections on clusters can also occur in absence of rain if dew is formed at night (Sawant *et al*., 2010). Spot infections on rachis or pedicles of young bunch leads to collapse of berries ahead of the infected area at later stages of development. Hence, the early shoot growth to fruit set stages is a high risk period for yield losses.

Most of the commercial table grape cultivars are highly susceptible to downy mildew and require frequent fungicide applications. A number of fungicides are registered in India for control of downy mildew but due to food safety considerations, there are limited fungicides choices available to growers for protection against the disease (Gessler *et al*., 2011). Furthermore, many of them belong to high risk group and the pathogen develops resistance to them leading to lack of required level of disease control (Sawant *et al*., 2016a and 2017). Use of low or nil risk chemicals and eco-friendly products have shown success in management of disease (Sawant *et al*., 2016b, 2016c). Chitosan, which is a naturally occurring linear polysaccharide composed of randomly distributed β-(1→4)-linked D-glucosamine (deacetylated unit) and N-acetyl-D-glucosamine (acetylated unit) has shown potential in control of plant diseases by inhibiting fungal growth and development and can also elicit defense responses in plants against various pathogens (Hadrami *et al*., 2010).

Similarly, use of microorganisms for induction of systemic resistance against plant pathogens in crop plants is well known. The induced systemic resistance (ISR) gives the plant an enhanced defensive capacity, which results in lesser disease severity (Van Loon *et al*., 1998; Van Loon and Bakker, 2005). Upon challenge by pathogen ISR results in alteration of host plant physiology and metabolic responses, leading to an enhanced synthesis of plant defense related bio-chemicals and enzymes (Naik and Singh, 2017; Ramamoorthy *et al*., 2001).

Trichoderma harzianum strain T39 is a commercial biocontrol agent for control of various pathogens on horticultural and field crops in greenhouse or open fields and also induce systemic resistance in many crops against gray mold caused by *Botrytis cinerea* (Elad, 2000; De Meyer *et al*., 1998). It also induce systemic resistance in grape vines against downy mildew caused by *Plasmopara viticola* (Perazzolli *et al*., 2008).

The increasing failures of high risk fungicides in controlling downy mildew in commercial vineyards in

Maharashtra due to fungicide resistant population and the number of fungicide residues being detected being observed was of concern to viticulture in India. To address these problems an attempt was made to evaluate micro-organisms and safer chemicals to reduce number of fungicide applications or use them along with multisite low risk fungicides.

MATERIALS AND METHODS

Antagonistic activity of chitosan against *P. viticola*

Four *P. viticola* isolates were selected for the study on the antagonistic activity against chitosan. Sensitivity to these isolates were earlier studied against azoxystrobin, dimethomorph and activated potassium salt of long chain phosphorous (Sawant *et al*., 2016a, 2016b and 2017).

Chitosan solution at 70% degree of de-acetylation (DD) was prepared by dissolving in 0.25 N HCl under continuous stirring at 50°C. Insoluble material was removed by centrifugation at 5000 rpm for 10 min and the chitosan in the supernatant was precipitated with 1N NaOH. It was then washed thrice with deionized water and then air dried. The stock solution of 10000 µg ml^{-1} was prepared by dissolving purified chitosan in 0.25 N HCl by stirring and the pH was adjusted to 5.6 with 1N NaOH. The solution was autoclaved and working concentration was obtained by appropriately diluting the stock solution with distilled water. Sensitivity test was done by leaf disk bioassay in 24 well plates. 15 mm discs were cut from leaf of 6^{th} position from the apex of healthy growing shoots of Thomson Seedless cultivar. The leaf discs were dipped for 1 minute in aqueous solution of chitosan concentrations 0, 1, 10, 100, 1000 and 10,000 µg ml^{-1}. Treated discs were placed upside down in a well containing 1 ml of solidified 0.5 per cent water agar. Leaf disks were inoculated with 10 µl sporangial suspensions containing 50,000 sporangia ml^{-1} at the centre of the disc. Plates were incubated at 22°C with alternating periods of 12 h light and dark. After six to eight days, the lesion diameter was measured and the per cent infected leaf area was determined, considering lesion in control as 100 per cent infected area. EC_{50} value was calculated by plotting the $\log_{10}$ fungicide concentration against the per cent infected leaf area.

Location and grape cultivar: The field trials were conducted at Dhondgavanwadi, Nasik, Maharashtra during fruiting growth phase 2014-15. The trial was conducted on 5 year old grape cultivar Tas-A-Ganesh planted at 10' x 6' spacing and trained to extended Y trellis. The vineyard was pruned on 13^{th} October 2014.

Control of downy mildew using chitosan: Ten applications of chitosan 70% degree of de-acetylation were made from 10.11.2014 to 17.12.2014. Applications were initiated before appearance of the disease. Thus, there were three preventive and seven curative applications. Unsprayed vines were maintained as control. Treatments were applied on 24 vines and disease observations were recorded on 40 shoots from the central 4 vines. Surrounding vines were maintained as guard vines. On each shoot 10 leaves and two bunches were taken for observations. Disease observations were recorded adopting 0 – 4 disease rating scale, where 0 = nil, 1 = up to 25%, 2 = 26 to 50%, 3 = 51 to 75% and 4 = more than 75% leaf area infected (Horsfall and Barratt, 1986). Per cent Disease Index (PDI) was calculated by following formula of McKinney (1923). Yield was recorded at harvest. The data was analysed by 't' test using SAS software.

$$PDI = \frac{\text{Sum of numerical ratings} \times 100}{\text{Number of leaves observed} \times \text{Maximum of rating scale}}$$

Integrated management using biological and safer chemicals

Microbial inoculum preparation: The *Trichoderma* isolates were grown on 200 ml potato dextrose broth (PDB) in Roux bottles seeded with 2 discs of 10 mm cut from 3-days old culture growing on PDA. Bottles were incubated on laboratory bench for 15 days under natural daylight conditions at 28 ± 4°C. The fungal growth was harvested, homogenized in a blender and filtered through double layer of muslin cloth. The suspension was diluted by adding required quantity of sterile distilled water, containing two to three drops of 0.05 per cent Tween 80 to provide a spore count of 5×10^6 conidia ml^{-1}. *Bacillus* isolates were cultured in nutrient broth (NB) in 250 ml conical flasks for 72 hr on an orbital shaker at 28°C and 150 rpm. The cells were harvested by centrifugation at 5000 rpm for 5 min, washed twice with sterile distilled water and then re-suspended in sterile distilled water to give a final count of about 1×10^8 CFU ml^{-1} as adjusted by absorbance of 0.08 to 0.1 at 600 nm.

Schedules: For integrated management trials, four schedules were prepared and implemented in the field with reduced fungicide applications as compared to general farmer practice (Table 1).

Schedule-1 (Farmers practice): Thirteen fungicide applications as practiced by the farmer. The fungicides and their doses (per L) used were dimethomorph 50 WP @ 0.5 g, mancozeb 75 WP @ 2 g, copper hydroxide 53.8 DF @1.5 g, (cymoxanil + mancozeb 8 + 64 WP) @ 2 g, (famoxadone 16.6% + cymoxanil 22.1 % SC) @ 0.5 ml, fosetyl Al 80 WP @ 2 g, (iprovalicarb + propineb 5.5 + 61.25WP) @ 2.25 g activated

Table 1. Details of treatment applications under different schedules

Date	Schedule 1 Farmer's practice	Schedule 2 ISR integrated with fungicides	Schedule 3 Chitosan integrated with fungicides	Schedule 4 APSP 96 % + mancozeb/copper hydroxide
07.11.2014	Dimethomorph	Dimethomorph & *Trichoderma/Bacillus*	Dimethomorph	--
09.11.2014	--	--	--	Mancozeb
12.11.2014	Copper hydroxide	--	Dimethomorph	--
14.11.2014	Cymoxanil + Mancozeb	Cymoxanil + Mancozeb	Cymoxanil + Mancozeb	--
15.11.2014	APSP + Mancozeb	--	--	APSP + Mancozeb/Copper hydroxide
16.11.2014	Famoxadone + Cymoxanil	--	--	--
17.11.2014	Fosetyl Al	Fosetyl Al	Fosetyl Al	Mancozeb/Copper hydroxide
18.11.2014	Dimethomorph	--	--	--
20.11.2014	--	--	--	APSP
22.11.2014	Iprovalicarb + Propineb	Iprovalicarb + Propineb	Iprovalicarb + Propineb	APSP
25.11.2014	Dimethomorph	Dimethomorph	Dimethomorph	APSP
26.11.2014	--	*Trichoderma/Bacillus*	--	Mancozeb/Copper hydroxide
30.11.2014	Cymoxanil + Mancozeb	--	--	--
03.12.2014	Dimethomorph	Dimethomorph	Dimethomorph	APSP
04.12.2014	--	*Trichoderma / Bacillus*	--	--
07.12.2014	Iprovalicarb + Propineb	--	--	--
13.12.2014	--	--	--	APSP + Mancozeb/Copper hydroxide
15.12.2014	--	--	Chitosan	APSP + Mancozeb/Copper hydroxide
19.12.2014	Dimethomorph	Dimethomorph	Chitosan	APSP
Total applications	13 fungicide	7 fungicide + 3 *Trichoderma/ Bacillus*	7 fungicide + 2 chitosan	6 mancozeb/COH + 8 APSP

potassium salt of long chain phosphorous (APSP) 96% @2g.

Schedule-2 (ISR): Four micro-organisms with potential to induce systemic resistance in grapevines against downy mildew from the culture collection of this centre were included under this schedule. These were *Trichoderma asperelloides* strain 5R, *T. asperelloides* (NAIMCC-F- 01812), *Bacillus* TS-45 and *Bacillus* DR-92 applied separately as four treatments. Three applications of one litre inoculum was applied as drench in the drip circle. The number of fungicide applications was reduced from 13 to 7.

Schedule-T3 (Chitosan): Fungicide applications were made at high risk period of downy mildew disease based on disease advisory data. Two applications of chitosan 2 g l^{-1} were made as foliar spray after fruitset and numbers of fungicide applications were reduced from 13 to 7.

Schedule-T4 (APSP): Application of activated potassium salt of long chain phosphorous (APSP) alone or in tank mix with as per disease risk based on disease advisory data. The high risk fungicides were replaced with APSP and only non-systemic multisite fungicides, mancozeb and copper hydroxide were used under this schedule. Eight APSP applications and 6 mancozeb or applications copper hydroxide were made. Thus, there were two treatments under this schedule.

In all schedules treatment applications were not necessarily made on the same day. Observations were recorded as given above. Ten replications per treatment were maintained. The data was analysed by RBD using SAS software. The PDI data was transformed using arcsine transformation. Results significant at $P = 0.05\%$ are discussed.

RESULTS AND DISCUSSION

Antagonistic effect of chitosan on *P. viticola*: The *in vitro* sensitivity (EC_{50}) of the four QoI and CAA fungicide resistant *P. viticola* isolates to chitosan was found to be 64.70 ± 12.76 µg ml^{-1}. These isolates had recorded EC_{50} of more than 110 µg ml^{-1} against kresoxim methyl (QoI group) and 54.28 to >100 µg ml^{-1} for dimethomorph (CAA group) and 539.9 to 643.5 µg ml^{-1} against activated potassium salt of long chain phosphorous (PSAP) (Sawant *et al*., 2016a, 2016b and 2017).

Bio-efficacy of chitosan against downy mildew: In the field trial, foliar application of chitosan significantly reduced PDI of downy mildew on both leaves and bunch (Fig. 1). The PDI in control increased to about 60 in leaves and bunch but in chitosan treated vines it was restricted to 26.56 on leaves

Table 2. Induction of systemic resistance in grapevines against downy mildew

Schedule No	Treatment	PDI on leaves	Bunch vine^{-1} (No)	Yield vine^{-1} (kg)	Berry diam. (mm)	TSS ^{0}B	PLW (%)
S1	Control (Farmer's practice)	4.5 (10.5)^{ab}	22.7^{e}	7.98^{d}	17.0	17.0	4.88 (12.76)^{abc}
S2	*Bacillus* TS-45	3.50 (10.0)^{a}	31.9^{abc}	12.00^{ab}	18.9	17.5	5.06 (12.99)^{bc}
S2	*Bacillus* DR-92	3.00 (9.3)^{a}	30.4^{b}	13.23^{a}	18.1	17.0	5.12 (13.09)^{bc}
S2	*T. asperelloides* (NAIMCC-F- 01812)	3.50 (10.1)^{ab}	37.9^{a}	10.26^{bc}	19.3	17.0	4.59 (12.36)^{bc}
S2	*T. asperelloides* strain 5R	3.25 (10.3)^{ab}	32.9^{abc}	10.00^{bc}	18.4	16.5	5.29 (13.30)^{c}
S3	Chitosan	4.25 (11.1)^{ab}	37.2^{a}	12.49^{a}	17.4	18.0	4.28 (11.92)^{a}
S4	APSP + Mancozeb	4.75 (12.2)^{ab}	26.3^{cde}	9.77^{cd}	17.4	19.3	4.56 (12.33)^{ab}
S4	APSP + Copper hydroxide	7.25 (15.5)^{b}	24.9^{de}	8.52^{cd}	18.6	18.0	4.59 (12.38)^{abc}
CD (p=0.05)		5.4	6.95	2.00	NS	NS	0.9

In the columns values followed by the same letters are not significant at *P=0.05* by Tukey's Studentized Range (HSD) Test.

and 18.14 on bunch. This shows that chitosan provides 57 per cent control on leaves and 70 per cent control on bunch. Chitosan is an efficient promoter of plant defense reactions and triggers an accumulation of phytoalexins, *trans*- and *cis*-resveratrol and their derivatives å-viniferin and piceid and also markedly induces chitinase and â-1, 3-glucanase activities in grapevine leaves which significantly reduce downy mildew disease (Aziz *et al*., 2006).

Integrated management: In the 'Farmers' practice'the disease PDI was 10.5 (Table 2). All treatments with reduced number of fungicide applications gave equal control of disease as farmers practice. Among the treatments PDI was lower in vines treated with the two *Bacillus* isolates TS-45 and DR-92 as compared to PDI in vines treated with + copper hydroxide. The number of bunches was highest in vines treated with chitosan and *T. asperelloides* (NAIMCC-F- 01812) which was higher than the number in vines treated with APSP + copper hydroxide or mancozeb and farmers practice. This may be due to less cluster necrosis in these treatments due to induction of systemic resistance. The number of bunch in vines treated with APSP along with mancozeb or copper hydroxide was equal to that in farmers' practice, while other treatments had more number of bunches as compared to farmers practice. Almost similar pattern was seen in yield per vine. The berry diameter and total soluble solids (TSS) were non-significant. Shelf-life studies indicated that the physiological loss in weight (PLW) in all treatments was equal to that in farmers practice. Among the different treatments PLW was lower in treatment with chitosan spray as compared to the ISR treatments showing that pre-harvest application of chitosan improve the shelf life of grape as has been shown in earlier studies also (Romanazzi *et al*., 2002). In an earlier studies it was reported that pre-harvest foliar applications with *T. asperelloides* strain 5R (earlier reported as *T. harzianum*) enhance shelf life of grapes, however, the same effect was not seen with soil application (Sawant and Sawant, 2010).

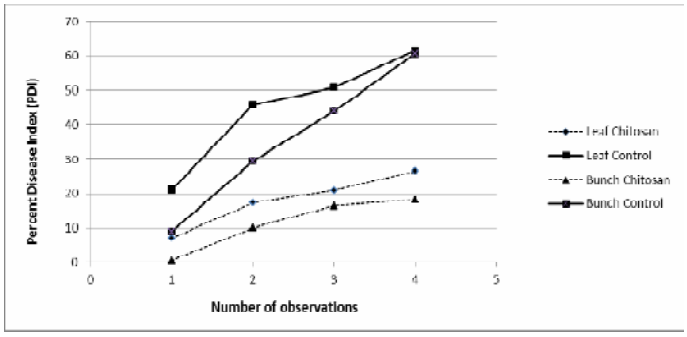

Fig. 1. Efficacy of chitosan in minimizing PDI of downy mildew on grape leaves and bunch. 1, 2, 3, & 4 observations were recorded on 24-Nov., 2-Dec., 13-Dec. and 19-Dec. 2014.

The study shows that soil application of selected efficient microorganisms can induce systemic resistance in grapevines and get better disease control than by use of fungicides alone. Soil application of microorganisms also resulted in savings of 6 fungicide applications. On the other hand, foliar application of chitosan was equally effective in yield and bettershelf-life of grapes. Foliar application of APSP in combination with non-systemic fungicides was also equally effective. Integrating soil application of microorganisms, with foliar applications of APSP and chitosan along with need based application of low risk fungicides indicates potential for management of fungicide resistance and fungicide residues in vineyards.

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Eco-friendly management of root rot of guar caused by *Fusarium solani* and *Rhizoctonia solani*

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ABSTRACT

Local isolates of 18 fungal and 19 bacterial biocontrol agents and five neem based formulations were evaluated at first *in vitro*, and then in green house to find the most effective ones for suppression guar root rot complex caused by *Fusarium solani* and *Rhizoctonia solani*. Maximum inhibition of *F. solani* and complete inhibition of *R. solani* was recorded in *T. viride* (S) and *T. harzianum* Gn1. The 17 *Bacillus subtilis* isolates and two *Streptomyces* isolates were highly effective against *F. solani* and *R. solani* in dual culture studies. Kernel extract and oil based neem formulations (0.2%) caused maximum growth inhibition of both *F. solani* and *R. solani*. Among the fungicides, Carbendazim, Mancozeb and Captan were effective. The biocontrol agents, neem formulations and fungicides found promising *in vitro* were then evaluated as seed treatments in greenhouse experiment. The highest seed germination and dry biomass and lowest disease incidence was recorded in *T. harzianum* Gn1 followed by *B. subtilis* isolate Ch-Kp-b-1. Among the neem based formulations tested, neem oil as seed treatment + soil drench resulted in good seed germination, reduced disease incidence and higher dry biomass. Among the fungicides Captan was found most effective in suppressing root rot of guar followed by carbendazim. The BCAs could establish and multiply well in the guar rhizosphere and significantly lowered the density of the both pathogens. The population density of *F. solani* and *R. solani* was lowest in *T. harzianum* Gn1and *B. subtilis* isolate Ch-Kp-b-1.

Key words: Guar, root-rot, *Fusarium solani*, *Rhizoctonia solani*, integrated management

Guar [*Cyamopsis tetragonoloba* (Linn.) Taub.] is an economically important drought tolerant annual leguminous crop in India where three forth of the global guar cultivation is carried out. Rajasthan is largest guar producing state in India where area under guar cultivation was 5.6 lakh hectares (Anonymous, 2003). Root- rot caused by *Fusarium solani* and *Rhizoctonia solani* is a major problem both in summer and *kharif* crops. These soil-borne pathogens being randomly distributed in soil and surviving as resistant resting structures are difficult to control by fungicides. This problem can be tackled through use of botanicals or biocontrol agents and their integration with fungicides. Keeping this in view, investigations were undertaken to evaluate the efficacy of bio-control agents, neem formulations and fungicides for suppression of *R. solani* and *F. solani in vitro* and in field for management of guar root rot, and the results are presented in this paper.

MATERIALS AND METHODS

In vitro evaluations of biocontrol agents

Eighteen potential fungal bio-control agents, 17 isolates of *B. subitlis* and 4 *Streptomyces* isolates isolated from disease suppressive rhizosphere soils of guar and other plants were evaluated *in vitro* against *F. solani* and *R. solani* by dual culture method (Dennis and Webster 1971), keeping three replications for each. The plates were incubated at 25±1°C. Observation on radial growth of the pathogen was recorded after 5 days on inoculation and per cent growth inhibition was calculated.

In vitro evaluation of neem formulations and fungicides

Five neem formulations (seed extract based, oil based-1, kernel extract based, achook and oil based-2) received from M/s Godrej Agrovet Limited, Mumbai was evaluated against *R. solani* and *F. solani* by poison food method at 0.2 per cent concentration *v/v* and per cent inhibition of the growth calculated. Four popular fungicides - Captan, Carbendazim, Copper oxychloride and Mancozeb were tested *in vitro* by poison food method against the two pathogens at 5 concentrations *viz*. 50, 100, 250, 500 and 1000 ppm.

Evaluation in green house

Five promising fungal BCAs, 5 *B. subtilis* isolates and *Streptomyces* sp. Ch-Br-b-2, 3 neem formulations (oil based-1, kernel extract based and oil based-2) and 3 fungicides (Carbendazim, Mancozeb and Captan) were included in the studies as seed treatments on a susceptible guar cultivar Pusa Nav Bahar. A mixture of sterilized soil + FYM was used. Culture(s) of *F. solani and R. solani* were separately multiplied on corn meal sand (1:1) mixture at 25±1° C for 10 days, then mixed in equal parts (1:1), added to the equal amount of sterilized garden soil and thoroughly mixed. This

inoculum-soil mixture was filled on top of sterilized soil + FYM in fresh earthen pots (25 cm face diameter). Thus, each pot contained 3 kg garden soil + 0.5 kg FYM + 1 kg inoculated soil. For each treatment 4 pots as four replications were maintained. All the pots were lightly irrigated and kept for 20 days to allow establishment of the pathogens and curing of soil before sowing.

BCAs seed treatments

Cultures of fungal BCAs were individually grown on 2 per cent malt extract agar (MEA), while of the bacterial BCAs on King's 'B' medium. The spores / colonies so developed were harvested by suspending in 20 ml water in each Petri dish and mixed with sterilized fine clay (10 g) to make slurry. This mixture was used for seed coating. The coated seeds were kept overnight in moist chamber, so as to enable the antagonists to establish on seeds.

Seed treatment with neem formulations and fungicides

Seeds were treated by soaking in the solution of individual neem formulation @ 0.2 per cent or with the fungicides for 30 minutes, air dried in shade and used for sowing. Drenching of neem formulations or fungicides was done after 30 days of sowing at the same concentration that was used for seed treatments by irrigating 50ml suspension around the plants in each pot.

Sowing and inter culture operations

The variously treated seeds were sown in pots @ 10 seeds pot^{-1}, keeping 4 replications for each treatment. Samples of 5 seeds were randomly drawn to determine inoculum density of the individual biocontrol agent in each bio-control agent's seed treatment. Just before sowing, soil samples were taken from each pot at the depth of 2 inches to determine initial population densities of the two pathogens.

Observations

Seed germination was recorded after 10 days of sowing. Observations on root-rot incidence were recorded at 45 and 60 days after sowing by counting total number of plants and root-rot infected plants in each treatment. Dry biomass weight of guar plants was recorded in each treatment after harvesting and oven drying of plants after 60 days of sowing. To determine the population of biocontrol agents and their possible effect on *F. solani* and *R. solani* soil samples from guar rhizosphere and around from both, diseased and healthy plants were collected after 45 and 60 days of sowing.

The population densities (c.f.u.) of all biocontrol agents and the two pathogens were determined by dilution plating (Warcup, 1950) on organism specific media. For *Trichoderma,* specific medium- Modified PDA Triton x-100 (Budge and Whipps, 1991) and for bacterial BCAs King's B medium (King *et al.,* 1954) was used. For *F. solani*, peptone PCNB medium (Nash and Snyder, 1962), and for *R.* solani modified rice agar medium (Mathur and Bohra, 2004) was used.

The data recorded in 2 runs of the experiment were subjected to pooled analysis of variance and least significant difference (critical deviation) determined at 5 per cent probability. Treatment means were compared using C.D. (critical difference) to determine efficacy of the different treatments.

RESULTS AND DISCUSSION

***In vitro* evaluation of BCAs**

Maximum and significantly high inhibition (75.1%) of the growth of *F. solani* was caused by *T. viride* (S) (Cent) followed by *T. harzianum* (Cumin) 15R95 (70.5 %), *T. harzianumn* Gn1 (63.6 %), *T. viride* (Cumin) 7R95 (58 %) and *T. viride* Tricho 20 (Akola) (52.2 %), respectively (Table 1). Complete growth inhibition of *R. solani* was obtained with *T. viride* (S) followed by with *T. viride* (Cumin) 7R95 and *T. harzianum* Gn1, where per cent growth inhibition was 78.4 and 64.7, respectively (Table 1).

Table 1. *In vitro* evaluation of biocontrol fungi against guar root-rot pathogens by dual culture method

Biocontrol fungi	Per cent growth inhibition*	
	Fusarium solani	*Rhizoctonia solani*
Trichoderma harzianum (Ginger) RAU Thj 89-2	40.5 (39.5)	43.1 (41.0)
Trichoderma longibrachiatum (Ginger) RAU Tlj 90-1	42.5 (40.7)	44.9 (42.0)
Trichoderma harzianum Gn1	63.6 (52.8)	64.7 (53.5)
Trichoderma viride (Cumin) 7R95	58.0 (49.6)	78.4 (62.3)
Trichoderma harzianum Tricho 28 (Ananthnag)	41.8 (40.2)	41.2 (39.9)
Trichoderma harzianum NRCG	49.5 (44.7)	48.4 (44.1)
Trichoderma harzianum (Cumin) 8R95	43.6 (41.3)	43.1 (41.0)
Trichoderma viride Tricho 20 (Akola)	52.2 (46.2)	51.6 (45.9)
Trichoderma harzianum Tricho 26	32.5 (34.7)	29.7 (33.0)
Trichoderma sp. (Guar)	39.4 (38.8)	56.6 (48.7)
Gliocladium virens (Maize)	52.2 (46.2)	54.7 (47.7)
Trichoderma harzianum (Cumin) 15R95	70.5 (57.1)	57.1 (49.1)
Trichoderma viride (Cumin) 1R95	48.4 (44.0)	55.5 (48.1)
Trichoderma harzianum (Akola) 21	38.4 (38.2)	32.2 (34.5)
Trichoderma harzianum (Akola) 23	47.7 (43.6)	32.9 (35.0)
Penicillium sp. (Gn) Chittor 1	41.2 (39.9)	30.9 (33.7)
Trichoderma harzianum (Coriander)	39.7 (39.0)	27.3 (31.5)
Trichoderma viride (S)	75.1 (60.0)	100.0 (90.0)
Control	00.0 (00.0)	00.0 (00.0)
SEm±	0.32	0.31
LSD (p = 0.05)	0.92	0.88

*Average of 3 replications; Values in parenthesis are ArcsinÖpercentage

Among the bacterial BCAs, *B. subtilis* isolates Cot-Coi-b-2, Ch-Kp-b-1 and Ch-Br-b-1resulted in hundred per cent growth inhibition of *F. solani* followed by isolates Cot-and-b-1, Cot-Hmg-b-2, Ch-Kp-b-3 and Cor-Kp-b-1 that caused 78.2, 69.2, 62.9 and 45.3 per cent growth inhibition, respectively (Table 2). Of the 4 isolates of *Streptomyces* tried, isolate Ch-Br-b-2 caused 82.9 per cent inhibition of the growth of *F. solani* followed by isolate Ch-Br-b-3 (74%) (Table 2). Maximum growth inhibition (86.9 %) of *R. solani* was recorded in Ch-Br-b-3 followed by isolate Ch-Br-b-2 (75.5%) and isolate Ch-Br-b-1 (58.8%), while, isolate Cot- Coi-b-1 caused least growth inhibition (26.6%) of *R. solani* (Table 2).

Table 2. *In vitro* evaluation of isolates of *B. subtilis* and *Streptomyces* spp. against guar root-rot pathogens by dual culture method

BCAs	Per cent growth inhibition*	
	Fusarium solani	*Rhizoctonia solani*
Bacillus subtilis Ch-M-b-1	27.1 (31.4)	35.7 (36.6)
Bacillus subtilis Ch-G-b-3	12.9 (21.0)	29.6 (32.9)
Bacillus subtilis Gr-Dr-b-1	5.5 (13.5)	19.6 (26.2)
Bacillus subtilis Gu-Ud-b-1	16.2 (23.7)	38.4 (38.2)
Bacillus subtilis Gu-Ud-b-2	2.9 (7.8)	28.4 (32.2)
Bacillus subtilis Gr-R-b-1	5.1 (13.5)	32.5 (34.7)
Bacillus subtilis Gr-R-b-2	5.5 (13.0)	30.7 (33.6)
Bacillus subtilis Gr-Mb-b-1	8.8 (17.2)	28.8 (32.4)
Bacillus subtilis Ch-Br-b-2	2.1 (8.4)	18.4 (25.4)
Bacillus subtilis Cor-Kp-b-1	45.3 (42.3)	48.4 (44.0)
Bacillus subtilis Ch-Kp-b-3	62.9 (52.4)	82.3 (65.1)
Bacillus subtilis Cot-Coi-b-2	100.0 (90.0)	100.0 (90.0)
Bacillus subtilis Cot-And-b-1	78.2 (62.1)	100.0 (90.0)
Bacillus subtilis Ch-Kp-b-1	100.0 (90.0)	100.0 (90.0)
Bacillus subtilis Cot-Hmg-b-2	69.2 (57.2)	100.0 (90.0)
Bacillus subtilis Ch-Br-b-1	100.0 (90.0)	78.8 (62.6)
Bacillus subtilis Ch-Br-b-2	12.2 (20.4)	35.1 (36.3)
Streptomyces sp. Cot-Coi-b-1	26.4 (30.9)	26.6 (31.0)
Streptomyces sp. Ch-Br-b-1	28.4 (32.2)	58.8 (50.0)
Streptomyces sp. Ch-Br-b-2	82.9 (65.5)	75.5 (60.3)
Streptomyces sp. Ch-Br-b-3	74.0 (59.3)	86.9 (68.8)
Control	00.0 (00.0)	00.0 (00.0)
SEm±	1.04	0.31
LSD (p = 0.05)	2.97	0.90

*Average of 3 replications; Values in parenthesis are ArcsinÖpercentage

Kernel extract based neem formulation caused maximum growth inhibition of both, *F. solani* (47.3%) and *R. solani* (64.2%), followed by oil based neem formulations (Table 3). Of the fungicides, both carbendazim and mancozeb were found to inhibit complete growth of *F. solani* as well as *R. solani* at all the tested concentrations (Table 4).

Evaluation of biocontrol agents, neem formulations and fungicides for management of root-rot in guar

The guar seed germinated well in all the treatments but there was significant reduction in germination in inoculated, untreated control (Table 5), where only 50 per cent germination was recorded. Highest germination (95%) was observed in *T. harzianum* Gn1 and *B. subtilis* Ch-Kp-b-1 followed by 87.5 per cent in *B. subtilis* Ch-Br-b-1. About 85

Table 3. *In vitro* evaluation of Neem based formulations against guar root-rot pathogens by poison food method

Type of neem formulation	Per cent growth inhibition*	
	Fusarium solani	*Rhizoctonia solani*
Seed extract based neem formulation (0.2%)	25.0 (29.9)	53.8 (47.2)
Oil based neem formulation-1 (0.2%)	28.5 (32.2)	25.9 (30.6)
Kernel based neem formulation (0.2%)	47.3 (43.4)	64.2 (53.2)
Achook 0.15% EC (0.2%)	20.3 (26.7)	30.8 (33.7)
Oil based Neem Formulation-2 (0.2%)	37.6 (37.8)	56.1 (48.4)
Control (Unamended)	00.0 (00.0)	00.0 (00.0)
SEm ±	0.10	0.19
LSD (p = 0.05)	0.32	0.58

*Average of 3 replications; Values in parenthesis are ArcsinÖpercentage

Table 4. *In vitro* evaluation of Fungicides against guar root-rot pathogens by poison food method

Conc. (ppm)	Per cent Growth Inhibition*							
	Carbendazim		Copper oxychloride		Captan		Mancozeb	
	F. solani	*R. solani*	*F. solani*	*R. solani*	*F. solani*	*R. solani*	*F. solani*	*R. solani*
1000	100.0	100.0	24.2 (29.4)	42.3 (40.5)	71.8 (57.9)	100 (90.0)	100.0	100.0
500	100.0	100.0	19.0 (25.8)	36.2 (37.0)	55.8 (48.3)	100 (90.0)	100.0	100.0
250	100.0	100.0	16.2 (23.7)	52.0 (46.1)	38.8 (38.5)	67.5 (55.2)	100.0	100.0
100	100.0	100.0	13.6 (21.6)	46.8 (43.1)	31.4 (34.1)	22.3 (28.2)	100.0	100.0
50	100.0	100.0	11.4 (19.7)	44.4 (41.7)	26.9 (31.2)	05.8 (13.9)	100.0	100.0
00	000.0	000.0	00.0 (00 .0)	00.0 (00 .0)	00.0 (00.0)	00.0 (00.0)	000.0	000.0
SEm±	0.23		0.17		0.37		0.22	
CD (p = 0.05)	0.71		0.53		1.14		0.68	

*Average of 3 replications; Values in parenthesis are ArcsinÖpercentage

per cent germination was recorded in the both *B. subtilis* Cot-and-b-1 and *Streptomyces* sp. Ch-Br-b-2, while 82.5 per cent seed germination was recorded in case of *B. subtilis* Cot-Coi-b-2, oil based neem formulation-2, carbendazim and captan. *T. viride* (S) and oil based neem formulation-2 resulted in 80 per cent seed germination followed by 77.5 per cent germination in *B. subtilis* Cot-Hmg-b-2 and kernel based neem formulation, 72.5 per cent in mancozeb and 70 per cent in *T. harzianum* (Cumin) 15R95 and *G. virens* (Maize). The lower germination (62.5%) was recorded in *T. viride* (Cumin) 7R95 among all seed treatments (Table 5).

The disease symptom started after 15-20 days of sowing and up to 45 days 70.4 per cent plants died in control (untreated seeds). At this stage all the treatments showed significantly less disease (5.9-35.3%) as compare to control. The treatment showing least disease (5. 9%) was *T. harzianum* Gn1 followed by *B. subtilis* Ch-Kp-b-1 and *B. subtilis* Ch-Br-b-1, where 6.2 and 8.5 per cent root-rot was observed respectively. The other isolate of *Trichoderma* and *Bacillus* showed 21.2 to 35.3 and 12.2 to 26.2 per cent disease, respectively as against 70.4 per cent disease in control. Oil based neem formulation-2 resulted in much reduced disease (18.7%), while, carbendazim and mancozeb resulted in 21.0 and 29.8 per cent root-rot. Of the 3 fungicides, captan showed good efficacy against the disease resulting into 17.7 per cent root-rot (Table 5).

At 60 days 95 per cent plants in untreated control had root-rot and most of these dried and disintegrated. At this time also *T. harzianum* Gn1 had the least disease (8.6%) followed by *B. subtilis* Ch-Kp-b-1, *B. subtilis* Ch-Br-b-1, *Streptomyces* sp. Ch-Br-b-2 and *B. subtilis* Cot-and-b-1, respectively, where disease incidence was observed 11.8, 15.2, 16.8 and 18.4 per cent respectively. In the remaining biocontrol agents 3 *Trichoderma* spp., *G. virens* (Maize) and *B. subtilis* Cot-Coi-b-2 about 30 to 56 per cent root-rot was recorded. Fungicide captan and oil based neem formulation-2 had significantly less disease (26.7 and 28.1 per cent respectively) as compared to carbendazim (37%) and mancozeb (47.8%).

All the treatments resulted in higher mean dry weight of the plants compared to non-treated control. The highest dry biomass weight was in *T. harzianum* Gn1 (9.7g) followed by *B. subtilis* Ch-Kp-b-1 (9.5 g), *B. subtilis* Ch-Br-b-1 (8.4 g), *Streptomyces sp.* Ch-Br-b-2 (8.1 g), *B. subtilis* Cot-and-b-1 (7.9 g), captan (7.8 g) and neem formulation 5 (7.3 g) (Table 6). *Streptomyces* sp. Ch-Br-b-2 resulted in good dry biomass weight (8.1 g) whereas, in 3 neem formulations, it was 5.4 (kernel extract based) to 7.3 g (oil based-2) and in case of 3 chemical fungicides it ranged from 5.0 (mancozeb) to 7.8 g (captan) (Table 5).

Table 5. Evaluation of biocontrol agents, neem formulations and fungicides for suppression of guar root-rot

Treatment	Per cent seed germination*	Per cent disease incidence*		Dry biomass of plants pot^{-1}* (g)
		45 DAS	60 DAS	
Trichoderma viride (S)	80.0 (63.7)	21.2 (27.4)	41.3 (40.1)	6.488
Trichoderma viride (Cumin) 7R95	62.5 (52.2)	35.3 (36.4)	55.6 (48.2)	2.918
Trichoderma harzianum (Cumin)15R95	70.0 (56.9)	34.3 (35.8)	48.1 (43.9)	3.428
Trichoderma harzianum Gn1	95.0 (80.7)	5.9 (14.0)	8.6 (17.1)	9.700
Gliocladium virense (Maize)	70.0 (56.9)	33.4 (35.3)	47.9 (43.7)	3.545
Bacillus subtilis Cot-Coi-b-2	82.5 (65.4)	19.5 (26.9)	30.1 (33.2)	6.798
Bacillus subtilis Ch-Kp-b-1	95.0 (80.7)	6.2 (14.4)	11.8 (20.0)	9.512
Bacillus subtilis Cot-And-b-1	85.0 (67.8)	12.2 (20.4)	18.4 (25.4)	7.912
Bacillus subtilis Cot-Hmg-b-2	77.5 (60.0)	26.2 (30.8)	42.5 (40.6)	5.095
Bacillus subtilis Ch-Br-b-1	87.5 (69.5)	8.5 (16.9)	15.2 (22.9)	8.477
Streptomyces sp. Ch-Br-b-2	85.0 (67.8)	10.9 (19.2)	16.8 (24.1)	8.167
Oil based neem formulation-1 (0.2%)	80.0 (64.1)	22.4 (28.2)	42.2 (40.4)	5.983
Kernel based neem formulation (0.2%)	77.5 (62.1)	22.7 (27.9)	42.3 (40.6)	5.448
Oil based neem formulation-2 (0.2%)	82.5 (65.4)	18.7 (25.6)	28.1 (32.0)	7.318
Carbendazim (0.1%)	82.5 (62.1)	21.0 (27.2)	37.0 (37.4)	6.532
Mancozeb (0.2%)	72.5 (58.9)	29.8 (33.1)	47.8 (43.7)	5.007
Captan (0.2%)	82.5 (65.8)	17.7 (24.8)	26.7 (31.1)	7.802
Control (untreated)	50.0 (44.9)	70.4 (57.0)	95.0 (74.2)	1.368
SEm±	3.3	0.4	0.6	0.093
LSD (p=0.05)	9.5	1.2	1.7	0.265

*Average of four replications, DAS = Days after sowing; Values in parenthesis are ArcsinÖpercentage

Table 6. Population densities of biocontrol agents and root rot pathogens in rhizosphere soil of guar

Treatment	Rhizosphere population (c.f.u. gram^{-1} soil)*					
	BCAs		*F. solani* (x10^5)		*R. solani* (x10^5)	
	45 DAS	60 DAS	45 DAS	60 DAS	45 DAS	60 DAS
Trichoderma viride (S)	10.0x10^5	11.0x10^5	2.3	1.6	1.0	7.0
Trichoderma viride (Cumin) 7R95	4.6 x10^5	19.0 x10^5	2.7	3.4	1.3	3.6
Trichoderma harzianum (Cumin) 15R95	5.1 x10^5	15.0 x10^5	4.9	6.2	2.0	5.0
Trichoderma harzianum Gn1	10.0x10^5	31.0 x10^5	2.7	1.3	1.0	2.0
Gliocladium virense (Maize)	1.0 x10^5	25.0 x10^5	3.7	3.6	2.0	1.6
Bacillus subtilis Cot-Coi-b-2	1.4 x10^9	1.2 x10^9	3.8	2.4	13.0	2.6
Bacillus subtilis Ch-Kp-b-1	1.9 x10^9	2.6 x10^9	2.1	2.8	1.3	1.8
Bacillus subtilis Cot-And-b-1	0.9 x10^9	1.7 x10^9	4.2	1.9	12.0	1.3
Bacillus subtilis Cot-Hmg-b-2	1.6 x10^9	1.0 x10^9	3.7	3.6	3.8	0.3
Bacillus subtilis Ch-Br-b-1	0.5 x10^9	1.4 x10^9	5.0	2.9	5.6	3.0
Streptomyces sp. Ch-Br-b-2	2.6 x10^9	2.3 x10^9	2.8	3.1	3.0	3.3
Oil based neem formulation-1 (0.2%)	-	-	2.7	4.8	2.6	1.0
Kernel based neem formulation (0.2%)	-	-	5.4	3.9	1.0	4.0
Oil based neem formulation-2 (0.2%)	-	-	1.9	3.2	2.3	3.6
Carbendazim (0.1%)	-	-	1.9	2.8	3.6	1.3
Mancozeb (0.2%)	-	-	1.6	4.0	12.0	3.6
Captan (0.2%)	-	-	1.3	2.4	1.3	3.0
Control (untreated)			6.9	7.2	33.0	36.0

Initial population of at sowing time : *F.solani* : 13.0 x 10^5 c.f.u. g^{-1} soil, *R.solani* : 3.1 x 10^5 c.f.u. g^{-1} soil, *Trichoderma* spp. : 1.7 - 2.0 x 10^6 c.f.u seed^{-1}, *B. subtilis* & *Streptomyces* sp. : 16.8 - 18.0 x 10^9 c.f.u. seed^{-1}
*Average of 3 replications

Population densities (c.f.u.) of the BCAs and the pathogens

Data presented in table 6 revealed that all the treatments significantly lowered the density of the two pathogens as compared to the untreated control. At 45 days, lowest population of *F. solani* was in captan (1.3 x 10^5) followed by mancozeb (1.6 x 10^5), carbendazim and neem formulation 5 (1.9 x 10^5). In different BCA treatments, it ranged from 2.3-5.0 x 10^5 c.f.u. g^{-1} soil. At 60 days the population of *F. solani* increased to 2.4 to 4.8 x 10^5 in neem and fungicide treatment, but in BCAs treatment it remained low to 1.3 to 3.6 x 10^5 c.f.u. g^{-1} soil except in *T. harzianum* (Cumin) 15R95, where it was 6.2 x 10^5 c.f.u. g^{-1} soil.

The initial population of *R. solani* at the time of sowing was 3.1 x 10^5 c.f.u. g^{-1} soil. The *R. solani* population at 45 days in *Trichoderma* spp. was 1.0-2.0 x 10^5, in *B. subtilis* treatments 3.8-13.0 x 10^5 and in neem and fungicide treatments was 1.0-12.0 x 10^5 c.f.u. g^{-1} soil as compared to 33 x 10^5 c.f.u. g^{-1} soil in untreated control.

At 60 days, the population density of *R. solani* in control increased and reached to 36.0 x 10^5 c.f.u. g^{-1} soil. While in neem formulation and fungicide treatments it remained low, ranging 1.3-4.0 x 10^5, but in *B. subtilis* and *Streptomycetes* spp. it ranged from 0.3-3.3 x 10^5 c.f.u. g^{-1} soil. With fungal BCAs, it was 1.6-7.0 x 10^5 c.f.u. g^{-1} soil.

The population of the fungal BCAs at this stage reached to 11-31 x 10^5 c.f.u. g^{-1} soil, of bacterial and actinomycetes ranged from 1.0-2.6 x 10^9 c.f.u. g^{-1} soil. Among the BCAs, *T. harzianum* Gn1 and *B. subtilis* Ch-Kp-b-1 developed to the maximum population densities at 60 days and these treatments had the lowest population of the two pathogens (Table 6).

Neem has been found to have superior antifungal activity against seed-borne pathogens of pea (Sharma *et al.*, 2003) and sunflower (Hussain *et al.*, 2000) and dry corm rot of colocasia caused by *R. solani* (Bhasker *et al.*, 2002). Suppression of soil- borne pathogens by *B. subtilis* strains has been reported (Roberti and Selmi, 1999) and also gray mold of tomato (Tsomlexoglon *et al.*, 2000). There are however, fewer reports about efficacy of use of strain of *Streptomyces* sp. A stain has been formulated for suppression of *Rhizoctonia solani* in tomato (Sabaratnam and Traquair, 2002). According to Weller *et al.* (2002) research on role of *Streptomyces* is warranted given their abundance in soil, ability to produce broad-spectrum antibiotics and grow under dry conditions.

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Development of *Mahua* (*Madhuca indica*) *Laddu* and its popularization in tribal areas

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ABSTRACT

Mahua flower is one of the most important non–timber forest produce (NTFP) in Jharkhand state, but, major part of *mahua* flower production (80 to 90%) is misused in the making and sale of unhealthy alcoholic liquor in Jharkhand state, just for quick and more earnings. As per collected information from the tribals, previously the same *mahua* flowers were consumed by tribals and others as *mahua laddu* also. But now-a-days, it is not in practice, because it has many drawbacks (look, shelf life, nutritive and sensory value *etc.*). Therefore, an on farm trial (OFT) was conducted to prepare *mahua laddu* scientifically with the objective to overcome all drawbacks of traditional *mahua laddu* and for healthy utilization of *mahua* flowers in place of harmful liquor. Three options were tried in OFT, each with 8 replications. In first option, as farmers' practice (FP), *mahua laddu* prepared with traditional method and second and third options were scientific methods *i.e.* Technological Option I (TO I) and Technological Option II (TO II). *Mahua laddus* were prepared with scientific method by using local resources and local facilities of rural areas. After developing technology for *laddus*, their nutritive value was estimated and sensory evaluations (of six parameters) were done through score card method. The economics were also calculated. The result revealed that *mahua laddus* prepared by TO II was richest in many nutrients among the three options. This *mahua laddus* were with protein (19.44 g), carbohydrate (135.38 g), energy (135.38 Kcal) and calcium (63.74 mg). The overall acceptability on the date of preparation of FP, TO I and TO II were 6.87, 7.77 and 8.75 respectively. So, it was clear that *mahua laddu* of TO II was liked most by farm women and others. After calculation, it was found that the cost of *mahua laddu* (per kg) of FP, TO I and TO II were 32.40, 60.00 and 58.00 rupees respectively. When, shelf life of *mahua laddus* were studied, it was found that *mahua laddus* of TO I and TO II had shelf life of 5 weeks from the date of preparation, whereas *mahua laddus* of FP had shelf life hardly of 1 week only. Traditional *mahua laddus* (of FP) was found on third position in the studied parameters (nutritive value, shelf life and sensory evaluation), except for economic point of view. Thus, the scientific *mahua laddus* (TO II) were popularized through the demonstration in many block level and district level *kisan melas*, exhibitions and through distribution of leaflets and handout written in Hindi among farmers and other persons, who visited *kisan mela* stall in bokaro and in other parts of Jharkhand state and the response was very encouraging.

Key words: *Mahua* flower, sterilized *mahua* syrup, nutritive value, sensory evaluation, shelf life

Jharkhand literally means "forest region", where 29 per cent (2.3 million hectares) of its land is under forest cover. More than half of the tribal population (28% of total population) depends on the non–timber forest produce (NTFPs) for livelihood. The four major NTFPs of Jharkhand are *karanj, chironjee*, lac and *mahua*. Among which, *mahua* generates maximum revenue (Gharai and Chakrabarti, 2009). Jharkhand produces 2 lakh metric tonne of *mahua* flowers (worth crores of rupees) annually (http://times of india.indiatimes.com cited on 17-7-2016). The flowers are produced during the leanest season of cultivation *i.e.* March-April (Patel and Naik, 2010, http//:www.banajata.org cited on18-4-2016) and consumed amongst locals as food. *Mahua* flowers are highly nutritious and eaten raw or cooked by the local tribal people (http//:www.banajata.org cited on18-4-2016). According to the National Institute of Nutrition, Hyderabad, the nutritive value of ripe *mahua* flowers per 100 g is, energy 111 Kcal, moisture 73.6 g, protein 1.4 g, fat 1.6 g, minerals 7 g, carbohydrate 22.7 g, calcium 45 mg, phosphorus 22 mg, iron 23 mg, carotene vitamin A 307 microgram and vitamin C 40 mg (Gopalan *et al*., 2004). There are many health benefits of *mahua* flowers *e.g*., its use as pain killer or act as analgesic and have shown a significant control of diabetes by control of hormones (http://www.satvikshop.com cited on 18-4-2016). Furthermore, tribes dry and press flowers to preserve it for 6 months for food and 2 years for wine (jharkhandindustry.gov.in cited on 18-4-2016). Women play a vital role in all work related to *mahua* processing in rural areas, from collection to marketing. They generally travel an average of 2 to 3 kms to collect *mahua* flowers and then it is sundried. The dried *mahua* flowers are either sold directly or processed to make liquor. In each stage, women's involvement is quite significant (Gharai and Chakrabarti, 2009). Production of liquor from *mahua* flowers is a traditional practice from centuries (Yadav *et al.*, 2009). In spite of the fact that *mahua* flowers are nutritious, major portion *i.e.* 80 to 90 per cent of its total production is misused in the production and sale of unhealthy alcoholic liquor, just for getting quick

earnings. As per the information provided by local tribes, the sun-dried *mahua* flowers were previously consumed as *mahua lattha* (similar to *laddu*) or *mahua laddu*. But, now-a-days, the consumption of *mahua laddu* is not in practice due to several reasons. First drawback of the traditional *mahua laddu* was it's black colour, which was not appealing. Second, it's characteristic strong smell and third drawback was its short shelf life. So, due to these drawbacks, gradually it's consumption became out of practice by local tribals. Therefore, the present study was undertaken and *mahua laddus* were prepared scientifically maintaining it's nutritive value, longer shelf life and sensory acceptability.

MATERIALS AND METHODS

In the On Farm Trial (OFT), three preparation methods with 8 replicates were evaluated. In first method *i.e.* Farmers' Practice (F.P.), *mahua laddu* was prepared as per traditional method of tribals. In second and third option *mahua laddues* were made with scientific intervention as Technological Option I (TO I) and Technological Option II (TO II) (Agrawal and Bhotmange, 2010) by using different local resources available in rural areas.

Farmers' Practice (F.P.)

Collection, sun-drying, stamens (jhilli) removal and cleaning of *mahua* flower

Local farm women collect fresh and ripe *mahua* flowers by hand picking of spontaneously dropped flowers on the ground from the tree and keep it for sun drying. After proper sun-drying, they store it in plastic bags. For preparing *mahua laddu*, already sun-dried *mahua* flowers are again sun-dried because the stored *mahua* flowers absorb moisture from atmosphere, as these are hygroscopic in nature (www.thelivelihoodschool.in cited on 17-7-2016). According to tribal people, its stamens (local people call it jhilli), are responsible for drossiness effect. So, after double sun-drying flowers are beaten with bamboo stick for the removal of stamens by winnowing. The next process is cleaning. But, tribals have no concept of thorough cleaning. They just clean it by through winnowing process.

Laddu making through farmers' practice

After cleaning of *mahua* flowers, both *mahua* flowers and wheat flour are roasted separately in big hot earthen pots, without using any medium like sand or oil. When both roasted materials are mixed well (in 2:1 ratio) homogenously. Then, *laddu*s are made out of it by rolling it in hand for making small round shape by taking few drops of water on the palm. They store these *mahua laddu* in plastic jar and keep it in cool and dry place in their home.

Laddu making through Technological Option I and II

In the scientific method, first four steps were same as in farmers' practice i.e. (i) collection of flowers (ii) sun drying (iii) repeated sun drying and (iv) stamens (jhilli) removal.

In both scientific process i.e. TO I and TO II, thorough cleaning of *mahua* flower was done. It was thoroughly washed with luke warm water repeatedly, until clean water start coming. Extra water is removed by pressing thoroughly and sun drying. Since, dried *mahua* flower are rich in sugar, it must be sterilized for complete removal of pathogens (Agrawal and Bhotmange, 2010). For sterilization *mahua* flowers are cooked in pressure cooker with potable water in 2:1 ratio. After first whistle, flame was kept at minimum for 30 minutes for proper sterilization. It is filtered through muslin cloth and the liquid portion is squeezed out. The filtered part (liquid portion) is known as *mahua* syrup.

In TO I and TO II, wheat flour/maize flour and groundnut flour were roasted in big hot earthen pot separately, without using any medium like sand or oil similarly as in Farmers' Practice. In TO I, *mahua laddu*s were prepared by using sterilized *mahua* syrup (in place of direct *mahua* flower), roasted wheat flour, roasted groundnut flour in ratio of 8:4:3 along with sugar as per requirement and GMS (an anti staling agent - 0.1% on the basis of total weight of raw ingredients). In TO II, *mahua laddu*s were prepared through same scientific process by using same raw ingredients, except maize flour, which was used in place of wheat flour. These ingredients are available in plenty in villages of Bokaro district.

For making *mahua laddu*s, sugar solution is prepared with the help of *mahua* syrup and sugar. Roasted wheat/ maize flour and groundnut flour are mixed well homogenously as per the technological options I and II separately. From this mixture, *laddu*s are prepared in the same way as in farmers' practice. These are then stored in plastic jars in cool and dry place separately.

Nutritive value, sensory evaluation, shelf life and cost analysis of *mahua laddu*

Nutritive value, sensory evaluation, shelf life and cost analysis of all three sets of *mahua laddu*s were done by common steps.

Nutritive value of *mahua laddu* were evaluated as per the method Gopalan *et al*. (2004). Nutritional composition of *mahua* flower is, moisture 19.8 per cent, protein 6.37 per cent, fat 0.5 per cent, total sugar 54.06 per cent, ash 4.36 per cent, calcium 8 per cent, phosphorus 2 per cent (Sunita and Sarojinj, 2013).

Sensory evaluation was done by farm women. As they were illiterate, simple score card method was used, because it is simple, easy to understand and easy to calculate. In sensory evaluation, six parameters were considered *e.g.* appearance, colour, texture, flavour, taste and overall acceptability. Sensory evaluation was done at three different intervals *i.e.* on the date of preparation, after 3 weeks and after 5 weeks from the date of preparation. On the date of preparation, all three sets of *mahua laddu*s were stored in simple plastic jar and kept at their home in cool and dry place and changes in appearance, colour, flavour, texture and taste were closely monitored upto 5 weeks from the date of preparation.

Cost analysis was done by calculating the cost of ingredients and preparation steps. Statistical analysis was done by standard method with the help of percentage, mean and standard deviation.

RESULTS AND DISCUSSION

Nutritive value of *mahua laddu*

Nutritive value of all three sets of *mahua laddu i.e.* Farmers' Practice, TO I and TO II are presented in table 1.

Protein, fat, carbohydrate, energy and calcium content were highest in *mahua laddu* prepared by Technological Option II followed by Technological Option I. It was interesting to note that *laddu* prepared by Technological Option II and Technological Option I were significantly quite rich in nutritive value.

Only the iron content was found less *i.e.* 1.98 mg 100^{-1}g in *mahua laddu* prepared by Technological Option II compared to Technological Option I (4.06 mg 100^{-1}g). So, on the basis of these findings, it was very clear that *mahua laddu* prepared by Technological option II was best and followed by Technological option II from nutrition point of view compared to Farmers' Practice.

Sensory evaluation

All three sets of *mahua laddu*s were kept for sensory evaluation at the three different intervals, *i.e.* on the date of preparation, after 3 weeks and after 5 weeks from the date of preparation.

Sensory evaluation on the date of preparation

First sensory evaluation was done on the date of preparation by farm women and finding is given in table 2.

Appearance: The appearance on the date of preparation of *mahua laddu* of TO II (score 8.87) was very appealing due to its light yellow colour, whereas *laddu* prepared by TO I was on the second position (7.55).

Colour: Colour of *mahua laddu* of TO II was of light yellow colour (score 8.75) and liked by all farm women most, whereas the colour of Farmers Practice was dark black and that's why it was on third position in preference. Colour of *laddu* prepared by TO I was of dark wheat colour (score 7.88) and on second position.

Texture: Both *mahua laddu*s of TO II (score 8.62) and of TO I (score 8.11) were firm and solid in texture and hence, maintained it's shape after keeping in plastic jar, whereas, by Farmers' Practice it was very soft and that's why not remained in shape, when kept in plastic jar and all *mahua laddu* stick with each other and became deformed. That's why texture of *mahua laddu* of TO II and of TO I were more preferred as compare to *mahua laddu* of Farmers practice.

Flavour: TO II based *mahua laddus* were most preferred because of having very pleasant cum sweet aroma due to the presence of both roasted maize and roasted groundnut. *Mahua laddu* of Farmers' Practice had strong characteristics sweet smell just like of fresh ripe *mahua* flower. So, it was on second position. *Mahua laddu* prepared by TO I had almost similar flavour as of TO II, but in lesser amount.

Table 1. Nutritive value of all three sets of *mahua laddu* (per 100 g)

Nutrient *Mahua laddu*	Protein (g)	Fat (g)	Carbohydrate (g)	Energy (Kcal)	Calcium (mg)	Iron (mg)
Farmers' Practice	3.92	1.28	56.04	251.43	27.47	1.44
Technological Option I	14.76	15.52	101.30	604.04	62.14	4.06
Technological Option II	19.44	22.04	135.38	818.18	63.74	1.98

Table 2. Mean value of six parameters of sensory evaluation through score card method on the date of preparation

Parameters/ *Mahua laddu*	Appearance	Colour	Texture	Flavour	Taste	Overall acceptability	Mean ± S.D.
Farmers' Practice	7.00	7.37	7.25	8.62	6.87	6.87	7.33±0.66
TO I	7.55	7.88	8.11	7.77	7.75	7.77	7.80±0.18
TO II	8.87	8.75	8.62	8.87	8.77	8.75	8.77±0.09

Yadav *et al*. (2009) conducted a study on value addition of *mahua* flower in which the potential of *mahua* as a nutra beverage was evaluated on the basis of total phenolic content (TPC) and antioxidant value and found that *mahua*-guava based product showed higher degree of protection against lipid peroxidation (Antioxidant property). Thus, these blending approaches could be adopted for the improvement of the antioxidant potential of *mahua*-based fortified products which enhanced the nutritional value of the final product, mask the unpleasant flavour of *mahua* and improve the texture of the product. A study conducted by Soni and Dey (2013) on value addition of *mahua* and found that currently several beverages and food products are marketed based on their antioxidant capacity.

Taste: TO II based *mahua laddus* had excellent taste followed by TO I, as per the feedback given by the farm women. *Mahua* syrup with roasted cereal flour (wheat or maize) and roasted groundnut flour improved the taste of *mahua laddu*, which was lacking in Farmers' Practice.

Overall Acceptability: When evaluation was done on the basis of overall acceptability TO II was found best (8.75) followed by TO I (7.77). Farmers' practice was on third position with score of 6.87. Patel and Naik (2008) on value addition of fresh *mahua* flower advocated that fresh fleshy corollas were also processed into jam, jelly and sauce and subjected to sensory evaluation and shelf life study. All the developed products were highly acceptable and stable at low temperature.

Sensory evaluation after 3 and 5 weeks from the date of preparation: To study the changes occurred in the parameters of sensory evaluation with time, continuous monitoring was done upto 5th week. Since, *mahua laddu* prepared through Farmers' Practice got spoiled hardly in one week, that's why only two sets of *laddu*s were left for monitoring sensory evaluation. Information related to these findings are given in table 3 and 4.

The appearance, colour, texture, flavour, taste and overall acceptability of TO II was better after 3 weeks of storage scoring 8.77, 8.50, 8.00, 8.55, 8.37, 8.555 respectively compared to *laddu* prepared by TO I, where score was 7.25, 7.75, 8.00, 7.44, 7.44, 7.62 respectively. After 5 weeks also the trend was similar and TO II performed better compared to TO I (Table 4).

The reasons behind better sensory score of TO II and TO I are due to first, thorough and proper cleaning of *mahua* flowers, second due to the use of sterilized *mahua* syrup in place of direct sun-dried *mahua* flowers and third, due to the use of anti-staling agent (GMS- Glycerol mono stearate). Therefore, it is very clear that just by using scientific method in place of traditional method, very good change occurred in the studied parameters of sensory evaluation.

Shelf life of three sets of *mahua laddu*: In the present study, it was found that sun-dried *mahua* flower is very rich in sugar and contained lots of dirt and dust particles. As in Farmers' Practice, cleanliness is not maintained, that's why just at the end of one week, lot of fungus developed over it, whereas, both *mahua laddu*s *i.e.* TO II and TO I were in very good condition even after 5 weeks from the date of preparation because these were developed from sterilized *mahua* syrup after thorough cleaning process and use of GMS. In another study conducted by Patel and Naik (2008) on *mahua* juice concentrate (MJC) and shelf life study revealed that the product (MJC) was stable at low temperature and high scores of sensory evaluation proved its acceptance as a food supplement. Utilization of the concentrate in confectionery (candy) and bakery products (biscuits, cakes, cookies etc.) at different concentration, proved the industrial applicability of the product. If these *mahua laddus* are stored in low temperature, the shelf life can be increased.

Cost analysis of all three sets of *mahua laddu*: After calculation, it was found that the cost of *mahua laddu* per kg prepared under Farmers' Practice, TO I and TO II were

Table 3. Mean value of six parameters of sensory evaluation through score card method after 3 weeks from the date of preparation

Parameters/ *Mahua laddu*	Appearance	Colour	Texture	Flavour	Taste	Overall acceptability	Mean±S.D.
Farmers' Practice	-	-	-	-	-	-	-
TO I	7.25	7.75	8.00	7.44	7.44	7.62	7.58±0.26
TO II	8.77	8.50	8.00	8.55	8.37	8.55	8.45±0.25

Table 4. Mean value of six parameters of sensory evaluation through score card method after 5 weeks from the date of preparation

Parameters/ *Mahua laddu*	Appearance	Colour	Texture	Flavour	Taste	Overall acceptability	Mean ±S.D.
Farmers' Practice	-	-	-	-	-	-	-
TO I	7.00	7.44	7.00	6.62	7.25	7.00	7.05±0.27
TO II	8.25	8.44	7.33	7.22	8.25	8.00	7.91±0.51

Rs. 32.40, 60.00, 58.00 respectively. In TO I and TO II raw ingredients used were same, except the cereal grain flour. In TO I, wheat flour was used, whereas in TO II maize flour was used. Since, in making of *mahua laddu* through Farmers' Practice method, only sun-dried *mahua* flower and wheat flour was used, that's why cost of Farmers' Practice was low. Whereas, in TO I and TO II total raw ingredients used were sun-dried *mahua* flower, either wheat or maize flour, groundnut, sugar and GMS, hence the cost was higher. The difference in price of TO I and TO II was very less because the difference of price of wheat flour and maize flour per kg was very less. Though, the price of *laddu* was higher in TO I and TO II, but the quality and shelf life was also far better compared to Farmers' practice. Hence, these can be graded superior. Patel and Naik (2010) working on value addition of *mahua* flowers found that about 80 per cent of juice was successfully extracted from fresh flowers and concentrated to produce a honey like liquid sweetener. The produced concentrate was analyzed and used for preparation of bakery and confectionary goods. In present study *mahua* syrup was used for preparing *laddu* in TO I and TO II.

Popularization of scientifically developed *mahua laddu*: For its popularization in rapid way and among large section of local rural people, *kisan mela* of different block of bokaro district was best suited. So in Chandanqyari block *kisan mela*, Petarwar block *kisan mela etc.* the scientifically developed *mahua laddu*s (TO II) was kept for sale and popularization and the response were very good. In district level *kisan mela* organised by Birsa Agricultural University, Ranchi, also, it was popularized and sold. In all *kisan melas*, leaflet and handout developed on this subject (written in Hindi language) was distributed among all farmers and other persons who came at stall of *kisan mela* and response was very encouraging. A study conducted by Kumari and Raghuvanshi, (2012) also popularized buckwheat, an underutilized cereal grain of Uttarakhand state through *kisan mela*.

Mahua tree is considered as life line for tribal community because it is multipurpose tree, source of food, medicine and income for them. Collection of *mahua* flowers is one of the most important sources of employment for the poorest of the poor in Jharkhand. Making *laddu* and it's maketing will be very useful for the nutritional and economic security point of view. More over, it will also restrict it's use for making liquor, which is harmful for the society.

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Agro-biodiversity among tribal homesteads of the Nilgiris district of Tamil Nadu

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ABSTRACT

Agro-biodiversity in this study referred as, variety of components such as cereals and millets, vegetables, fruits, spices and condiments, plantation and livestock present in the tribal homesteads of the Nilgiris district. The study was conducted at tribal areas of Nilgiris district, which is known as biodiversity hot spot. The list of tribal respondents from selected village was obtained from horticulture department. A sample size of 100 homesteads respondents were fixed for the study. The technique proportionate random sampling was followed for the selection of respondents from three habitations *viz*., kunjappanai, mantharai, thuthikarai. Two ecological indices were used to analyse the agro-biodiversity *viz*., species diversity among tribal homesteads based on Shanon-Wiener index and species richness in tribal homesteads by using Margalef Index. Majority of the tribal homesteads had medium level of diversity and species richness. A participatory group approach is needed in the tribal areas to conserve agro-biodiversity.

Key words: Homestead, species diversity and richness, tribal

Homestead farming is a very old tradition that has evolved over a long period of time from the practice of the hunters/gatherers and continued till now. It was started as a system for the production of subsistence crops for the household with or without involvement of cash crops. Tribal homesteads are traditional agro-forestry systems in which perennial, annual crops are grown and the seasonal crops are grown as intercrop often without any definite space arrangement. Tree crops and other components like vegetables, birds, and domestic animals are essentially important components in tribal's homesteads. Homestead farming is a highly complex and dynamic combination of different crops and livestock for achieving food, nutritional and economic security of the tribal's through the efficient utilization of available resources like land, water, solar energy and manpower.

MATERIALS AND METHODS

The list of tribal respondents from selected village was obtained from horticulture department. A sample size of 100 homesteads respondents were fixed for the study. The technique proportionate random sampling was followed for the selection of respondents from three habitations *viz.*, Kunjappanai, Mantharai, Thuthikarai from Kothagiri taluk of Nilgiris district.

The Shannon-Wiener index

The Shannon-Wiener index is a popular diversity index also knows as Shannon–Weaver index and the Shannon entropy. The measure was originally proposed by Claude. Shannon-Wiener Index is the most commonly used diversity index in plant communities and it takes a value of zero when there is only one species in a community and a maximum value when all species are present in equal abundance.

The following equation used for this study looks at the diversity of species in tribal homesteads.

$$H = \sum_{i=1}^{s} pi \ln pi$$

where,

S= No. of species

i = No. of individuals

pi = proportion of species i relative to the total number of species

ln = Natural logarithm.

H= The Shanon-Wiener Index

The Margalef index

The index 'Margalef' is a commonly used index to assess species richness in plant communities. The following equation used for this study looks at the richness of species in tribal homesteads.

$$Da = S-1 / Ln(n)$$

Where,

S= Total no. of taxa

N= No. of individual in all species

Da = Margalef Index

After obtaining the Margalef Index values for each homestead separately, the mean index has been worked out in order to find the species richness.

Agro-biodiversity and tribal homesteads

Agro-biodiversity in this study referred as, variety of components such as cereals and millets, vegetables, fruits, spices and condiments, plantation and livestock present in the tribal homesteads of the Nilgiris district.

Homesteads represent a promising land use system which is common in hilly areas especially in tribal areas. In this present study attempt was made to understand the diversity and species richness of the tribal homesteads, for which around 100 homesteads were studied in the Nilgiris district of Tamil Nadu.

RESULTS AND DISCUSSIONS

Agro-biodiversity among tribal homesteads of the Nilgiris district

For the present study, following ecological indices were used to analyze and to get a clear picture of the agro-biodiversity in the tribal homesteads, which was used by Bishwajit *et al.* (2013). The indices are listed below.

a. Species diversity among tribal homesteads based on Shanon-Wiener index

b. Species richness in tribal homesteads by using Margalef Index

Species diversity among tribal homesteads based on Shanon-Wiener index

Species diversity was estimated in the tribal homesteads using Shanon-Wiener Index (Jayasree *et al*., 2013) and calculated the mean index separately for each homestead. Based on the mean score, tribal homesteads were categorized as low, medium and high levels of diversity and the results are given in table 1.

Table 1. Species diversity among tribal homesteads based on Shanon-Wiener index (n=100)

Diversity category	Mean index	No. of respondents (%)
Low level	0 to 0.20	38
Medium level	0.21 to 0.25	41
High level	0.26 to 0.28	21
Total		100

It could be observed from table 1 that, majority (41%) of the tribal homesteads had medium level of diversity index (0.21 to 0.25) followed by 38 per cent of the homesteads had low level of diversity and only 21 per cent of the tribal farmers had high level of diversity on their homesteads. It shows the declining rate of agro biodiversity.

It was observed that agro-biodiversity components such as cereals and millets, vegetables, fruits, medicinal plants, plantation crops and livestock were found in majority of the homesteads.

Further, it was noticed that besides other components, almost all the homesteads were covered with tree crops. Integrating trees on homesteads provides a viable solution for many problems in tribal areas such as depletion of agricultural lands and landslides. Tree crops serve as wind breaker, source of organic matter, shade and soil binder to prevent soil erosion, while generating additional income. The finding of present study is in line with the findings of Ashok Kumar (2011) who had also reported that planting tall growing tree crops on bunds is very common in tribal homesteads.

Cereals and millets and vegetables were not commonly grown in all the homesteads. The reason might be that, short duration crops like cereals and vegetables were not much preferred by the tribal's in their homesteads. Also, the tribal respondents expressed that cereals and millets required frequent care and intercultural operations which is very difficult in hilly areas. So they preferred to cultivate trees and other perennial crops.

Species richness in tribal homesteads of the Nilgiris district

In this study, species richness is referred as number of different species present in the following components *viz*., cereals and millets, vegetables, fruits, spices and condiments, plantation and livestock in the tribal homesteads of the Nilgiris district

Species richness was estimated in the tribal homesteads separately by using Margalef Index and the results are given below.

Table 2. Species richness based on Margalef index mean value (n=100)

Richness category	Mean index	No. of respondents (%)
Low level	0 to 2.82	26
Medium level	3 to 4.99	46
High level	5 to 5.86	28
Total		100

The table 2 revealed that, majority (46%) of the homesteads had medium level of species richness followed by high level (28%) and low level (26%).

Nearly fifty per cent of the homesteads had medium level of richness. During the survey it was observed that different fruit crops were commonly found in most of the homesteads such as banana, mango, orange, papaya and jack. With regard to plantation crops, tea, coffee and silver oak trees were commonly grown in all the homesteads. Among all the spices and condiments, pepper, ginger, coriander, cardamom and turmeric were commonly cultivated in most of the tribal homesteads. The vegetable crops such as carrot, tapioca, chilies, beans, cabbage and brinjal were predominantly grown in tribal homesteads. The reason might be due to the suitable climate in the hilly areas to support cultivating those crops with minimal cultivation practices.

The rest of the crops particularly, cereals and millets like crops in *viz*., ragi, varagu, sorghum and thinai were commonly cultivated in separate fields, which are far-off from their homesteads. This might be the reason for the less species richness in the tribal homesteads.

It can be observed from the research findings that, majority of the tribal homestead had medium level of both species diversity and richness. Especially 38 per cent of the homesteads had low level of species diversity. It shows the declining face of agro-biodiversity in tribal homesteads farming. There was a lack of scientific knowledge of the gardeners, an absence of proper planning and no specific objectives and goals. During the homesteads visit it was learnt that most of the crop species were not utilized for the past few decades and led to species loss. A participatory group approach is needed in the tribal areas to conserve agro-biodiversity. Government should provide subsidies and constant encouragement by giving trainings on planning and monitoring of homesteads farming systems.

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Organoleptic and proximate evaluation of processed pearl millet based instant *Upma* mix

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ABSTRACT

In the present study pearl millet was subjected to processing treatments *viz.* blanching, extrusion and germination and utilized in development of gluten free instant *Upma* mix along with incorporation of soybean grits. All types of instant *Upma* mixes were found to be organoleptically acceptable. Taste of blanched pearl millet based instant *Upma* mix was liked very much by panelists. Proximate evaluation was evaluated with standard procedures of AOAC. Maximum amount of crude protein was analyzed in germinated pearl millet based instant *Upma* mix 15.79 g 100 g^{-1}, whereas maximum ash and crude fibre content in extrusion processed pearl millet based instant *Upma* mix i.e. 2.92 and 2.67 g 100 g^{-1}, respectively, gluten was not present in developed instant *Upma* mixes. Developed product will not only cater to nutritional needs of gluten intolerance sufferers but also lead to diversification in pearl millet utilization and availability of low cost alternative.

Key words: Pearl millet, convenience, gluten free, proximate, celiac disease, instant *upma* mix

Upma is a traditional south Indian recipe. Being wheat based, it renders itself unsuitable for celiac disease sufferers. Pearl millet based instant *Upma* mix can provide convenience to celiac patients as well as normal people by means of minimum handling prior to consumption along with providing satisfaction to deprived palates of celiac disease patients.

Pearl millet grains are nutritionally comparable and even superior to other major cereals with respect to energy, protein, vitamins and minerals. Besides they are rich source of dietary fiber, phytochemical and micronutrients, hence, they are termed as "Nutri-Cereals". Pearl millet constitutes an important staple crop, especially for marginalized household for whom coarse cereals account for a larger share in daily diets than wheat (Ramaswami, 2002). Pearl millet (*Pennisetum glaucum*) is a gluten free grain and is the only grain that retains its alkaline properties after being cooked which is ideal for people with wheat allergy/celiac disease. Being gluten free, pearl millet products can conveniently be used by celiac or people suffering from gluten intolerance (Yadav *et al*., 2014). Celiac disease brings along several other complications like anemia, growth failure, osteoporosis and many others. To counter the complications, their diet needs to be supplemented with other grains (Kapur *et al*., 2003).

Processing of pearl millet grains alters the bioavailability of macro and micro nutrients. Digestibility of micronutrients improves upon processing by softening the food matrix, release of protein bound micronutrients and thus, facilitating their absorption. Processing influences the inherent factors that interfere with mineral absorption, such as dietary fiber and phytate. The main reasons of processing are to eliminate micro-organisms and to extend shelf life along with improving the nutritive value of pearl millet for its diversified uses.

Over the past several years, convenience foods have been popular and such food based upon pearl millet may emerge as promising products in the market. Convenience foods require minimum handling, such as mild heating/ warming for ready-to-eat products or rehydration in hot/ cold water for dehydrated foods (Arya, 1992). Sood *et al.* (2010) prepared ready to use *Upma* mixes by using cereals, pulses, vegetables/ fruits and nuts with soy-whey, which were found organoleptically acceptable. Balasubramanian *et al.* (2014) developed *Upma mix* using pearl millet semolina. Pearl millet grains were hydro-thermally treated to reduce anti-nutritional factors and inactivate lipase activity. Developed instant *Upma* mix was organoleptically acceptable and was stable for a period of six months.

The major objectives of this study were:

- To standardize instant *Upma* mix using blanched, germinated and extruded pearl millet incorporating soybean grits.
- To carry out organoleptic evaluation of develop instant *Upma* mix
- To study the nutritional composition of developed processed and unprocessed pearl millet based gluten free instant *Upma* mix.

MATERIALS AND METHODS

White pearl millet variety HHB-256 was procured from Bajra section of Department of Genetics and Plant Breeding, CCS Haryana Agricultural University, Hisar, while other ingredients were procured from the local market. The present investigation was conducted in the Department of Foods and Nutrition, I.C. College of Home Science, CCS Haryana Agricultural University, Hisar. Raw materials were then cleaned, washed, dried and stored in clean and hygienic condition for further use where as pearl millet grains were further subjected to processing treatments *viz.* Blanching, Germination and Extrusion.

Processing treatments

Blanching: Blanching was done by the process of Chavan and Kachare (1994). Distilled water was brought to boiling to 98°C in an aluminium container. The grains were subjected to boiling water (1:5 ratio of seeds to boiling water) for 30 seconds and dried at 50°C for 60 minutes.

Germination: Soaked seeds (12 hours) were kept in petridishes lined with wet filter paper for germination in an incubator at 30°C for 48 hours. Seeds were kept moist by sprinkling distilled water frequently.

Extrusion: Extrusion processing was done according to the method of Sihag *et al*. (2015). Grains were preconditioned to adjust the feed moisture content. The moistened grains were kept for 48 hours for preconditioning in airtight containers to equilibrate moisture. After 48 hours of conditioning, grains were fed into feeder hopper that contained screw auger to transport materials at uniform rate into the barrel. The single screw extruder consists of one screw in the barrel to transport the ingredients through its three zones, *viz.* feeding zone, kneading zone, and cooking zone. The temperature of cooking zone was maintained at 110°C. The material was finally extruded through a 3 mm diameter die, where it expanded due to sudden evaporation of water from plasticized mass. Finally extrudates were milled in the milling machine to obtain grits.

Table 1. Combination of ingredients for making instant *Upma* mix

Ingredient	Control (Unprocessed)	Type-I (Blanching)	Type-II (Extrusion)	Type-III (Germination)
Pearl millet grits (g)	90	90	90	90
Soybean grits (g)	10	10	10	10
Bengal gram (g)	5	5	5	5
Ghee (g)	3	3	3	3
Peas (g)	2	2	2	2
Carrot (g)	2	2	2	2
Onion (g)	2	2	2	2
Curry leaves (nos.)	3-4	3-4	3-4	3-4
Red chilli (tsp)	¼ tsp	¼ tsp	¼ tsp	¼ tsp
Salt (g)	3	3	3	3

Proximate composition: Proximate composition including moisture, crude protein, crude fat, ash and crude fiber were determined by standard methods (AOAC, 2000). Gluten content was assessed using hand washing method (AACC, 2000).

Standardization and development of instant *Upma mix*

Chopped and peeled vegetables
(carrot, onion, peas, green chilles and curry leaves)
↓
Blanched vegetables (1-2minutes)
↓
Dried vegetables in oven at 50°C
↓
Roasted bengal gram dal, mustard seeds and
Pearl millet suji (PMS) separately
↓
Added salt, chilli powder to taste
↓
Packed and sealed polyethylene pouches
↓
Upma mix (RTC)

Fig. 1. Flow chart of *Instant Upma Mix*

Preparation schedule for making *Upma* from *Upma Mix*:

Ingredients	Amount
Upma mix	100 g
Water	250 ml

Method

- Heat ghee in skillet and added *Upma* mix to it.
- Add boiled water and mixed quickly to avoid lumps formation.
- Cover the skillet with a lid.
- When all the water was absorbed by the mixture, the *Upma* was done.

RESULTS AND DISCUSSION

Data in table 2 presents the mean scores of organoleptic characteristics of gluten free instant *Upma* mixes. All the types of instant *Upma* mix ranked as 'liked moderately'. Mean scores in colour, appearance, aroma, texture and taste were highest for Type-I instant *Upma* mix than other types. Taste of

Table 2. Mean scores of organoleptic characteristics of gluten free *Upma* reconstituted from instant *Upma* mix based on processed and unprocessed pearl millet

Type of instant *Upma* mix	Colour	Appearance	Aroma	Texture	Taste	Overall Acceptability
Control (Unprocessed)	7.4±0.16	7.4±0.14	7.4±0.16	7.5±0.24	7.4±0.16	7.4±0.16
Type-I (Blanching)	7.8±0.23	7.7±0.21	7.8±0.20	7.9±0.20	8.0±0.23	7.8±0.20
Type-II (Extrusion)	7.6±0.22	7.5±0.13	7.4±0.21	7.8±0.22	7.5±0.22	7.5±0.26
Type-III(Germination)	7.4±0.15	7.4±0.18	7.6±0.13	7.4±0.15	7.3±0.10	7.4±0.21
CD (P<0.05)	0.56	0.52	0.54	0.60	0.53	0.61

Values are mean ± SE of ten independent determinations

Table 3. Proximate composition and gluten content of instant *Upma* mix (g $100g^{-1}$ on dry matter basis)

Treatment	Moisture	Crude protein	Crude fat	Ash	Crude fiber	Wet gluten	Dry gluten
Unprocessed (Control)	5.17±0.14	15.68±1.41	9.95±0.87	2.54±0.21	2.07±0.12	0.03±0.002	-
Blanching	5.28±0.08	15.57±1.02	7.50±0.71	2.69±0.24	1.27±0.15	0.04±0.002	-
Extrusion	4.79±0.03	15.40±1.56	8.91±0.35	2.92±0.34	2.67±0.21	0.02±0.003	-
Germination	5.96±0.12	15.79±1.67	7.26±0.21	2.39±0.33	2.38±0.09	0.02±0.002	-
CD (P<0.05)	0.37	0.16	0.34	0.13	0.71	0.03	-

Values are mean ± SE of three independent observations

blanched pearl millet based instant *Upma* mix was liked very much by the panelists. Balasubramanium *et al*. (2014) developed pearl millet *Upma* mixes, which scored in the range of 6.7-8.1 for overall acceptability.

Data in respect of proximate composition and gluten content of instant *Upma* mix is presented in Table 3. Findings of the present study are similar to the results reported by Balasubramanium *et al*. (2014) regarding the proximate composition of pearl millet based instant *Upma* mix developed by them. Significant (P<0.05) differences were observed in moisture content of unprocessed, extruded and germinated pearl millet based instant *Upma* mix. Poonam (2002) also reported that moisture content of pearl millet decreased significantly during various heat treatments like extrusion processing. In the present investigation maximum amount of crude protein was present in germinated pearl millet based instant *Upma* mix. The possible reason for increase in protein content in germinated pearl millet based instant *Upma* mix might be due to protein synthesis. During germination, degradation of storage protein takes place for development of seed embryo but at the same time, synthesis of new protein and other nutrients take place. Germination may be a desirable processing technique to increase the protein content of millet grain (Fasasi, 2009). All processing techniques resulted in significant (P<0.05) reduction in fat content. Non-significant differences were observed in crude fiber content of developed processed and unprocessed pearl millet based instant *Upma* mix.

Therefore, it is concluded that pearl millet has the potential to cater increasing needs and demands of gluten free convenience foods along with providing convenience in acquisition, storage, preparation and consumption of foods attributing to the busy lifestyle, increased women employment away from homes and several other factors. Instant *Upma* mix can serve as a low cost gluten free alternative, which can easily be developed at household as well as commercial level. It is remarkable that despite the grain being an ancient food, research on pearl millet and its food value is in its infancy and its potential vastly untapped. Research results so far are promising, showing the grain to have great aptitude and versatility and more and more uses for millet are being discovered every year, including its potential benefits.

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