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Contents

REVIEW ARTICLE

ROLE OF SULPHUR IN OILSEED CROPS: A REVIEW

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Abstract: India is largest producer and consumer of vegetable oil in the world. Oilseeds have been the backbone of agricultural economy of India since long. Oilseed crops are primarily grown for edible oil. Sulphur is an important nutrient for the higher yield and quality of oilseed crops. Sulphur plays a remarkable role in protein metabolism. It is required for the synthesis of proteins, vitamins and chlorophyll and also sulphur containing amino acids such as cystine, cysteine and methionine which are essential components of protein. Among the sulphur supplying sources, gypsum and elemental sulphur are being abundantly used in sulphur deficient soils.Use of high analysis sulphur free fertilizers, heavy sulphur removal by the crops under intensive cultivation and neglect of sulphur replenishment contributed to widespread sulphur deficiencies in arable soils. Due to Sulphur Deficiency, plants leaves becomepale green, yellowish green or complete yellowcolouration.The leaves are smaller and narrower and stem growth is affected.

Keywords: Sulphur, Growth, Oilseed, Quality

INTRODUCTION

India is one of the major oilseed grower and importer of edible oils. India's vegetable oil economy is world's fourth largest economy after USA, China & Brazil. The oilseed accounts for 13% of Gross Cropped Area, 3% of the Gross National Product and 10% value of all agricultural commodities. In India, oilseeds are cultivated over an area of 24.65 million hectares with a production of 31.31 million tones (Anon, 2018). Gujarat ranks third in the production of oilseeds in India whereas Madhya Pradesh tops the list and Rajasthan is in second position.The diverse agro-ecological conditions in the country are favourable for growing annual oilseed crops, which include 7 edible oilseeds (groundnut, rapeseed & mustard, soybean, sunflower, sesame, safflower and niger) and two non-edible oilseeds (castor and linseed) (Table 1).

Oilseed crops generally are one of the most important crops in the world. Their role in human diet and industrial application cannot be underestimated. The major oilseed crops include soyabean,coconut, oil palm, sesame, rapeseed, sunflower, safflower, olive seed, *etc*. The byproducts (hull, meal and oil) of oilseed crops had been integrated into human and animal diets due to its nutrient compositions. Majority of the oilseed meal consists of proteins and high contents of essential amino acid which are beneficial to human health and wellbeing. Likewise, the fat yields of oilseed crops are generally high, though varied from crop to crop (specie) and methods of extraction; high polyunsaturated fatty acids contents also prevent against coronary heart disease. Apart from the food value of oilseeds, several industrial products such as biodiesel, fertilizer, medicine, cosmetics, animal feeds, fibers, paint, button *etc*. have also been reported. Oilseed crops are promising crop with high potentials to improve human diets, prevent malnutrition and food insecurity and to provide employment through income generation in the society. Several researches had been carried out on the extraction of oil from oilseeds using different methods, production of improved breed of oilseed and detoxification of oilseed meals.It is known that one of the essential nutrients in human consumption is oil or fat, derived from the plant or animal sources. However, there is a limited increase in animal fat production. Therefore, in order to meet the increasing need in oil production is bound to the improvement and growth in oil plants. Oil is important due to the fact that not only it isan energy source in human consumption but also it is essential in the use of A, D, E, K vitamins and it contains oleic, linoleic (78%) fatty acids. Protein, oil, fiber and carbohydrate composition and energy value of different oilseed crops are given in table 2(Singh, 1999).

Sulphur (S) ranks 13th in terms of abundance in the earth's crust and is the fourth major plant nutrient after Nitrogen (N), Phosphorus (P), and Potassium (K). Generally, the oilseed crops require as much S as they need P. Sulphur is used as soil amendment for amelioration, as plant nutrient for increasing yield and quality of crop produce, as chemical agent to acidulate other nutrient and as pesticides (Kanwar and Mudahar, 1986). Sulphur plays an important role in the nutrition of oil seed crop and it act as a constituent of sulphur containing amino acids cystine, cysteine and methionine. (Parmar *et al.,* 2018 and Gangadhara, 1990). They considered that the oil seeds require more sulphur than other crops; its concentration and uptake vary with the availability of sulphur in soil. The overall requirement of sulphur for oil seed crops is as high as phosphorus (Aulakh and Pasricha, 1988). In oil seed crops, it is also involved in the formation of glucocides or glucosinolates which on hydrolysis

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increase the oil content. One of the main functions of sulphur in proteins or polypeptides is the formation of disulphide bonds between polypeptide chains. Disulphide linkages are important in stabilizing and determining the configuration of proteins. The application of sulphur increased the uptake of various macro and micro nutrients in groundnut (Singh, 1999). Sulphur deficiencies in soil and plant are recognized as a wide spread problem. Crop grown in sulphur deficient soil have poor utilization of N, P, K and a significant reduction in sulphur content in plants. Sulphur is also essential for the formation of chlorophyll and improves root growth besides this is involved in the formation of vitamins and enzymes required for the plant to conduct its biochemical processes (Jat*et al.*,2017 and Scherer *et al.,* 2008). Ultimately it only reduces the grain yield and quality of produce but also make a sharp impact in agro-based economy (Patil *et al.,* 2014 and Fismesa*et al.,* 2000). Sulphur can be applied to the soil through any suitable sulphur carriers *viz.* gypsum, elemental sulphur, ammonium sulphate and potassium sulphate *etc.* The choice depending on crop, local availability, price and need for other nutrients. Among the sulphur supplying sources, gypsum and elemental sulphur are being abundantly used in sulphur deficient soil.

Physiology and biochemistry of sulphur nutrition

The importance of S as plant nutrient has been recognized since the middle of the last century. Plants satisfy their S requirements from soil, atmosphere, irrigation water, and application of S containing fertilizers. An adequate supply of S to plant can improve yield, quality, and efficiency of other nutrients utilization besides reducing the incidence of diseases and is as important as phosphorus for oilseed crop. The functions of S, their requirement, yield responses and quality improvement and deficiency symptoms are discussed here.

Sulphur requirement

The oilseeds require more amount of S for its growth and development than other crops. The crops require S to make specific amino acids and various metabolites containing S, protein synthesis and process required for efficiency from other inputs. Being comparatively a drought tolerant with low transpiration, the oilseeds are susceptible to nutritional disorders especially S due to insufficient supply. The S requirement of oilseed crops in the field has been worked out by several workers (Banu *et al.,* 2017; Tandon 1991a, 1991b; Aulakhand Pasricha, 1988; Pasricha *et al.,* 1988 and Kanwar *et al.,* 1983).Based on the number of published data,Aulakh and Pasricha (1988) and Dhage*et al.* (2014) reported that the S uptake ranged from 5 to 20 kg t^{-1} of oilseeds and ranked the S requirement of oilseeds crucifers>sesame= sunflower>legumes > linseed. Sulphur requirement of oilseeds can be met through a number of S-containing materials such as gypsum, elemental S, pyrite and phosphogypsum. Generally, application of 30-40 kg S ha^{-1} to groundnut was more beneficial (Patel *et al.,* 2018;Kale, 1993 and Patra *et al.,* 1995).Application of S increased 4.5 to 7.5 kg groundnut yield per kg ha^{-1} of nutrient applied (Singh and Chaudhari, 1997).

Functions of Sulphur

The sulphur accounts for 0.1-0.5% dry wt. of the oilseeds where it is present in both organic and inorganic form. The sulphate uptake is slightly lower than phosphate. The S is mainly taken up by plant through roots as sulphate (SO_4^{-2}), but S also can be absorbed by leaves as SO_2 gas from the atmosphere. However, this gaseous S has to be subsequently transformed into the sulphate. In the chloroplast, the sulphate is first reduced to sulphide and thenincorporated into cysteine. A major portion of cysteine S is transferred to methionine, and bulk of these two is incorporated into proteins, where cysteine is responsible for secondary structure. The sulphide which is not incorporated into protein convert back to sulphate and stored in the leaves and to a lesser degree, in the seeds and can be mobilized whenever necessary. The S is required for the synthesis of protein, oils and vitamins. About 90% of the reduced S is required for the protein as it is constituent of methionine (21% S), cysteine (26% S), cystine (27% S). Some 50% of the total sulphur content of proteins are in methionine. Cystine is formed by the oxidation of two molecules of cysteine. Sulphur is also important for chloroplast and chlorophyll synthesis owing to its role in protein synthesis and important enzymes and redox reactions. Sulphur is also associated with flowering, nodulation and the quality of oilseeds and especially of groundnut crop. The S deficiency disrupt nitrogen metabolism, reduce protein quality, and induces carbohydrates accumulation.

Effect of Sulphur on crop quality and yield

The economic value of oilseeds is determined by its yield and quality, which are the resultant in part of the grower's ability to exploit the plant genetic make-up and part of less tractable components of the environment in which it is growing.Sulphur is involved in oil synthesis, and oil storage organs which are rich in S.The composition of oil and meals of most of the oilseeds has received greater attention from quality point of view.The oil in low erucic acid is nutritionally more desirable for human consumption. Low glucosinolate meals are acceptable for feed for livestock.The S is a part of oil compounds and its application increases the oil content in most of the oilseeds. On an average 5% increase in oil content has been observed with S fertilization (Naiknawar*et al.,* 2015 and Pasricha *et al.,* 1987).Effect of sulphur application on cell division, enlargement and elongation resulting in overall improvement in plant organ associated with faster and uniform vegetative growth of the crop (Noman *et al.,* 2015). Patel *et al.* (2018) reported that

the application of sulphur increase the protein content, oil content, haulm yield and pod yield in groundnut at Anand, India (Table 3)(Patel *et al.*, 2018).

Deficiency symptoms of sulphur

The S deficiency causes production of pale green, yellowish green or complete yellowcolouration of the leaves (Fig. 1).The S deficiency resembles those of nitrogen, but appear first on younger leaves due to its lesser mobility in plant. The S stored in the older leaves as sulphate is easily mobilized and transferred to growing organ to a certain extent, but not the one that has already been incorporated into organic compound. However, the S mobilized from older leaves is not sufficient to maintain the normal growth, as a result the youngest leaves remain small and more or less yellow owing to lack of protein and chlorophyll. The S content of older leaves is thus somewhat higher than the young leaves. The S deficient plants are stunted and have a rigid erect appearance. The leaves are smaller and narrower, stem growth is affected. As the oilseeds has very high requirement for S, the S-deficiency in oilseeds can reduce the yields from 20 to 40% even without or mild visible symptoms. The visible symptoms are the indication of severe stress of S-deficiency and fertilization is must to sustain the production. The deficiency symptoms available for individual oilseeds are given in table 4.

Determination of Sulphur Deficiency

Soil Analysis: A number of chemical methods have been developed and tested for estimating the available sulphur status of soils. The important thing is the selected method should be accurate, precise, rapid and highly correlated with crop response to sulphur application. The most often used method in India and some other places involve extraction of soil sulphur with 0.15 percent solution of $CaCl_2$. Soils containing less than 10 ppm sulphur are considered to be low or deficient in plant available sulphur by this method (Williams and Stainbergs,1959). Globally, monocalcium phosphate is also a popular extractant.

Plant Analysis: Plant analysis is carried out by standard analytical methods. Normally, if leaves of cereal plants contain less than 0.2% sulphur are considered to be deficient in sulphur and require sulphur application for optimal growth and yield production. S is estimated in plant digest obtained by dry ashingby wet digestion with HNO_3 and $HClO_4$(Chaudhary and Cornfield, 1966).

Sources of sulphur useful for various oilseed crops

There are now many sources of S fertilizers, but the comparative response of various S sources on oilseeds has been studied by a few workers (Banu *et al.*, 2013; Bandopadhyay and Samui, 2000; Chaubey*et al.*, 2000 andDutta and Patra, 2005). The most commonly used S fertilizers are ammonium sulphate (24% S), single superphosphate (12% S), Gypsum (13% S), pyrite (22-30% S), elemental S (85-100% S). The other S sources are phosphogypsum (16% S), potassium sulphate (18% S), magnesium sulphate (13% S), pressmud (2-3% S), sulphur sludge (10-16% S). The S fertilizers containing SO_4^{-2}-S were effective for most of the oilseed crops, (Tandon, 1991a). The gypsum is reported to be efficient for groundnut and mustard.

As the elemental S require oxidation before its uptake by plants hence should be applied 20-25 days before planting. The efficacy of elemental S increases with increase in soil pH but not in soil with high sodium. Pyrite is also a slow-release fertilizer (Tiwari *et al.*, 1984) and suitable for calcareous and alkaline soils. It also requires application 20 to 25 days before planting in moist soil. The gypsum and phosphogypsum are suitable for both alkaline and acid soils but more suitable for crop requiring high Ca such as groundnut (Rao *et al.*, 2013). The ammonium sulphate is most suited for non-nitrogen fixing oilseed crops and corrects both the N and S deficiencies. The singlesuperphosphate is a good source of S and P and required for integrated P and S application in most of the oilseeds.

How to apply the S fertilizers?

Broadcast or dribble banded, broadcast and subsequent incorporation, band placement, seed placement and banded near the seed are the major methods of application of sulphur fertilizers in soils. Among these methods broadcasting and band placement methods are widely used.

Managing sulphur fertilization in soil using 4R nutrient stewardship principle (Right Source of nutrient applied at the Right Rate, Right Time, and Right Place) is a modern tool to enhance the use efficiency of S-fertilizers.

Scientific management practices can be adopted to avoid the losses of the nutrient element S. A combination of soluble SO_4^{-2} and elemental S may be useful to provide both an immediate and a prolonged source of plant nutrition.

Table 1. Common Oilseed crops grown in India**.**

Common name	Botanical name	Others name
Groundnut	*Arachis hypogaea*	Moongphali, Chinabadam, Bhoising, Kadalcayi, Bhu mung, Verusanaga.
Rapeseed-Mustard	*Brassica compestris (Sarson)* *B. juncea (Raya)*	Rai, Sorse, Sasuve, Aasur, Kadugu, Mohori, Sorisa,
Niger	*Guizotiaabyssinica*	Kala til, Ramtil, Gurellu, Karale, Kattelu, Valasulu

Sunflower	*Helianthus annuuas*	Surya mukhi, Suraj mukhi, Surya kantihi
Sesamum	*Sesamum indicum*	Til, Acchellu, Rasi, Nuvvulu
Safflower	*Carthamumtinctoris*	Kardi, Kushmaginzalu
Soyabean	*Glycin max*	Bhatmas, Garikalai, Muth
Caster	*Ricinus cummunis*	Aranda, Erandi, Rendi, Haralu
Linseed	*Linumusitatissimum*	Tichi, Tishi, Java, Alsi, Seeme, Pesi

Table 2. Composition and energy value of oilseeds.

Composition (%)						**Energy(Cal/100 g kernel)**
Oilseeds	**Moisture**	**Protein**	**Oil**	**Fiber**	**Carbohydrate**	
Groundnut	5.0	28.5	47.5	2.8	13.3	595
Mustard	8.5	20.0	39.7	1.8	23.8	541
Niger	4.2	23.9	39.0	10.9	17.1	515
Sunflower	5.5	19.8	52.1	1.0	17.9	620
Sesamum	5.3	18.3	43.3	2.9	25.0	563
Safflower	5.5	13.5	25.6	34.9	17.9	356
Soyabean	8.1	43.2	19.5	3.7	20.9	432

Table 3. Effect of sulphur levels on protein content, oil content, haulm yield and pod yield of groundnut**.**

Sulphur level	**Protein content (%)**	**Oil content (%)**	**Haulm yield (kg ha^{-1})**	**Pod yield (kg ha^{-1})**
0 kg S ha^{-1}	27.00	41.76	3704	1745
20 kg S ha^{-1}	27.29	43.08	4141	1956
40 kg S ha^{-1}	27.48	43.75	4349	2083

Table 4. Deficiency symptoms of major oilseed crops**.**

Crops	**Deficiency Symptoms**
Rapeseed-Mustard	Deficiency symptoms first occur on younger leaves. Chlorosis of leaf margins, development of purple pigmentation. Inward curling of young leaf lamina giving rise to a cupped appearance followed by scorching and withering.
Groundnut	Stunted growth, and general yellowing of plants. Delay in maturity. Acute sulphur deficiency causes the entire plant turn to yellow.
Sunflower	Yellowing spreads from the base to the apex. Growth of plants is reduced. The size of capitulum is severely restricted. Inflorescence may remain covered within the bracts. Maturity of flowers is often delayed.
Sesame	Growth is retarded, leaves are smaller and fully emerged leaves first turn pale and then golden yellow. Number of flowers and pods are reduced; hence yield is reduced.
Canola	Younger leaves are lime-green, often with interveinal chlorotic mottles and pale leaf margins. Leaves are cupped or roll inwards and become thickened and crisp and brittle.

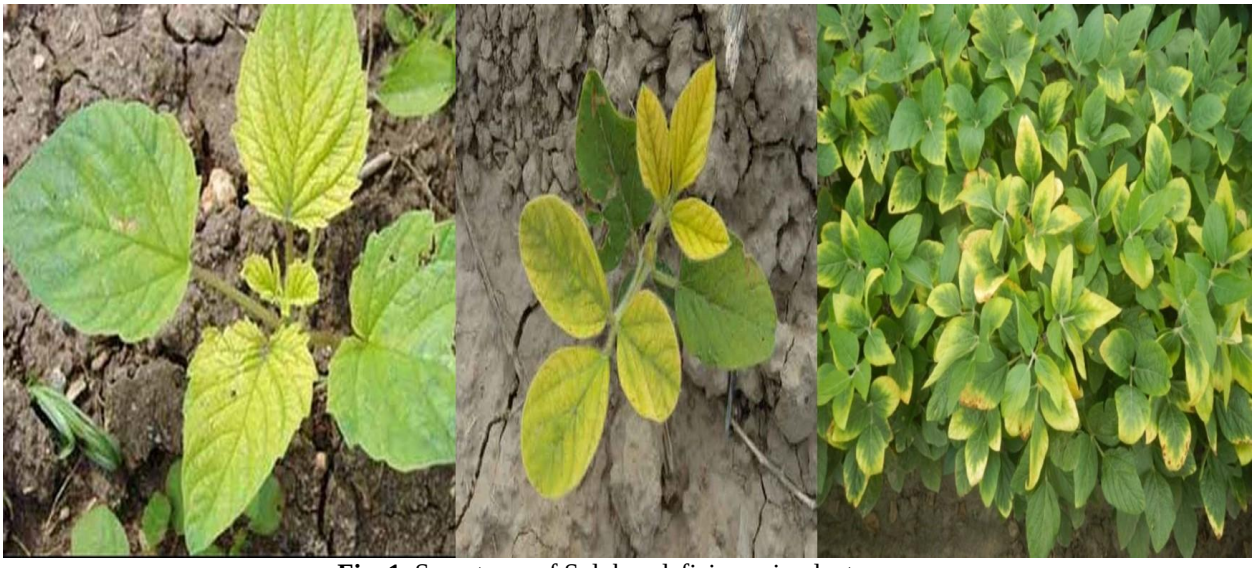

Fig. 1. Symptoms of Sulphur deficiency in plants

CONCLUSION

The application of sulphur @ 30 to 40 kg ha^{-1} significantly influence the growth, yield, nutrient uptake and economics of oilseed crops in different agro climatic zone of the country. Sulphur applications has pronounced effect on quality parameters of oilseeds.

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REVIEW ARTICLE

SOME MEDICINAL PLANTS USED FOR NERVOUS DISORDERS: A REVIEW

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Abstract: Use of plants for curing human ailments is an ancient practice. Recently there is revival of interest. Ethno botanical field surveys have been done from different parts of developing countries of the world. It reflects concern about the possible loss of valuable information on traditional medicine. Neurological disorders are often not considered common diseases. They are mental illness like epilepsy which is the most serious chronic disorder affecting millions of people. Other's like Parkinson's, Alzhemirs, Meningitis and Stroke. Nervous disorders also affect speaking, movement, breathing, mood and memory. Herbal medicines are a holistic medium. Growing of these important herbs will add to the terrestrial diversity of the ecosystem and help in conservation of Biodiversity. *Centella asiatica, Avena sativa, Lagenaria sicerana, Cassia tora, Cassia fistula* are some of the important plants used in nervous disorders. The different medicinal plant varieties can be studied with biochemical properties and a taxonomic classification can be made based on medicinal uses and on the biochemical relationship drawn. Tissue Culture studies along with molecular characterization can also be done. Important germplasm of the medicinal plants will add to the terrestrial biodiversity and the most effective medicinal plant used for nervous disorder can be obtained.

Keywords: Nervous disorders, Medicinal plants, Biodiversity, Ethnobotany, Mental illness

INTRODUCTION

Most of the population depends on traditional medicine for primary health care, however, neurological disorders are often not considered as common diseases and many people with mental illnesses, like epilepsy, are severely affected by health related stigma and discrimination. Epilepsy is the most common serious chronic brain disorder, estimated to affect at least 10 million people.

Others like Parkinson's, Alzhemirs, Meningitis and Stroke. Nervous disorders also affect speaking, movement, breathing, mood and memory. Neurological disorders affect the brain and spinal chord.

Ethnomedicine and Ethno pharmacology can bring promising results capable of adding value to the very rich natural resources of the country.

Taking into account the existing knowledge on the medicinal properties of plants for treatment of neurologic disorders, it is believed that research in the areas of ethnomedicine and ethnopharmacology is required.

Medicinal value present in tissues produces physiological action on body. Alkaloids (in the form of C,H,O,N), Glucosides, essential oils, fatty oils, mucilages, tannins, gums are present in the plants.

Use of plants for curing human ailments is an ancient practice.

Recently there is a revival of interest. It reflects concern about possible loss of valuable information on traditional medicine.

Most population depends on traditional medicine for primary health care, so study of medicinal herbs is essential (Sinha[5], 1997).

Plants used in nervous disorders:

1. Nervous disorders: *Avena sativa, Aegle marmelos, Acorus calamus*
2. Hypochondria: *Aegle marmelos*
3. Epilepsy: *Datura metel, Emblica officinalis, Evolvulus alsinoides, Ferula asafoetida*
4. Melancholia: *Aegle marmelos*
5. Hysteria: *Valeriana*
6. Mania: *Datura*
7. Depression: *Annona squamosa*
8. Insanity: *Datura metel. Bacopa monieri, Anacardium occidentale*
9. Sedative: *Annona muricata*
10. Anxiety: *Bacopa monieri,Rosmarinus officinalis*
11. Hypnotic: *Annona muricata*
12. Insomnia: *Biophytum*
13. Hysteria: *Cassia occidentalis*
14. Epilepsy: *Cassia fistula*
15. Narcotic: *Papaver somniferum*
16. Paralysis: *Strychnos nux vomica*
17. Hypnotic: *Hyacyamus niger*
18. Stimulant: *Panax ginseng*
19. Neuralgia: *Aconitum napellus*
20. Increases brain power: *Loranthus longifera*
21. Anxiety: *Rauwolfia serpentine*

Parts of plants used for medicinal purpose:

Datura metel: root
Papaver somniferum: dried juice, latex of unripe capsule.
Strychnos nux vomica: seeds
Hyoscyamus niger: leaves, flower tops, seeds.
Valeriana: essential oil, root.
Panax ginseng: root
Bacopa monnieri: leaf
Ferula asafoetida: root
Aconitum: seeds

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REVIEW:

***Rauwolfia serpentina*:**

In *Rauwolfia* the root is used in humans to treat hypertension, insanity. It is also used for relief of central nervous disorder including anxiety and excitement (Erheni A. H., Obadoni B. O., 2015). It is used for insomnia, mental disorders, and aggressive behavior. It calms the central nervous system and reduces anxiety, irritability and aggression. It can be used for the treatment of schizophrenia, epilepsy, psychosis and other mental disorders (Jagdev Singh[17], 2015).

Aegle marmelos:

In *Aegle marmelos* various studies have shown presence of flavonoids in phytochemical screening which are responsible for anxiolytic effect through benzodiazepine receptors. Therefore, flavonoids present in *Aegle marmelos* may be responsible for the anti–anxiety activity. Various studies on *Aegle marmelos* have shown presence of phyto constituents other than flavonoids like tannic acid, phenols, marmesinin, ascorbic acid, eugenol, skimmianine and saponin etc which may possess anxiolytic properties. *Aegle marmelos* can be a safe and effective drug for the treatment of number of anxiety disorders. The fruit contains ethanolic extracts. These are used to care fatigue, anxiety, depression. The fruit has steroids, coumarin and alkaloids.

Rosmarinus officinalis:

Rosmarinus officinalis L. has several therapeutic applications in folk medicine in curing or managing a wide range of diseases including depression. The extract of *R. officinalis* produced an antidepressant like effect, since the acute treatment of mice with the extract reduced the immobility time swimmining test and tail suspension test in mice as compared to a control. The results suggest that the anti depressant action of *R.officinalis* is mediated by an interaction with the monoaminergic system and that this plant should be further investigated as an alternative therapeutic approach for the treatment of depression (Daniele G Machado[15], 2009). Rosemary diterpenes have been shown in recent years to inhibit neuronal cell death induced by a variety of agents both in vitro and in vivo. The multifunctional nature of the compounds from the general antioxidant-mediated neuronal protection to other specific mechanisms including brain inflammation and amyloid beta formation is discussed (Solomon Habtemarian[34], 2016).

***Evolvulus alsinoides*:**

Bioactivity guided purification of n-BuoH solube fraction from two new compounds, 2,3,4-trihydroxy3methylbutyl 3-2 propeonate and 1,3-di-O-caffeoyl quinic acid methyl ester along with 6 known compounds, caffeic acid, 6-methoxy-7-O-beta-glucopyranoside coumarine, 2-C-methyl erythritol, Kaemferol-7-O-beta-glucopyranoside.The structure of new compounds were elucidated by spectroscopic analysis ,while known compounds were confirmed by direct comparison of their NMR data with those reported in literature. This is the first report of the presence of phenolic constituents in *Evolvulus alsinoides* (Prasoon Gupta[13], 2007). *Evolvulus* is effective nootropic agent. It is mainly indicated in loss of memory, sleeplessness, treatment of epilepsy (Anupama[35], 2016). The isolated compounds were screened for anti stress activity in acute stress induced biochemical changes in adult male Sprague-Dawley rats. Stress exposure has resulted in significant increase of plasma glucose, adrenal gland weight, plasma creatine kinase and corticosterone levels. The compounds displayed most promosing antistress effect by normalizing hyperglycemia, plasma corticosterone and adrenal hypertrophy.

***Avena sativa*:**

Avena sativa is mainly used for spasmodic and nervous disorders with exhaustion. Cardiac weaknesses, spermatorrhea problem, the nervous debility of convalescence are common symptoms of homeopathic *Avena sativa* (Shastho Totho[11]). In male function neurasthenia, homeopathic *Avena sativa* has a selective influence upon the nerve system of the genitor-urinary apparatus. Because of its selective power upon the total nervous structure which supplies the reproductive organs. Nervous palpitation of the heart, insomnia, nervous excitement and mental weakness or failure and general debility caused by masturbation can be easily removed using this remedy.

***Datura metel*:**

Producing and selecting interspecific hybrids of *Datura* for high scopolamine production was successfully done. The leaves of *Datura metel* contain 0.2-0.5% tropane alkaloids, the flowers 0.1-1.0% and the seeds 0.2-0.5%. Scopolamine is major constituent in mature leaves. Other alkaloids are hyoscyamine, norhyoscyamine, norscopolamine, hydroxyl-6-hyoscyamine and metelodine. They increase the heart rate, induce relaxation and motor inhibition in smooth muscles, decreased secretions and induce dilation of the pupils of the eyes. In vitro production of scopolamine and hyoscyamine is feasible though uneconomical. Cultures of hairy roots of *Datura metel* are the most productive (Plant Resouces of Tropical Africa).

***Annona*:**

Some neuropharmacological are there in effects of the ethanol extract of the leaves of *Annona diversifolia.* Intraperitoneal administration of the extract delayed the onset of clonic seizures induced by petylenetetrazole and delayed the time in the Rota-red and swimmimng test. In addition the extract augmented the duration of sleeping time induced by sodium pentobarbital. These results indicate that the ethanol extract of the leaves of *A. diversifoloia* has depressant activity on the central nervous system (M E Gonza lez Trujano[19], 1998).

***Acoros calamus*:**

Chewing the rootstock of *Acorus calamus* plant can cause visual hallucinations, possibly because of the presence of alpha-asarone or beta-asarone. *Acorus calamus* shows neuroprotective effect against stroke and chemically induced neurodegenaration in rats. Specifically,it has protective effect against acrylamide-induced neurotoxicity. Both roots and leaves of *Acorus calamus* have shown antioxidant properties. *Acorus calamus* roots and rhizomes have been used in Indian system of traditional medicine for hundreds of years and it is highly valued as a rejuvenator for the brain and nervous system. *Acorus calamus* rhizome constituents, particularly alpha and beta asarone possess a wide range of pharmacological activities such as sedative, CNS depressant, behavior modifying, anticonvulsant, acetyl cholinesterase inhibitory and memory enhancing (Jina Pattanaik[25], 2013).

***Bacopa monnieri*:**

Several studies have suggested that *Bacopa monnieri* extracts have protective effects in animal models of neurodegeneration. The herbal supplement and extract has effect on memory, anxiety and brain health. It is also used for epilepsy, nootropic substances, Alzheimer's disease and memory improvement. It helps in anxiety reduction, attention deficit hyperactivity disorders. The whole plant standardized dry extract has role on cognitive function and affects its safety and tolerability in healthy elderly study participants. The study provides further evidence that it has potential for safely enhancing cognitive performance in the aging (Carlo Calabrese[36], 2008).

***Ferula asafoetida*:**

The oleo gum resin of *Ferula asafoetida* has recently found to have neuroprotective properties in animal models and humans (Asma K[16], 2015). *Asafoetida* has been used as a sedative and stimulant. It is widely used in Indian system of medicine like Ayurveda. *Asafoetida* has been held in great esteem among indigenous medicines, particularly in Unani system (Poonam Mahendra[28], 2012).

***Embilica officinalis*:**

Embilica officinalis is helpful in the following health conditions: Memory loss, mental fatigue, anxiety with mental irritability and restlessness, depression with aggressive reactions, attention deficit hyperactivity disorder (Jagdev Singh[17], 2015). Amla is helpful in following health conditions; Brain and nerves-headache with burning sensation, migraine with pulsing and throbbing pain, memory loss, mental fatigue, vertigo. Psychological diseases-anxiety with mental irritability and restlessness, depression with aggressive reactions, insomnia, violent mental agitation (Jagdev Singh[17], 2015).

***Valeriana*:**

Valerian extract can cause sedation by increasing brain's GABA level. GABA is an inhibitory neurotransmitter, and in large enough quantities it can cause a sedative effect. Results from an in vitro study suggest that valerian extract may cause GABA to be released from brain nerve endings and then block GABA from being taken back into nerve cells. In addition Valeran's valerenic acid inhibits an enzyme that destroys GABA another way that valerian can improve your GABA levels and promote a great night's rest. Scientists have found that valerian root increases the amount of a chemical called gamma aminobutyric acid (GABA) in the brain. GABA helps regulate nerve cells and calms anxiety. Drugs such as alpazolam and diazepam also work by increasing the amount of GABA in the brain. The valerenic acid and valerenol contained in valerian root extract act as anti-anxiety agents. It's pretty amazing that an herbal remedy like Valerian root can have the same anti-anxiety effects of prescription drugs without all the serious side effects of psychotropic drugs. Valeriana root have sedative and anxiolytic effects.

***Cassia*:**

Study evaluated the effect of *Cassia fistula* on sleeping time and level of anxiety in male albino mice. The aqueous extract of fruit increased sleeping time and decreased levels of anxiety in mice. Investigations have revealed several biological activities such as antidepressant activities of *Cassia occidentalis*. (Manikandaselvi V[21], 2016). Leaf poultices of Cassia fistula are also used for fascial massage in affections of the brain and applied externally in paralysis, rheumatism and gout (Rajan Singh[27], 2016).

Papaver somniferum:

Papaver somniferum is the species of plant from which opium and poppy seeds are derived. It is the source of natural and semi synthetic narcotics. It is the source of several pharmaceutical benzylisoquinoline alkaloids including morphine, codeine and sanguinarine. The hairy root cultures accumulated three times more codeine than intact roots. Narcotics are used therapeutically to treat pain but they alter mood and behavior significantly (Da Cheng Haeo[30], 2015).

***Strychnos nux vomica*:**

Nux vomica is a plant. The seed is used to make medicine. It is used for nerve conditions and depression. *Nux vomica* dried seeds contains two principles alkaloids-Strychnia and Brucia. It is useful for people doing mental work or under stress (Manisha Bhatia, 2010).

***Hyoscyamus niger*:**

The Application areas of *Hyocyamus niger* are epilepsy, meningitis and dementia. *Hyoscyamus* is a remedy with some common mental and emotional themes running through all its various expressions (David A Johnson[31], 2009).

***Panax ginseng*:**

The root of *Panax ginseng* has been a popular medicine. Ginsenosides are neuroprotective. This review considers publications dealing with the various actions of *P.ginseng* that are indicative of

possible neurotherapeutic efficacies in neurodegenerative diseases and neurological disorders such as Parkinsons disease, Alzhemirs disease, Huntingtons disease and amyotrophic lateral sclerosis and multiple sclerosis (I K Hyun Cho[22],2012). Ginseng has been used as a traditional modern medicine for over 2000 years and is recorded to have antianxiety, antidepressant and cognition enhancing properties. The molecular mechanisms of the neuroprotective effects of ginseng in Alzhemirs disease including beta amyloid formation, major depression and Parkinson's disease is discussed (Wei-Yi-Oug[32], 2015).

***Aconitum*:**

Aconite is one of the best remedies for waves of fear or outright panic. It is wild in alpine Himalayas of Kashmir and Nepal at an altitude of about 3600m. Root is used for nervous disorders, neuralgins, dropsy and as sedative. Pure roots contain the alkaloids pseudaconitinine, chasmaconitine, indaconitine and bikhaconitine. The efficacy of the drug is based on the di-ester alkaloids - aconitin, mesaconitin and hypaconitin (C P Khare).

***Loranthus longifolia*:**

Loranthus longifolia protects central nervous system against electromagnetic radiation on rat. It has been widely used for the treatment of brain diseases, particularly in South West China. Hence, the present neuroprotection model was designed to investigate its neuroprotective properties against hydrogen peroxide induced oxidative stress in NG-108-15 cells. The aqueous extract exerts marked neuroprotective activity (Daniel Zin Hua[29], 2012).

DISCUSSION

Several medicinal plants have been identified having properties for dealing with nervous disorders. *Aegle marmelos* can be a safe and effective drug for anxiolytic effects. The fruit contains the ethanolic extracts. The fruit has steroids and alkaloids. There is need to further study the antidepressant action of *Rosmarinus officinalis*, the rosemary diterpenes have shown to inhibit neuronal cell death (Daniele G Machado[15], 2009). Compounds have been reported from *Evolvulus alsinoides* which are most promising in antistress effect by normalizing hyperglycemia, plasma corticosterone and adrenal hypertrophy (Anupama[35], 2016). Hybrids of *Datura metel* have high scopolamine production which increases heart rate and induces relaxation in muscles and the in vitro production of scopolamine is feasible, as hairy roots of *Datura metel* are the most productive.

Further work on *Acorus calamus* needs to be done as leaves which show antioxidant properties and has neuroprotective effect (Jina Pattanaik[25], 2013). The whole plant dry extract of *Bacopa monnieri* has a role on cognitive function and is tolerable in elderly patients (Carlo Calabrese[36], 2008). *Ferula asafoetida* is neuroprotective and an indigenous medicine in Unani and Ayurveda (Poonam Mahendra[28], 2012). Which is used in household always? The molecular characterization work still needs to be done on *Embilica officinalis* which is helpful for violent mental agitation (Jagdev Singh[26], 2015). The GABA action of *Valeriana* roots without any side effects and used as a psychotropic drug. The important fact of *Papaver somniferum* for mood swings is that hairy root cultures have three times more codeine than intact roots.

Ginseng has been used for more than 2000 years and is useful as antianxiety, anti depressant and in Parkinson's disease. The pure roots of Aconite contain the alkaloids for fear and panic (I K Hyun Cho[22], 2012). *Loranthus* is neuroprotective against hydrogen peroxide induced oxidative stress (Daniel Zin Hua[29], 2012).

CONCLUSION

Certain genera like *Cassia* may be of value in conservation of drug plant resources. Further identification of medicinal plants through such classification might help in drug formulations, drug substitution and for systemizing our knowledge about medicinal plants. Family wise and disease wise break up of drug plants would systematize the survey and structure based grouping of useful plants. Geographical regions of the world likely to yield useful information on disease incidence/distribution and variation of the pattern of plant use, diversity of a plant species is proportional to its medicinal use. Medicinal value present in tissues produces physiological action on body. Alkaloids and glucosides can be use (kokate[2], 2008). Different plant varieties to be studied with biochemical parameters and a taxonomic classification can be made based on medicinal uses and on the biochemical relationship drawn. Tissue culture studies and molecular characterization of all the species are to be done. Important germplasm of the above mentioned plants will add to the terrestrial biodiversity and the most effective medicinal plant used for nervous disorder can be obtained.

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RESEARCH ARTICLE

CHARACTERIZATION OF POTENTIAL PGPR'S ISOLATED FROM RHIZOSPHERE OF WHEAT FROM TRANS-HIMALAYAS AND THEIR EFFICACY ON SEED GERMINATION AND GROWTH PROMOTION OF WHEAT UNDER NET HOUSE CONDITIONS

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Abstract: In the present study, the diversity of rhizobacterial isolates from rhizosperic soils under wheat cultivation in districts of Solan and Sirmour of Himachal Pradesh a Himalayan belt of India. Phenotypic and genotypic characteristics of the PGPR isolates were recorded to categorize and identify the bacteria. In total seventy three rhizobacterial isolates were isolated from different locations of both the districts of which some sites were rainfed and some sites were irrigated. The characteristics of the bacterial isolates were determined using the colony morphology, gram staining as well as biochemical properties. After screening for PGP attributes *in-vitro* conditions. Three isolates (Kn-7, De-21 and Dh-7) were found hyperpotential for PGP attributes such as production of siderophore, P-solubilization, ammonia, HCN and growth regulators. These three isolates had shown maximum PGP potential *in- vitro* conditions and thus were selected to construct bioformulations for the wheat crop under net house conditions.

Keywords: Wheat, PGPR, Rhizosphere, PGP Attributes, Growth Promotion of wheat

INTRODUCTION

Wheat is a commercially important crop belonging to gramineae family. At present India is the second largest producer of wheat after China. The use of microorganisms in agriculture is at a low level despite the investment in scientific work. Microbial inoculants can be used as an alternative to chemical fertilizers in view of the damaging effect of pesticides, fungicides and insecticides. Plant Growth Promoting Rhizobacteria (PGPR) is such groups of bacteria that colonize the rhizosphere and improve plant growth. The use of PGPR can be used in the future to enhance agricultural production. PGPR's also played an important role in enhancing the root and shoot growth, and act as efficient microbial competitors in the root zone. Significant effects have been observed in wheat. The use of PGPR reduces soil borne pathogens and thus enhances plant growth. Himachal Pradesh is an important Himalayan state for wheat cultivation the state has unique pattern of terraced cropping system.

Agriculture status of Himachal Pradesh

Himachal Pradesh is situated in the north-western part of Himalaya. Most of the geographical area of the state comes under forest, pasture, and grazing land, agriculture is possible only on less than ten percent of the state's net area. The physiography and climatic condition in the state favours diversified potential for farming and allied activities. Due to the undulating terrain condition ranging from plains to high hills, mixed farming is predominant. Most of the farming activities are concentrated along the channels of major rivers and their tributaries. Different crops are being cultivated in the state. Among the cereals, wheat, rice, maize, and barley are important. The state also produces pulses and oilseeds. Cash crops are also becoming important, since fair amounts of potatoes, ginger, tea, and peas come from the state. Fruits, dry fruits, and a variety of vegetables are grown in the state Kant S (1995). Mechanisms that can promote plant growth include production of phytohormones, biological nitrogen fixation and increased solubility of insoluble elements in soil (Rovera *et al*., 2008). Interest in the beneficial rhizobacteria associated with cereals has increased recently and several studies clearly demonstrated the positive and beneficial effects of PGPR on growth and yield of different crops especially wheat at different environment under variable ecological conditions (Mehnaz *et al.*, 2010, Zhang *et al.*, 2012).

PGPR

PGPR are free living bacteria that resides in the rhizosphere region in the soil. They either directly or indirectly assist rooting. They play different roles in the soil which proves beneficial for plant health and productivity.

The mechanism by which PGPR exerts their beneficial effect on plants can be very diverse. They can establish themselves on root surface or inside the roots. PGPR can be classified into extracellular plant growth promoting rhizobacteria (ePGPR) that may exist in the rhizosphere, on the rhizoplane or in the spaces between the cells of root cortex. The bacterial general such as *Agrobacterium, Arthrobacter, Azotobacter, Azospirillum, Bacillus, Flavobacterium,*

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Pseudomonas and *Serratia* belong to ePGPR. The other category is intracellular plant growth promoting rhizobacteria (iPGPR) that locates generally inside the specialized nodular structures of root cells (Figueiredo *et al.*, 2011). It belongs to the family of *Rhizobiaceae* includes *Allorhizobium*, *Bradyrhizobium*, *Mesorhizobium* and *Rhizobium*, endophytes and *Frankia* species both of which can symbiotically fix atmospheric nitrogen with the higher plants.

PGPR are free living or symbiotic associated bacteria that reside in rhizospheric soil or intracellularly as endophytes. They play very important roles in the soil which proves beneficial for plant health and productivity. They colonize the rhizosphere and protect plants from its pathogens, by producing secondary metabolites such as antibiotics, volatile compounds that suppress harmful pathogenic bacteria and fungi by different mechanisms. PGPR's also produce siderophores (iron chelating compounds), and phytohormones (Auxins, Gibberellins and Cyatokinins), can fix atmospheric nitrogen, and help in providing nutrition uptake by solubilizing phosphate and produce biologically active substances which influence the plant growth and development (Mayak *et al.*,1999).

Keeping in mind the present study was planned to isolate the native strains from rhizosphere of wheat grown on different soils of Solan and Sirmour districts of Himachal Pradesh. These bacteria were characterized and screened *in vitro* for PGP potentials. Furthermore to evaluate the efficacy of selected strains of PGPR in seed germination and growth promotion of wheat under net house conditions.

MATERIALS AND METHODS

Sample Collection, isolation and purification of PGPR: Soil samples were collected from wheat rhizosphere from different locations of Solan (Kandaghat, Deothi, Dharja) and Sirmour (Rajgarh, Habban, Pulwahal) districts of H.P. Nitrogen-free medium (Jensen medium), Luria Bertani agar, King's B agar and Nutrient agar medium were used for isolation of PGPR by serial dilutions method followed by purification on the same solid media with a repeated plating method.

Colony morphology and pigment production

Colony morphology (form, elevation, and margin, and opacity, surface) and the production of pigment was checked on Nutrient agar medium at 28±2 ^{0} C.

Biochemical characterization

Biochemical characteristics of the purified PGPR's isolates like Gram reaction, catalase reactions, methyl red, Voges–Proskauer test, citrate utilization, casein hydrolysis (Subba Rao, 1977).

Bio assays for Plant Growth Promoting Attributes

P-Solubilization

For estimation of phosphate solubilizing capacity of PGPR isolates. Pikovskaya agar plates (Pikovskaya's, 1948) with known amount of inert phosphorus ($Ca_3(PO_4)_2$). Phosphate solubilization expressed in terms of mm diameter of yellow colored zone produced around well/bit at 28^0C after 72h.

Siderophore production

Siderophores production was detected by chrome azurol-S (CAS) plate assay method (Schwyn and Neilands, 1987). 25 ml of CAS dye was mixed with 250 ml of nutrient agar and mixed well before pouring. 100µl of 72 h old culture supernatant of each test bacteria was placed on prepoured chrome azurol-S agar (CAS) plates. Plates were incubated at 28^0 C for 72 h. Production of siderophore was expressed in terms of mm diameter of pinkish/orange halo zone produced around the well at 28^0 C in 72h.

Ammonia production

Ammonia production was checked according to Lata and Saxena (2003). PGPR's isolates were grown in peptone water (5 ml) in tubes. Tubes were incubated at 28^0 C for 4 days. 1ml of Nessler's reagent was added to each culture tube. Presence of faint yellowish to brown color (+) indicated small amount of ammonia and deep yellow (++) to brown color (++++) indicated large amount of ammonia production.

HCN production

PGPR's isolates were screened out for the production of hydrogen cyanide (HCN). (Bakker and Schippers, 1987) bacterial cultures were streaked on prepoured plates of nutrient agar medium amended with 1.4 g/l glycine. Whatman No.1 filter paper strip were soaked in 0.5 per cent picric acid followed by 2 per cent sodium carbonate and were placed in the lid of each petriplates. Petriplates were sealed with parafilm and were incubated at 28^0 C for four days. Uninoculated control with picric acid paper strips was kept for comparison of results. Plates observed for change of color of filter paper from yellow (-) to brown (+++) to dark brown (+++++). Intensity of color developed indicated as high production of volatile HCN.

Plant Inoculation and Root Colonization

Pot experiment in net house conditions

Pot experiment was conducted under the net house conditions. Plastic pots having (20 cm diameter, 20 cm deep) were used for this experiment containing 3 kg of sterilized soil. Fresh culture of each isolates was used for the each treatment. Total twenty five treatments were used for the study in which individual, consortia of each isolate and recommended dose of fertilizer were used in each treatment. Seeds were surface sterilized with 0.1% $HgCl_2$ for the prevention of surface fungal/bacterial contamination. Sterilized seeds were coated with bacteria by dipping the seeds of wheat in liquid bioformulation. The pot experiment contained control (no bacterial inoculation) and inoculation with bacterial culture in the form of individual and

consortia of isolates. Pots were incubated in net house conditions at temperature 16-20^{0}C (day/night) for 50 days and after a week germination percentage and shoot length were calculated as seedling growth parameters (Meena *et al.*, 2016). Three individual isolates (Kn-7, Dh-7 and De-23) and their consortia (Kn-7+ Dh-7 + De-23) along with different doses of fertilizer.

Estimation of chlorophyll content of leaves (Withem *et al.*, 1971)

Fresh leaves of wheat were collected from the field and were weighed 1gm of each treatment. Dip the leaves in 80% acetone in a test tube, keep it overnight as such and record the optical density on next day at two wavelengths i.e. A_{663} and A_{645}. Chlorophyll a, chlorophyll b and total chlorophyll in mg/g of tissue was calculated by formula,

mg of chlorophyll a/g tissue =12.70 (A_{663})-2.69(A_{645}) ×V/ w×1000

mg of chlorophyll b/g tissue = 22.9 (A_{645})- 4.68(A_{663}) ×V/w×1000

mg total chlorophyll /g tissue=20.0 (A_{645})+8.02 (A663)×V/w×100

Where, A= optical density

V=final volume of 80% acetone chlorophyll extract (10ml)

W= fresh weight in gm of tissue extract (1gm)

RESULTS

Isolates from soil samples

Total seventy three PGPR's isolates were isolated from different rhizosperic soil samples of wheat from different locations of Solan and Sirmour districts of Himachal Pradesh on specific nutrient media (Nitrogen-free medium (Jensen media), Luria Bertani agar, King's B agar and Nutrient agar medium). Out of total seventy three isolates of PGPR, twenty nine hyperpotential PGP rhizobacteria were selected for further studies after primary (qualitative) screening *in-vitro* conditions.

Table 1. Characterisation of isolates isolated from rhizospheric soil of wheat from different sites (Deothi , Kandaghat, Dharja, Habban, Rajgarh and Pulwahal) of Solan and Sirmour districts of Himachal Pradesh

Sr.no.	Isolate	Isolated from	Colony size and shape	Colony color	Cell shape	Gram reaction	Catalase
1	**De-1**	**Deothi**	Medium, Round	White	Small rods	+	+
2	**De-2**	**Deothi**	Medium, Round	Milky white	Small rods	+	+
3	**De-3**	**Deothi**	Medium, Round	White	Small rods	+	+
4	**De-4**	**Deothi**	Medium, Round	White	Small rods	+	+
5	**De-5**	**Deothi**	Large, Round	Dark yellow	Cocci	+	+
6	**De-6**	**Deothi**	Medium, Round	White	Small rods	+	+
7	**De-7**	**Deothi**	Medium, Round	White	Small rods	+	+
8	**De-8**	**Deothi**	Large, Round	Off-white	Small rods	+	+
9	**De-9**	**Deothi**	Medium, Round	Milky white	Small rods	+	+
10	**De-10**	**Deothi**	Medium, Round	White	Small rods	+	+
11	**De-11**	**Deothi**	Large, Round	Off-white	Small rods	+	+
12	**De-12**	**Deothi**	Medium, Round	Milky white	Small rods	+	+
13	**De-13**	**Deothi**	Medium, Round	White	Small rods	+	+
14	**De-14**	**Deothi**	Small, Round	Milky white	Small rods	+	+
15	**De-15**	**Deothi**	Medium, Round	White	Cocci	-	+
16	**De-16**	**Deothi**	Medium, Round	Milky white	Small rods	+	+
17	**De-17**	**Deothi**	Medium, Round	Off-white	Small rods	-	+
18	**De-18**	**Deothi**	Medium, Round	White	Cocci	+	+
19	**De-19**	**Deothi**	Small, Round	Off-white	Small rods	-	+
20	**De-20**	**Deothi**	Medium, Round	Off-white	Small rods	+	+
21	**De-21**	**Deothi**	Medium, Round	White	Small rods	-	+
22	**De-22**	**Deothi**	Medium, Round	Milky white	Small rods	-	+
23	**De-23**	**Deothi**	Medium, Round	White	Cocci	+	+
24	**De-24**	**Deothi**	Medium, Round	Milky white	Small rods	-	+
25	**Kn-1**	**Kandaghat**	Small, Round	Yellowish green	Small rods	-	+
26	**Kn-2**	**Kandaghat**	Medium, Round	Greenish yellow	Small rods	-	+
27	**Kn-3**	**Kandaghat**	Medium, Round	Dark yellow	Medium rods	-	-
28	**Kn-4**	**Kandaghat**	Medium, Round	Yellowish green	Small rods	-	-
29	**Kn-5**	**Kandaghat**	Small, Round	Greenish	Small rods	-	-

				yellow			
30	**Kn-6**	**Kandaghat**	Medium, Round	Greenish yellow	Cocci	-	+
31	**Kn-7**	**Kandaghat**	Medium, Round	Yellowish green	Small rods	-	-
32	**Kn-8**	**Kandaghat**	Small, Round	Greenish yellow	Small rods	+	-
33	**Kn-9**	**Kandaghat**	Medium, Round	Dark yellow	Thin rods	-	-
34	**Kn-10**	**Kandaghat**	Medium, Round	White	Small rods	-	+
35	**Kn-11**	**Kandaghat**	Small, Round	Yellowish green	Small rods	-	-
36	**Kn-12**	**Kandaghat**	Medium, Round	Yellowish green	Cocci	-	-
37	**Kn-13**	**Kandaghat**	Medium, Round	Greenish yellow	Oval	-	-
38	**Kn-14**	**Kandaghat**	Medium, Round	White	Small rods	-	-
39	**Kn-15**	**Kandaghat**	Small, Round	Greenish yellow	Medium rods	-	+
40	**Kn-16**	**Kandaghat**	Medium, Round	Greenish yellow	Oval	+	-
41	**Kn-17**	**Kandaghat**	Medium, Round	Yellowish green	Small rods	-	-
42	**Kn-18**	**Kandaghat**	Medium, Round	Greenish yellow	Small rods	-	-
43	**Kn-19**	**Kandaghat**	Small, Round	Yellowish green	Cocci	-	-
44	**Kn-20**	**Kandaghat**	Medium, Round	White	Small rods	-	+
45	**Kn-21**	**Kandaghat**	Small, Round	yellowish	Small rods	-	-
46	**Kn-22**	**Kandaghat**	Small, Round	Greenish yellow	Small rods	+	-
47	**Kn-23**	**Kandaghat**	Small, Round	Yellowish green	Small rods	-	-
48	**Kn-24**	**Kandaghat**	Small, Round	Yellowish green	Small rods	-	-
49	**Pul-1**	**Pulbahal**	Large, Slimy	White	Small rods	-	-
50	**Pul-2**	**Pulbahal**	Large, Slimy	Creamish	Small rods	-	-
51	**Pul-3**	**Pulbahal**	Large, Slimy	White	Small rods	+	-
52	**Pul-4**	**Pulbahal**	Large, Slimy	White	Small rods	+	-
53	**Hb-1**	**Habban**	Medium, Round	Creamish	Small rods	+	-
54	**Hb-2**	**Habban**	Small, Round	yellowish	Small rods	-	-
55	**Hb-3**	**Habban**	Medium, Round	yellowish	Cocci	+	+
56	**Hb-4**	**Habban**	Small, Round	Creamish	Small rods	-	+
57	**Hb-5**	**Habban**	Medium, Round	yellowish	Small rods	+	+
58	**Hb-6**	**Habban**	Medium, Round	Creamish	Small rods	-	+
59	**Rj-1**	**Rajgarh**	Medium, Round	White	Small rods	+	+
60	**Rj-2**	**Rajgarh**	Small, Round	Cream	Small rods	-	-
61	**Rj-3**	**Rajgarh**	Medium, Round	Cream	Small rods	+	+
62	**Dh-1**	**Dharja**	Medium, Round	Creamish	Thin rods	-	+
63	**Dh-2**	**Dharja**	Large, Round	Creamish	Small rods	-	-
64	**Dh-3**	**Dharja**	Large, Slimy	Creamish	Medium rods	-	-
65	**Dh-4**	**Dharja**	Medium, Round	Creamish	Small rods	-	-
66	**Dh-5**	**Dharja**	Large, Round	Creamish	Small rods	-	-
67	**Dh-6**	**Dharja**	Large, Round	Creamish	Cocci	-	-
68	**Dh-7**	**Dharja**	Large, Round	Creamish	Thin rods	+	+
69	**Dh-8**	**Dharja**	Medium, Round	Creamish	Small rods	-	-
70	**Dh-9**	**Dharja**	Large, Round	Creamish	Small rods	-	-
71	**Dh-10**	**Dharja**	Large, Round	Creamish	Small rods	-	-

72	**Dh-11**	**Dharja**	Large, Round	Creamish	Thin rods	-	-
73	**Dh-12**	**Dharja**	Large, Round	Creamish	Small rods	-	-

Plant growth promoting attributes

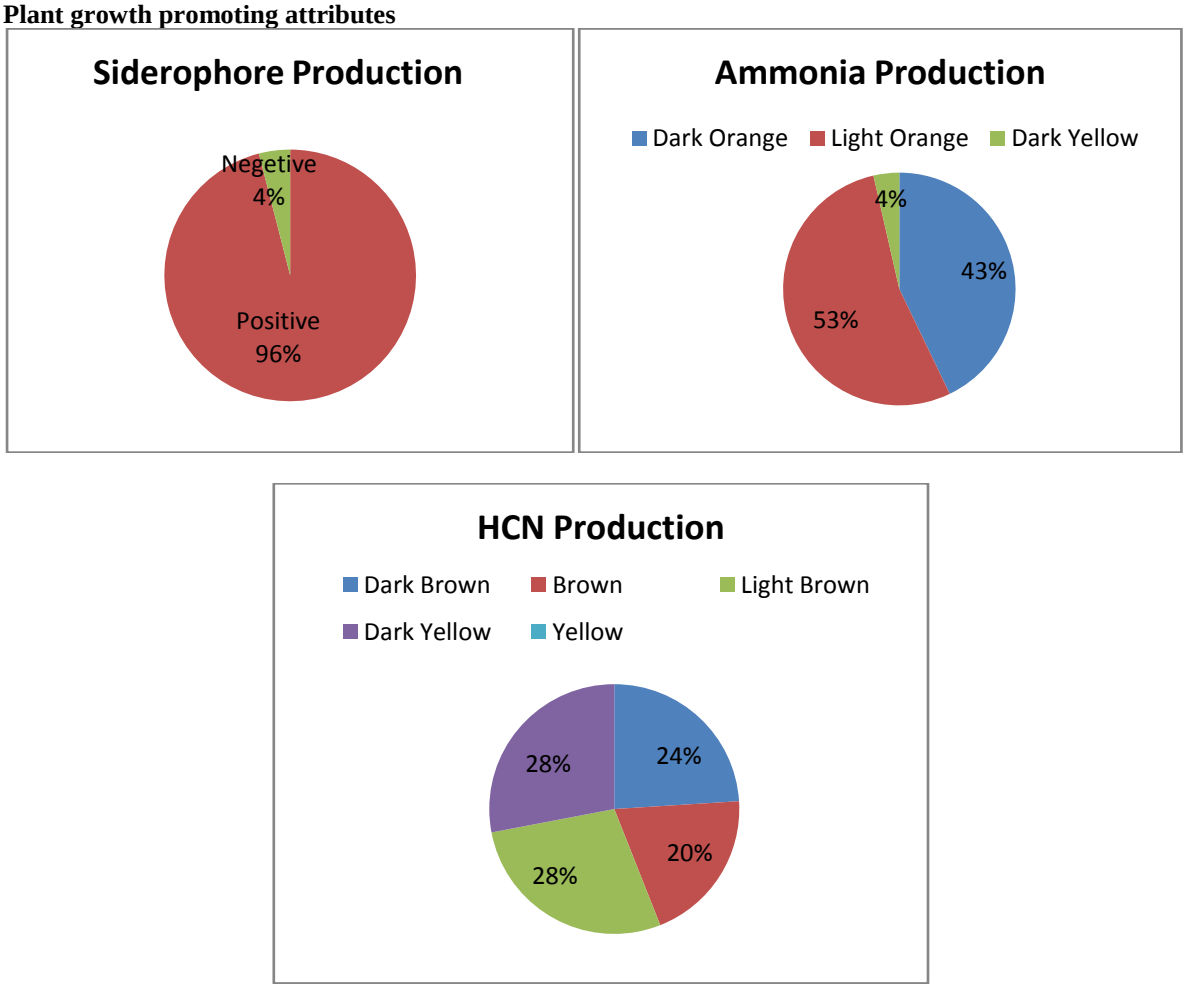


Fig 1. Plant Growth Promoting attributes of bacterial isolates from the wheat rhizosphere in the mid Himalayan zone of Himachal Pradesh

P-Solubilization

All the twenty nine isolates of PGPR showed positive for inorganic P-Solubilization *in vitro* conditions. Phosphorus (P) is one of the major essential macronutrients for biological growth and development. Rhizobacteria offer a biological rescue system capable of solubilizing the insoluble inorganic P of soil and make it available to the plants. The ability of some rhizobacteria to convert insoluble phosphorus (P) to an accessible form, like orthophosphate, is an important trait in a PGPB for increasing plant growth and yield (Saharan and Nehra 2011).

Siderophore production

Out of twenty nine isolates 96% isolates showed positive and 4% showed negative results for siderophore production. Siderophores are small iron carriers, chemically high-affinity iron chelating compounds secreted by PGPR's and are among the strongest soluble Fe^{3+} binding agents known. Comprehensive information on the role of siderophores in increasing iron oxide solubility and promoting dissolution in soils requires the consideration of the rates of various processes such as siderophore exudation, the uptake, and the degradation rates (Scavino and Pedraza, 2013).

Ammonia production

Out of twenty nine isolates 43% isolates showed dark orange, 53% light orange and 4% showed dark yellow color in case of ammonia production. Ammonia production is also a important traits of PGPR because huge amount of free nitrogen is present in the environment but this form of nitrogen is unavailable for the plants, so in ammonia production the free nitrogen is converted to ammonia by the enzyme nitrogenase present in the bacteria, which is the suitable absorbable form of nitrogen for the plant growth.

HCN

Out of twenty nine isolates 24% showed dark brown, 20% brown, 28% light brown and 28% dark yellow coloration of filter paper strip. A secondary

metabolite produced commonly by rhizospheric bacteria is Hydrogen Cyanide (HCN), a gas known to compatible mechanism for biological control of major plant pathogens (Heydari *et al.*, 2009). Hydrogen Cyanide is a poisonous gas produced by rhizobacteria as it has toxic properties.

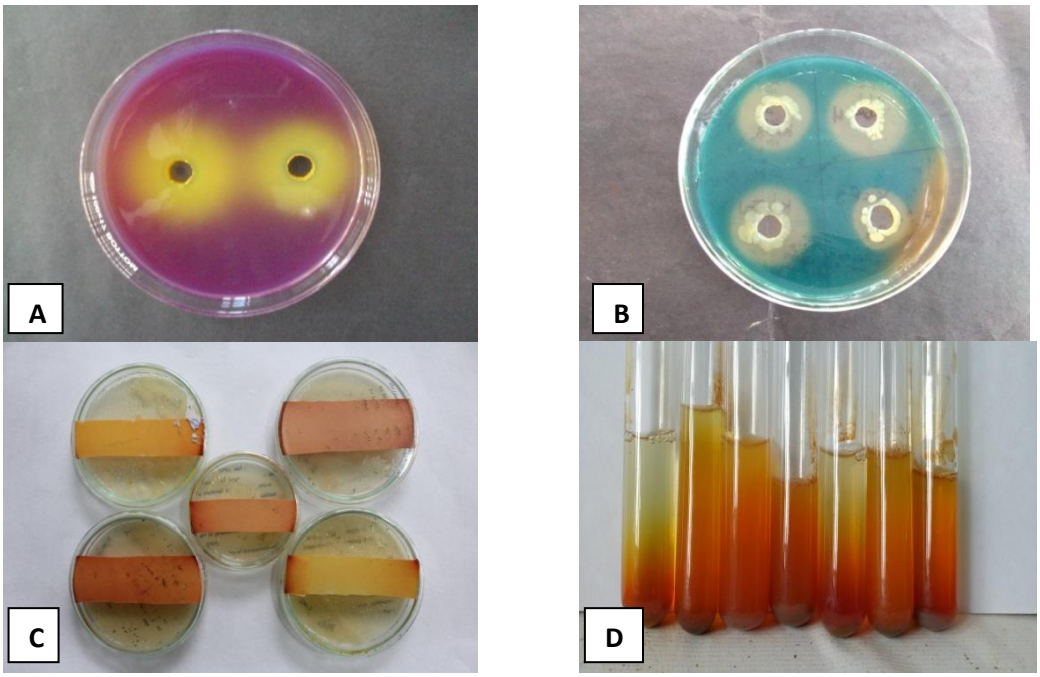


Plate 1. Production of Siderophore (A), Phosphate solubilization (B), HCN and Ammonia (C,D) shown by PGPR's isolates on PVK, CAS –agar, peptone water and nutrient agar supplemented with glycine at 28± 2^{0}C respectively.

Pot experiment

Out of twenty nine isolates of PGPR, three hyperpotential isolates Dh-7 (*Bacillus pumilus*), Kn-7 (*Pseudomonas putida*) and De-21 (*Stenotrophomonas maltophilia*) were selected for pot experiment because of their hyperpotential among PGP attributes. The results indicated that the percentage of the seed germination and shoot elongation of wheat was significantly increased under the influence of the consortia of the PGPR along with different doses of fertilizer in comparison with the control.

Seed Germination percentage

The germination percentage of seeds was 100% in each treatment except control plants with 50% NPK, Kn-7+De-21 with 50% NPK and Dh-7+De-21 with 50% NPK showed 93.33% germination. All the seeds were germinated after one week of sowing. Survival percentage of wheat plants in each treatment was 100 % along with control plants (figure 2).

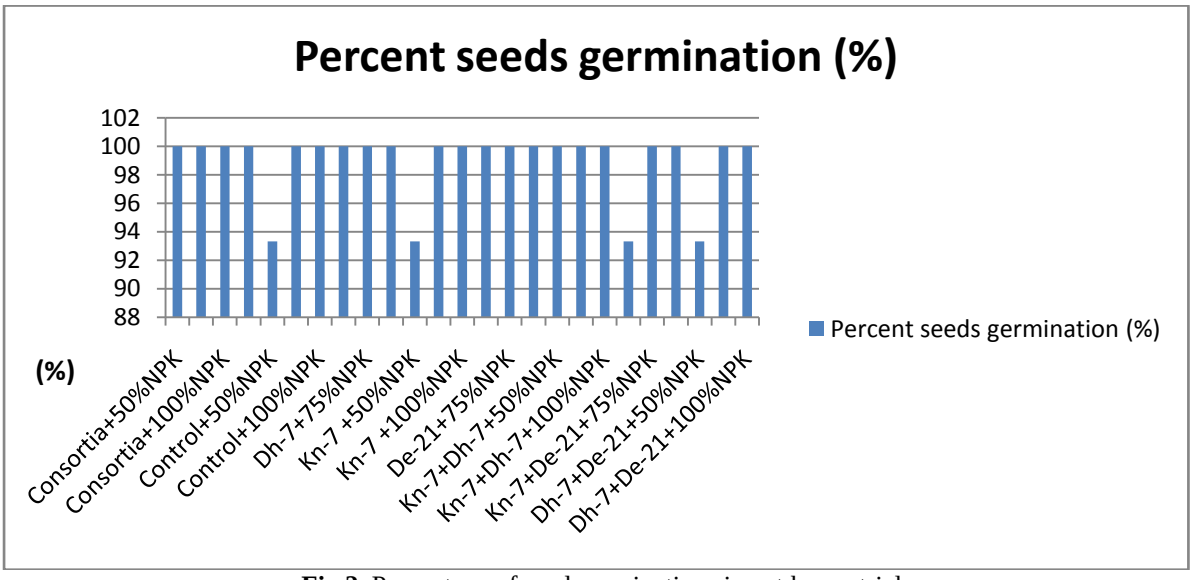


Fig 2. Percentage of seed germinations in net house trial

Shoot growth

On an average, in all the treatments with bioformulation of individual and consortia of isolates Dh-7, Kn-7 and De-21, there was an increase in plant height as compared to control after 45 days of seed sowing of *Triticum aestivum*. All the formulations showed significant increase in plant height after regular interval of time. The maximum height (19.83 cm) was recorded in consortia of three isolates with 100% NPK dose followed by (19.24 cm) consortia of three isolates with 50% NPK dose after 45 days of seed sowing. Minimum plant height was recorded in individual treatment of Kn-7 with 50% NPK dose i.e. 14.58 cm. It was found that shoot growth enhancement was observed in the treatments of consortia of three isolates (Kn-7, Dh-7 and De-21) with 100% NPK dose and consortia of three isolates without NPK dose this is due to that these strains were effective when applied collectively instead individually they were found to solubilize inorganic phosphate in the rhizosphere of wheat plants so they provide available form of phosphorus to the plants, production of siderophore which chelate the iron from the rhizospheric environment and made available to the plants so no iron deficiency was there for the PGR treated plants.

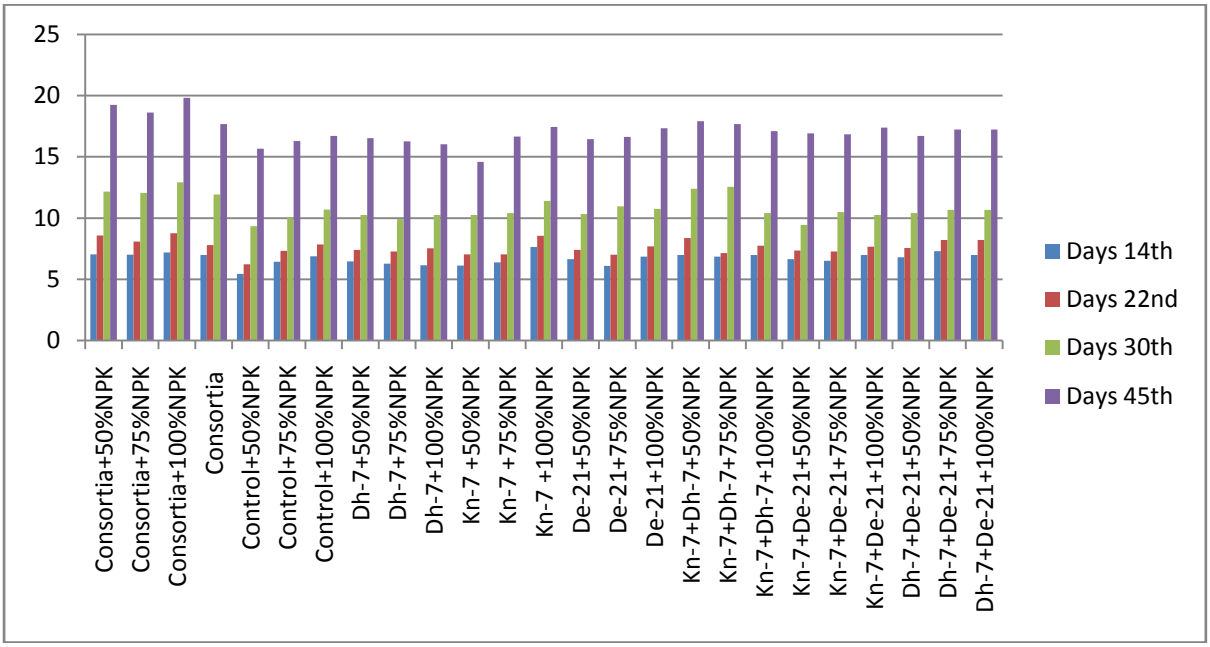
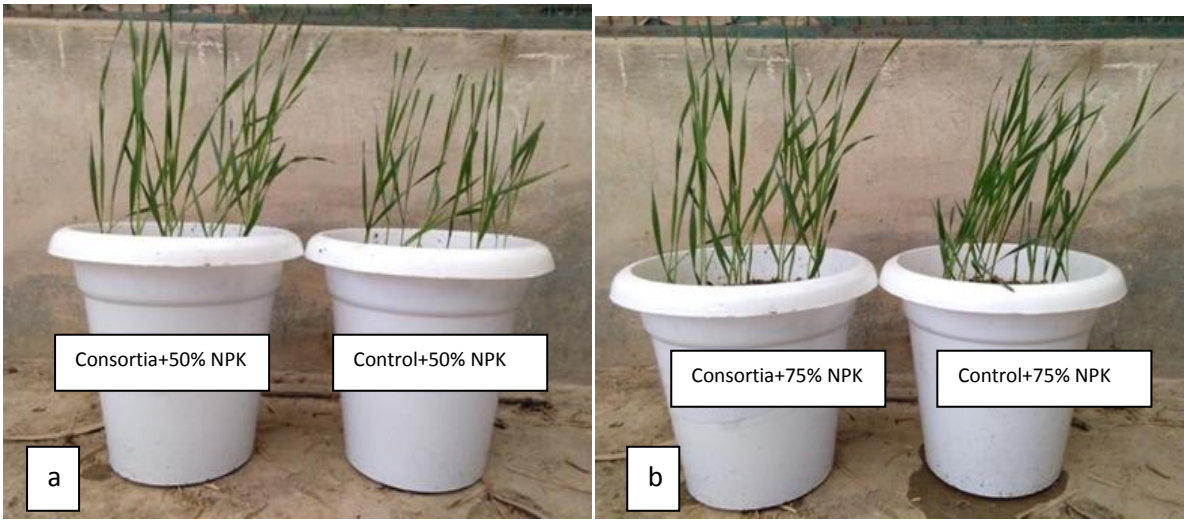


Fig 3. Effect of PGPR formulation on the shoot growth promotion of wheat seedlings in net house experiment



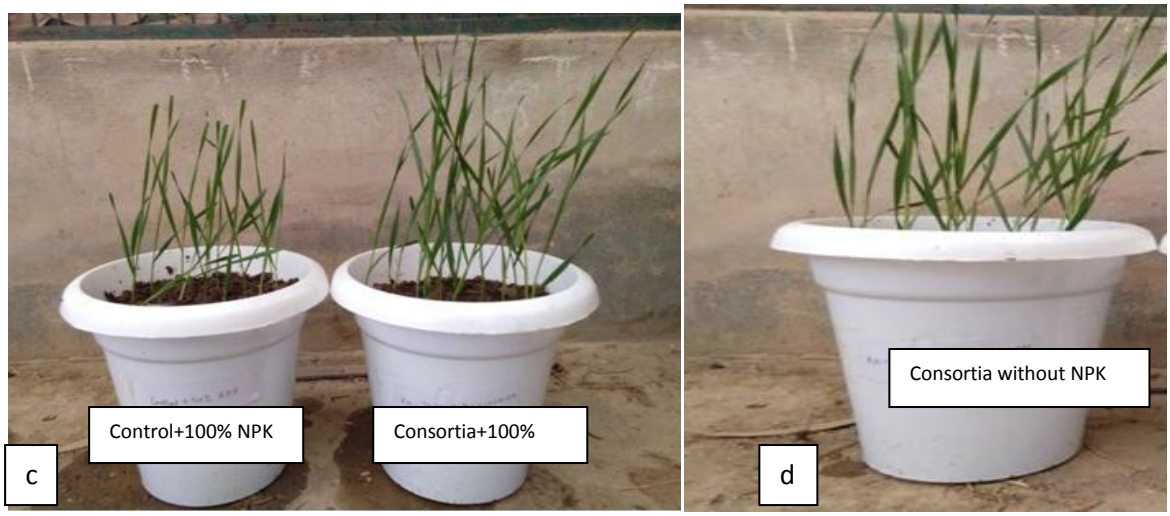


Plate 2. Effect of PGPR formulation on the shoot growth promotion of wheat seedlings in net house experiment

Plate 3. Overview of net house experiment at UHF Nauni- Solan (H.P)

Chlorophyll content of leaf

All the treatments comprised of PGPR significantly increased the chlorophyll content of leaves (a, b and total) over uninoculated control plants after 30 days of plantation (Fig 4). The chlorophyll content 'a' ranged from 0.11 to 0.13 mg/g fresh weight, whereas maximum chlorophyll content (0.138 mg/ml fresh weight) was noted in treatment consortia of three isolates with 100% NPK and minimum (0.115 mg/g fresh weight) was found in treatment Dh-7 with 50% NPK dose. Overall the chlorophyll content 'b' of leaves ranged from 0.21 to 0.26 mg/g fresh weight where the maximum chlorophyll content (0.264 mg/g fresh weight) was noted in treatment De-21 with 100% NPK dose and minimum was recorded for treatment Dh-7 with 50% NPK dose (0.210 mg/g fresh weight). The increase in total chlorophyll content of leaves was found in the range of 0.33 to 0.38 mg/g fresh weight. While the maximum chlorophyll content (0.386 mg/g fresh weight)) was noted for consortia with 100% NPK dose and minimum (0.336 mg/g fresh weight) was noted for treatment Dh-7 with 50% NPK dose and De-21 with 50% NPK dose.

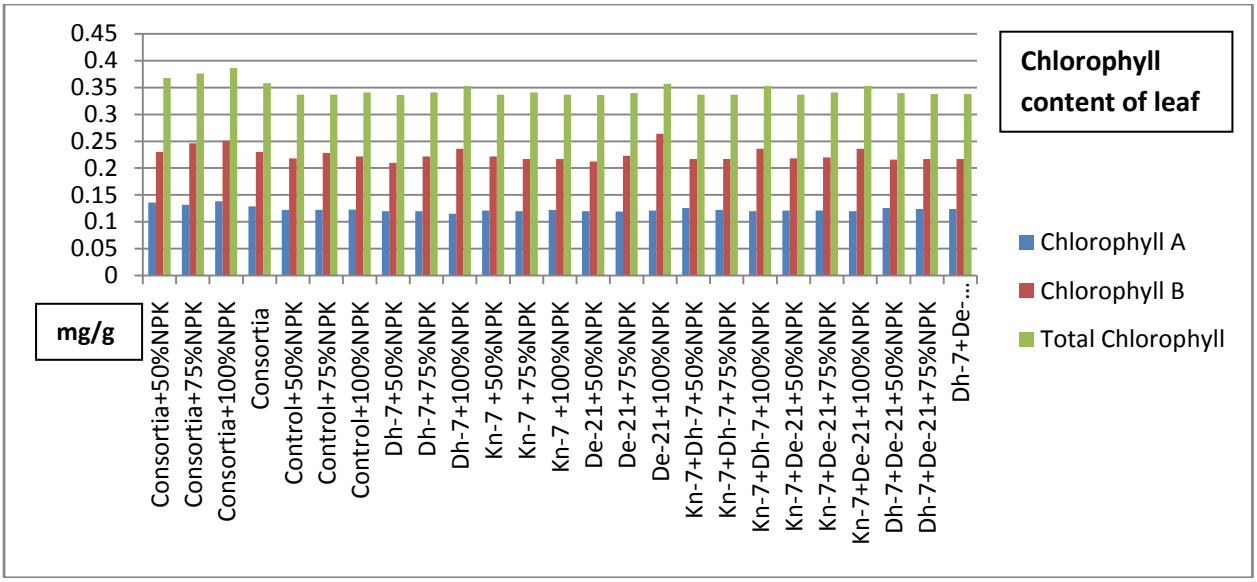


Fig 4. Effect of PGPR formulation on the chlorophyll content of leaves of wheat plants in net house experiment

DISCUSSION

The principal mechanism for mineral phosphate solubilization is the production of organic acids, and acid phosphatase play a major role in the mineralization of organic phosphorus in soil (Gaur *et al.,* 2005). Phosphorus (P) is one of the major plant nutrients, that promote shoot and root growth of plants. Chemical fertilizers are main source for phosphorus supplying in agricultural systems but about 75-90 % of added phosphorus to the soil is being fixed by Fe, Al^{3+} and Ca^{2+} complexes (Tarun *et al.*, 2006). The phosphate solubilization is the most common mode of action implicated by PGPR that increases nutrient availability to host plants (Thakur, 2014). Gupta (2012) also reported that the population of phosphate solubilizing microorganisms, in general, varied from 20-24% of the total population, however in some soils it may be 85% of the total population. In another studies conducted by Kundu *et al*., (2002) reported that about 16% of the total bacterial population in rhizosphere of wheat was P-solubilizer.

The organisms used were siderophoregenic pyoverdin-producing *Pseudomonas putida* and *Pseudomonas aeruginosa* strains from two diverse habitats. Inoculation with siderophoregenic PGPR increased percentage germination, shoot height, shoot and root length, weight of spikelets, chlorophyll content, grain yield and iron content in wheat crop (Sarode *et al.*, 2013, Mishra *et al.,* 2013). Similar study was demonstrated that *Acinetobacter calcoaceticus* isolated from wheat rhizosphere produces catechol type of siderophores during exponential phase, which is influenced by iron content of medium (Sarode *et al.,* 2009).

Previous studies Chaiharn *et al.,* (2008) reported the production of ammonia by phosphate solubilizing microorganisms, more than 64% of the isolates were found to produced ammonia. Another study by Ahmad *et al.,* (2006) reported that the several plant growth promoting rhizobacteria were found to produce ammonia in peptone water amended with Nesseler's reagent.

Although Hydrogen Cyanide acts as a general metabolic inhibitor, it is synthesized, excreted and metabolized by hundreds of organisms present in the rhizosphere mainly Plant Growth Promoting Rhizobacteria (PGPR) (Zeller *et al.,* 2009). The similar study demonstrated that the HCN production is found to be a common trait of *Pseudomonas* (88.89%) and *Bacillus* (50%) in the rhizospheric soil of wheat a biocontrol metabolite in *Pseudomonas* species (Saharan and Nehra, 2011). Another previous study was demonstrated that the *Pseudomonas fragi* CS11RH1 (MTCC 8984), a psychrotolerant bacterium produces hydrogen cyanide (HCN) in the presence of glycine (Seval kumar *et al.*, 2009).

In this study seeds inoculation with the individual and consortia of bacterial culture has been found to improve the percentage seed germinations with different doses of fertilizer this was due to nutrient uptake of wheat seedlings via promotion of the plant growth and increased root surface area or the general root architecture of the treated seeds. Seeds inoculated with the bacterium has been found to improve the growth and nutrient uptake of wheat seedlings via promotion of the plant growth and increased root surface area or the general root architecture (Lucy *et al.,* 2004). (Laid *et al.,* 2016) Results analysis of PGPR effects of actinomycetes isolates on growth parameters show that the isolates have a significant effect on the germination rate of the treated and untreated seeds of the same degree by bioformulation. Analysis of variance revealed a very highly significant effect on germination rate for

foliage, and significant for shoot growth and root length.

Rana *et al.,* (2011) reported that number of other *Bacillus* spp. isolated from wheat rhizosphere have also been investigated for their growth-promoting property in wheat having similar effects on dry weight. All the nine selected PGPR significantly increase the root length (cm), dry root weight (g plant-1), shoot length (cm) and dry shoot weight (g plant-1) as compared to media (uninoculated) and control (uninoculated). Three PGPRs identified as *Bacillus anthracis* (A29), *Serratia proteamaculans* (A28) and *Psychrobacter maritimus* (A18) were performed best in growth chamber and selected as best potential strains the others (Amara *et al.,* 2015). Hayat *et al.,* (2012-13) also reported that *Bacillus, Enterobacter, Pseudomonas* and *Serratia* sp. were very good PGPRs with PGP traits like IAA production, phosphate solubilization and N_2-fixation and are also being used for crop production as bioinoculants. The same results were concluded by Adesemoye *et al.,* (2010) who reported that PGPR's applied along with fertilizers promote plant growth.

According to Zahra *et al.,* (2012), use of rhizobacterial inoculants as biofertilizer significantly improved the growth parameters of cereals. Various researchers reported that under controlled conditions, root and seed inoculation with PGPRs enhance root growth through PGP activity. Similar results are presented by Shaharoona *et al.,* (2008), who reported improved

efficiency of nutrients uptake by inoculation of PGPRs which resulted in increased root growth and hence efficient uptake of nutrients by plants.

Importance

Inoculations of wheat seeds with consortia of (Dh-7, Kn-7 and De-21) along with recommended dose of fertilizes has direct positive effect on increase in shoot growth, percentage of seed germination and chlorophyll content of the leaves under net house conditions.

CONCLUSION

This study that has provided an insight into the rhizobacterial community present in the rhizosphere of wheat in different locations of Solan and Sirmour district in the mid hill zone of Himachal Pradesh, India. We have demonstrated that efficient inorganic P-solubilizer, siderophore, ammonia and HCN producing rhizobacterial isolates were present among the natural population in the rhizosphere of wheat in this area. These characteristics are considered as important PGP attributes. In the present study we have been found that the consortia (*Pseudomonas putida*, *Stenotrophomonas maltophilia* and *Bacillus pumilus*) with the recommended doses of fertilizer (NPK) have been found effective in positively improving the seed germination percentage, chlorophyll content of leaf and increase in shoot growth of tested wheat plants. It is an environment friendly and cost effective technology.

ACKNOWLEDGMENTS

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RESEARCH ARTICLE

POPULATION STRUCTURE OF VEGETATION IN URBAN ENVIRONMENT OF SARGUJA, CHHATTISGARH, INDIA

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Abstract: The present study was conducted in different directions(east, west, north and south) of Ambikapur to explore the urban vegetation in terms of species status, population structure and regeneration potential of species. A total of 10 tree species distributed into 6 families were recorded in east direction, 9 tree species with 4 families in west direction, 12 tree species comprised of 9 families in north direction, and 11 tree species belonging to 8 families were recorded in south direction. The tree density ranged between 170-240 trees/ha across the site being highest under north direction and least at east direction. The rarity and commonness of the species in urban setup reflected that majority of the species are rare in occurrence in different stratum while the intermediate, moderately high and common (high frequency) species class was almost negligible in the entire site in most of the vegetation stratum. Population structure of the species also revealed the younger vegetation stand in all the direction due to absence of the different size classes of the species. The regeneration of the species was not found up to the mark in all the direction. Therefore, there are needs for the conservation priority to manage the urban landscape for better management and planning.

Keywords: Structure, Population dynamics, Regeneration, Urban vegetation

INTRODUCTION

The vegetation such as tree, shrub and herb present in cities play vital role in improving urban environment. It plays a key role in maintaining various components of ecosystem and the biodiversity (Jim and Chen, 2009). Tree species primarily provide fresh air/ oxygen and are essential for social life to the residents of a locality; therefore, it is necessary to have a detailed record of trees of an urban locality. Trees have good potential of tapping atmospheric carbon through photosynthesis and thus reduce atmospheric carbon (Yadav et al. 2017; Jhariya et al. 2019). Tree species also help in mitigating climate change and reduces urban temperature. Plants store carbon in terms of live biomass, which becomes a part of the food chain and enters into the (Gavali and Sheikh, 2016). Urban forests sequester carbon and affect the emission of $CO_2$by reducing its level from urban areas. Thus, urban forests play a major role in managing the increase in level of CO_2. Urban trees store carbon derived from CO_2, which is the major gas contributing to global climate change (Saral et al. 2017).

The net save in carbon emissions that can be achieved by urban planting can be up to 18 kg CO_2/year per tree and this benefit corresponds to that provided by 3 to 5 forest trees of similar size and health (Francesco and Alessio,2011). As in urban area level of pollution is very high, this vegetation helps in pollution control.Vegetation in urban areas, particularly trees, directly removes air pollution and can also provide barriers between sources and exposed populations. Urban trees perform important ecological function in cities by sequestering carbon and reducing automobile pollution. Urban vegetation can provide numerous benefits beyond air quality improvements, including temperature and storm water regulation, noise reduction, aesthetic improvements, and environments conducive to physical exercise and experiencing nature. These co-benefits or ecosystem services of urban vegetation have been associated with improved physical and mental health and community vitality (Baldauf and Nowak, 2014).Therefore, the present investigation is carried out to record the urban vegetation structure, population dynamics and associated ecological attributes in Sarguja (Chhattisgarh), India to enrich the information for better management and conservation of these resources under the urban setup.

MATERIALS AND METHODS

The vegetation was analysed in different sites (i.e., east, west, north and south) of Ambikapur, Sarguja. The trees, saplings, seedlings and shrubs were analysed by randomly laying quadrats of size 10 m x 10 m. The girth at breast height (i.e., 1.37 m above the ground) of all the trees and saplings in each quadrat was measured.While in the case of seedling the numbers of individuals by species were recorded separately. The shrubs were measured at collar height.For herb a quadrat of 50 cm x 50 cm was laid and number of individuals was counted in various seasons.

The species rarity or commonness of the species was calculated as the frequency class of the species (Raunkiaer, 1934; Hewit and Kellman, 2002). As per

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frequency classes, the species were categorized as A, B, C, D, and E, where A represents rare (0–20%), B represents low frequency (20–40%), C represents intermediate frequency (40–60%), D represents moderately high frequency (60–80%), and E represents high frequency or common (80–100%).The population structures of vegetation were developed by using thevarious girth classes of tree species. The total number of individuals corresponds to these girth classes was calculated at species level for various direction (Saxena and Singh, 1984; Tripathi et al. 1991; Jhariya et al. 2012). Besides to seedling (A) and sapling (B) classes, three more tree size classes viz., C(31.5−70.0 cm), D(70.1−110.0 cm) and E (>110 cm) were arbitrarily established at species level through graphical representation. The regeneration potential of urban vegetation was determined as per the Khan et al (1987).

RESULTS AND DISCUSSION

Species status and vegetation statistics

The present study revealed that total 10 species of tree species distributed into6 families were recorded in east direction, in west direction total 9 tree species with 4 families, in north direction 12 tree species comprised of 9 families, and in south direction 11 tree species belonging to 8 families were recorded (Table 1). The tree density ranged between 170-240 trees/ha across the site being highest under north direction and least at east direction. The north direction was found rich in number of species, families as well as density as compared to other direction.Under sapling layer, in west direction 3 species with 1 family was found while in north and south direction 2 species with 1 family was recorded. The sapling density was highest in west direction while least towards north direction (Table 1). Seedling stratum revealed that the higher number of species and families were recorded towards north direction and least at south direction. The higher seedling density was found in north direction while least at east direction. Higher shrub species was found in the both east and north directions while least at south direction (5 species). The highest shrub density was recorded at east direction and least towards south direction (Table 1).The herbaceous vegetation reflects substantial seasonal variation across the site in different season in terms of herb richness, families and density.During rainy season, a total of 5 herb species belonging to 4 families were found in the east direction.In west direction total 7 herb species with 4 families were recorded, in north direction 9 herb species with 7 families, and in south direction 5 herb species belongs to 4 families were noticed. The density of herb was found higher towards north direction in rainy season. During winter season the highest number of species and families were recorded at north direction while least at east direction. The herb density during winter was also found more in north direction and lowest in both east and south directions, respectively. In summer season, higher number of herb species was found in west direction, and higher family was found both in west and north direction, respectively. The least number of species and families were recorded towards south direction. The density of herb in summer was found highest at north and lowest at south direction, respectively (Table 1).

Table 1. Vegetation statistic at different direction in an urban setup of Sarguja

Attributes	East	West	North	South
Tree				
Number of species	10	9	12	11
Number of families	6	4	9	8
Number of individuals (per hectare)	170	220	240	190
Sapling				
Number of species	0	3	2	2
Number of families	0	1	1	1
Number of individuals (per hectare)	0	50	20	40
Seedling				
Number of species	3	4	8	2
Number of families	1	1	4	1
Number of individuals (per hectare)	30	400	420	70
Shrub				
Number of species	7	6	7	5

Number of families	5	6	6	4
Number of individuals (per hectare)	570	260	450	230
Herb				
Rainy				
Number of species	5	7	9	5
Number of families	4	4	7	4
Number of individuals (per hectare)	138000	114000	152000	90000
Winter				
Number of species	3	6	7	4
Number of families	3	3	5	3
Number of individuals (per hectare)	72000	94000	104000	72000
Summer				
Number of species	5	12	9	4
Number of families	3	7	7	2
Number of individuals (per hectare)	100000	134000	184000	88000

The urban vegetation reflects significant variation among them. The vegetation in different stratum shows rich diversity in terms of number, species and family across the sites. The present findings were comparable withOgwu et al. (2016), they reported 20 tree species with 12 families during their investigation. Further, Agbelade et al. (2017) mentioned 69 species with 29 family of tree species in urban centre whereas 20 species with 12 family of tree species in peri-urban area of Nigeria.

Species rarity and commonness

The tree layer reflectstotal 10 speciesin east sitewhich revealed all the species found in class Arepresenting rare frequency (Table 2). Towards west direction total 9species occurred, which revealed only two frequency classes *viz*., class A (comprised of 77.78% species) and class B(22.22% species). Towards north direction 12 species occurred, in class A 91.67% species found representing rare frequency,in class B 8.33% species recorded. In south direction 11 species occurred, in class A 90.91% species was found representingrare frequency, in class B 9.09% species was recorded, representing low frequency. It reflected from the Table 2 in tree layer the frequency classes C,D, E were totally absent in all the direction.Thesapling layer reflects that towards east direction no presence of species under this layer while in other directions sapling were represented only in rare class (A) while the class B, C, D and E was totally absent. The seedling layer showed rare and low frequency classes for west and north direction while east and south direction reflects only by class A. The frequency class C, D and E were totally absent in the entire site studied (Table 2).

Total 7 shrub species occurred in east direction (Table 2), 57% species found in class A representing rarefrequency, 14% species found in class B showing low frequency and 29% species found in classD representing moderately high frequency while in C and E class species were totally absent. Towards west direction 6 species occurred, in class A 67% species was found, in class B 17% specieswas recorded, and in class C 17% species found showing intermediatefrequency while in class D and E species were totally absent. At north direction 7 species occurred, in class A57% species found, in class B 29% species recorded, representing low frequency in class D 14%species representing moderately high frequency while in class C and E species were absent. At south direction 5species occurred, in class A 60% species was found, in class B 40%species was recorded, while species were totally absent in C, D and E classes (Table 2).

During rainy season (Table 2)5 herb species in east direction was recorded in which 60%species found in class B and remaining in class C (40%). Towards west direction 7 speciesoccurred, in class A 57.14% species were found and in class B42.86% species were recorded. At north site 9 species occurred, in which class A comprised of 55.56% species, in class B 11.11% species was recorded, and in class C33.33% species were found. In south direction 5 species occurred, in class A 60% species were found and remaining in class B (40%). In winter season 3 species occurred in east site, 33.33% species in class A and in class B 66.67% species were found. At west direction 6 species occurred, 50% species in each frequency class i.e., A and B were found. Towards north direction 7 species occurred, inclass A 71.43% species were found, and remaining by class B

(28.57%). At south direction4 species occurred, in class A 75% species were found, and in classB 25% species was recorded. Herbs in summer season at east direction reflected 2 species,of which 80% species was in class A and 20% species in class B. At west direction 6 species occurred which was distributed only frequency class A (100%). Towards north direction 9species occurred, in class A 66.67% species were found, and in classB 33.33% species were recorded. South direction contains 4 species, in class A 75% species was found, and remaining (25%) in class B. The size class C, D, E are mostly absent in case of herb layer across the site in all the season except rainy season in east and north direction, respectively (Table 2).

The species occurrence and life-forms are generally associated with specific climatic regimes. In present investigation it was found that species class A (species rarity) was prevailing in different direction of urban landscape. The subsequent classes were decreasing in terms of number of species presence and even nil or negligible in higher frequency class. These findings were also supported by Oraon and Jhariya (2018). They also found the similar trend of species distribution as per the frequency class.

Table 2. Species rarity or commonness in an urban setup of Sarguja

Species	Sites	Rare (A)	Low frequency (B)	Intermediate (C)	Moderately high (D)	High frequency (E)
Tree	East	10	0	0	0	0
	West	7	2	0	0	0
	North	11	1	0	0	0
	South	10	1	0	0	0
Sapling	East	0	0	0	0	0
	West	2	0	0	0	0
	North	2	0	0	0	0
	South	2	0	0	0	0
Seedling	East	3	0	0	0	0
	West	1	3	0	0	0
	North	6	2	0	0	0
	South	2	0	0	0	0
Shrub	East	4	1	0	2	0
	West	4	1	1	0	0
	North	4	2	0	1	0
	South	3	2	0	0	0
Herb (Rainy season)	East	0	3	2	0	0
	West	4	3	0	0	0
	North	5	1	3	0	0
	South	3	2	0	0	0
Herb (Winter season)	East	1	2	0	0	0
	West	3	3	0	0	0
	North	5	2	0	0	0
	South	3	1	0	0	0
Herb (Summer season)	East	4	1	0	0	0
	West	12	0	0	0	0
	North	6	3	0	0	0
	South	3	1	0	0	0

Relationship of density to GBH

Tree species density-GBH distribution followed a linear model [Y = lin. (a − bx)] on all the direction of the Ambikapur. The sites thus exhibited a small structure as 15-67.74% individuals had ≤10 cm girth, and only 6.45–24.14% were in girth classes exceeding 10 cm GBH (Figure1), while 28.13–70% individuals were exceeding >50 cm GBH. When data were pooled, the woody species density was related to GBH according to:

(1) East direction: Y = lin. [5.714x+5.714]
(2) West direction: Y =lin. [-33.57x+225.7]
(3) North direction: Y = lin. [-35.71x+231.4]
(4) South direction: Y = lin. [-1.071x+45.71]

Linear relationship between density and GBH reflected structure vegetation where only 28.13 70% individuals were in classes >50 cm GBH. Jhariya and Yadav (2018) reported that the sites possessed small structure (≤10 cm girth) as 72.66% and 73.87% individuals, respectively in natural and teak stand. Further the relationship between density and girth class revealed 1.27-6.22% individuals were distributed in girth class ＞50 cm. This may be because of higher biotic pressure and faster turnover in the study sites (Jhariya and Yadav, 2018). Similarly, Oraon (2012) mentioned up to 1.5–3.7% of individual species was in girth classes exceeding 50 cm GBH and nearly 89-94% individuals had girth class of ≤10 cm. Jhariya (2014) reported that 86.37–91.71% individuals had ≤10 cm girth, 8.29–13.63% in girth of more than10 cm, while 1.58–2.18% individuals were represented with exceeding girth class of>50 cm in an forest stand.

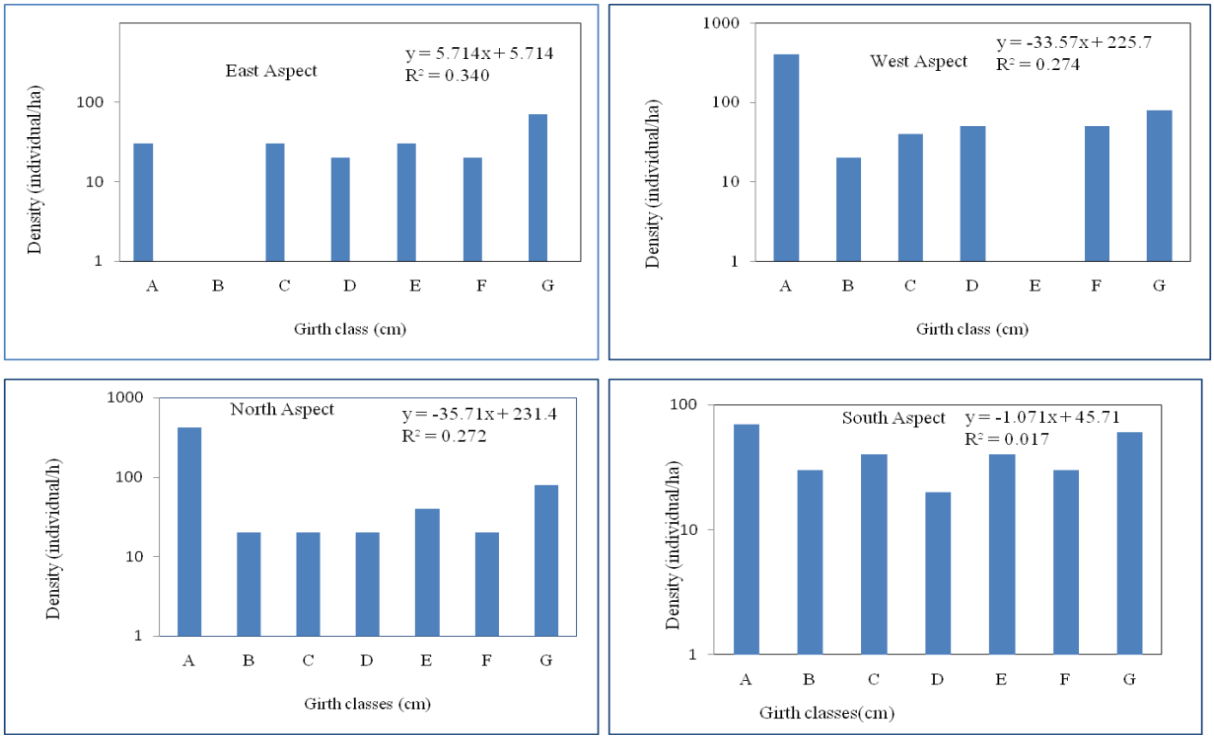
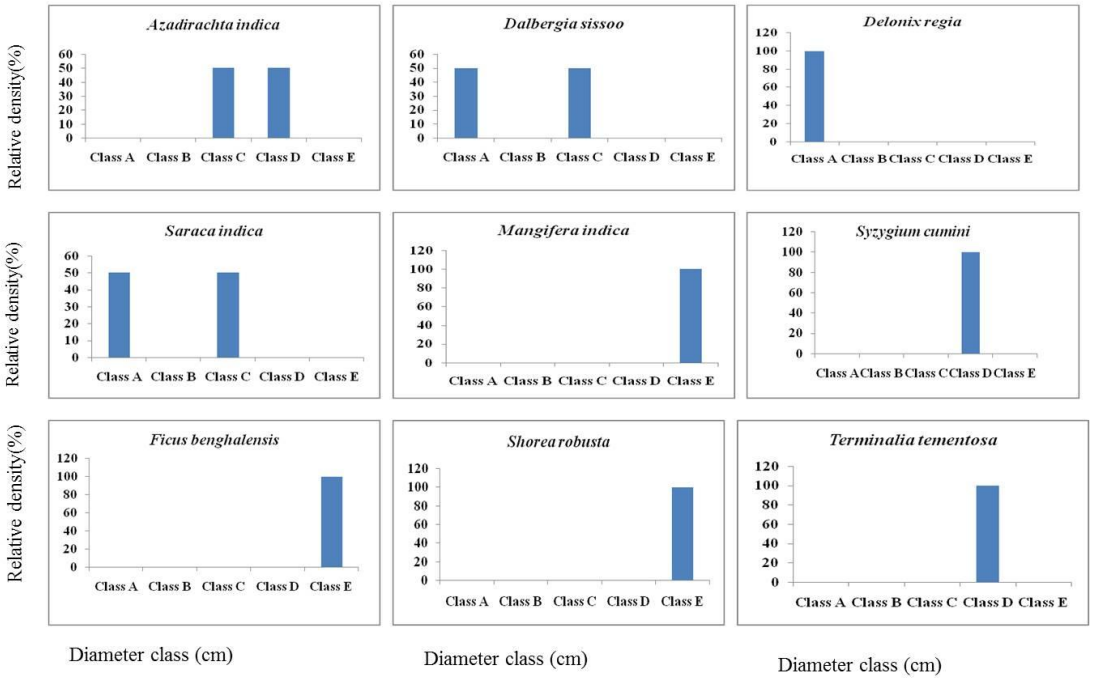


Figure 1. Tree species density and GBH relationship of urban vegetation in Sarguja

Population Structure of Urban Vegetation

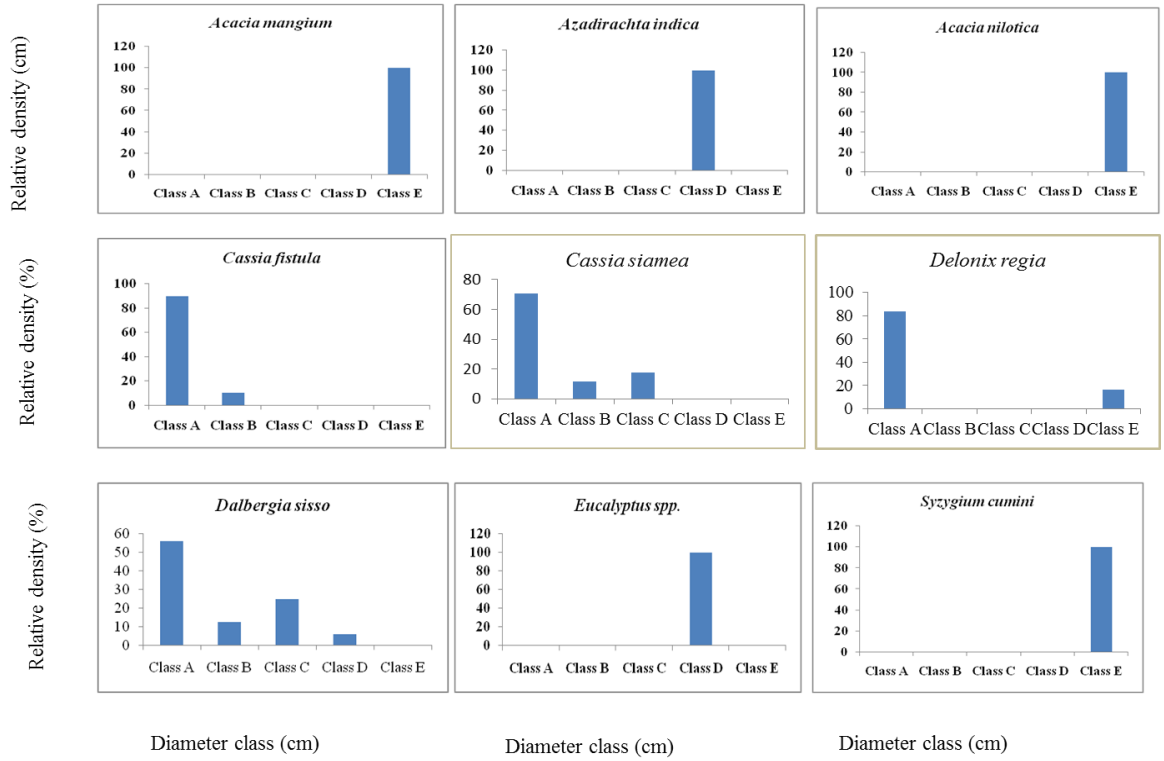
Population structure of tree species towards east site reflected class A was represented by *Dalbergia sissoo, Delonix regia,Saraca indica,* no species found in class B, class C was represented by *Azadirachta indica,Dalbergia sissoo, Saraca indica,* class D was represented by *Azadirachta indica, Syzygiumcumini, Terminalia tomentosa,* class E was represented by *Mangifera indica, Ficus benghalensis,Shorea robusta, Terminalia bellerica, Terminalia chebula.* In east aspect class E was represented also represented by few species but have poor regeneration (Figure 2).

Figure 2 Population structure of tree species in east direction of urban setup



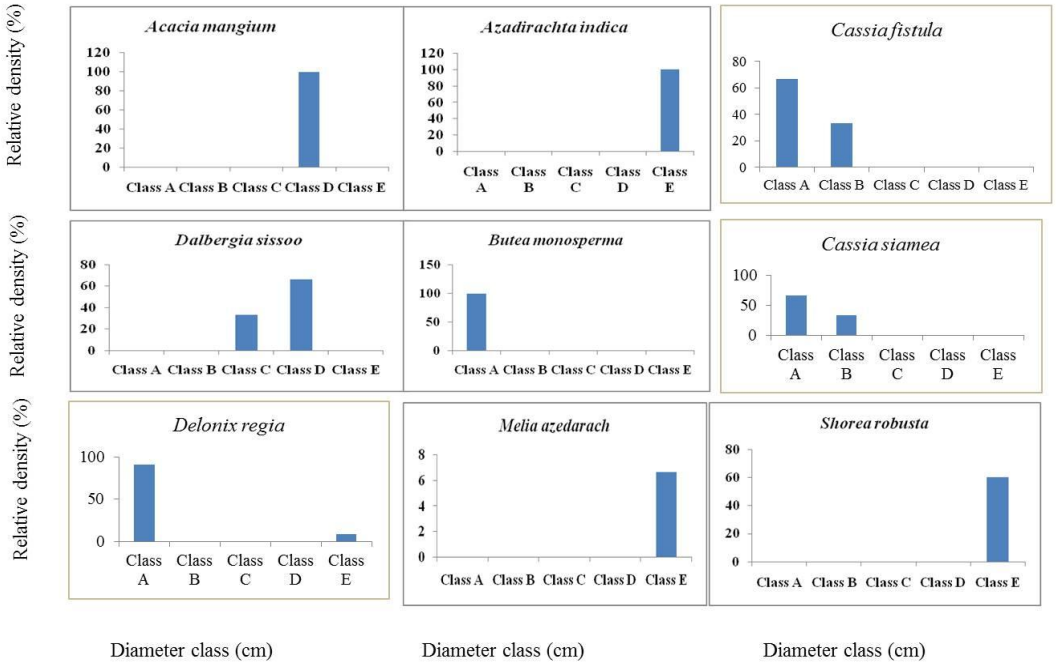
Towards west direction class A was represented by *Cassia siamea, Cassia fistula, Dalbergia sissoo, Delonixregia,* class B was represented by *Cassia siamea, Cassia fistula, Dalbergia sissoo,* class C wasrepresented by *Cassia siamea, Dalbergia sissoo, Zizyphus mauritiana,* class D was representedby *Azadirachta indica, Dalbergia sissoo, Eucalyptus spp.*,andclass E was represented by *Acacianilotica, Acacia mangium, Delonix regia, Syzygium cumini.* In Class E *Acacia nilotica, A. mangium,Delonix regia, Syzygium cumini* found which is higher than other classes. *Cassia siamea, Cassiafistula, Dalbergia sissoo, Delonix regia* shows good regeneration in this site (Figure 3).

Figure 3 Population structure of tree species in west direction of urban setup



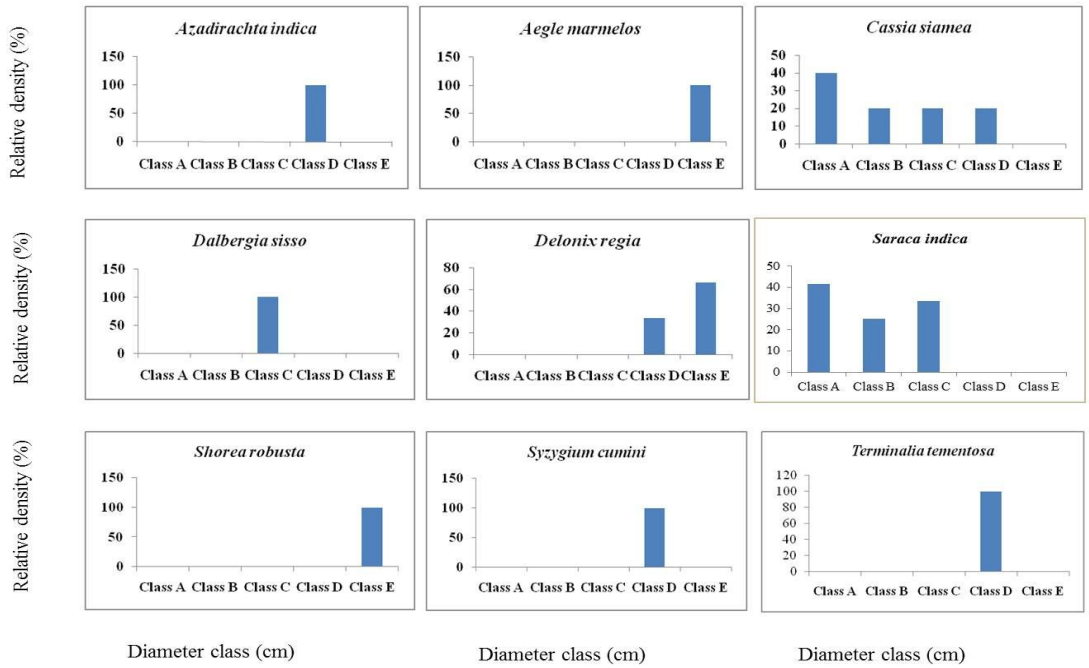
North direction (Figure 4) reflected class A was represented by *Butea monosperma, Cassia siamea, Cassiafistula, Delonix regia, Ficus benghalensis, Pongamia pinnata*, class B wasrepresented by *Cassia siamea, Cassia fistula, Tectona grandis*, class C was represented by *Bauhiniaracemosa, Eucalyptus spp., Dalbergia sissoo, Mangifera indica*, class D was represented by*Acacia mangium, Dalbergia sissoo, Eucalyptus spp., Mangifera indica*, Class E was representedby *Delonix regia, Melia azedarach,Tamarindus indica, Shorea robusta* and *Syzygium cumini*.

Figure 4 Population structure of tree species in north direction of urban setup



At south direction (Figure 5), class A was represented by *Cassia siamea* and *Saraca indica*, class Bwas represented by *Cassia siamea* and *Saraca indica*, class C was represented by *Cassia siamea,Dalbergia sissoo* and *Saraca indica*, class D was represented by *Azadirachta indica, Bombaxceiba, Cassia siamea, Delonix regia, Ficus racemosa, Syzygium cumini* and *Terminaliatomentosa*, Class E was represented by *Aegle marmelos, Delonix regia* and *Shorea robusta.*

Figure 5 Population structure of tree species in south direction of urban setup



During present study species representing single class towards east direction are *Delonix regia*representing class A, *Syzygium cumini* and *Terminalia tomentosa* representing class D, *Ficusbenghalensis*, *Mangifera indica*, *Shorea robusta* representing single class E. In west direction*Eucalyptus spp*. and *Azadirachta indica* representing class D while *Acacia mangium*, *Acacianilotica* and *Syzygium cumini* representing only class E. In north site *Butea monosperma*representing class A, *Acacia mangium* represents class D while *Azadirachta indica*, *Meliaazedarach*, *Shorea robusta* represent single class E. In south direction species showing singleclass were *Dalbergia sissoo* represents class C, *Azadirachata indica*, *Syzygium cumini*,*Terminalia tomentosa* represent class D while *Aegle marmelos*, *Shorea robusta* represents class E.Similar findings were also reported by Kittur et al. (2014). They reported that the highlydisturbed sites comprised of seedlings of size class (A) and saplings of size class (B), representedby very few species. Younger andolder trees were more abundant, whereasintermediate-aged trees were distributed sporadically (Jhariya et al. 2012). Similarly, Kumar et al (2017) reported the proportion of seedling size class (A) was found to be dominant while the older size classes (D) and (E) were totally absent in various directions.

Comparative family wise distribution of vegetation layer

A sum of 11 families was recorded for tree stratumamong which the family Fabaceae was found to be dominating followed by Combretaceaefamily (Figure 6). In Sapling layer only single family i.e., Fabaceae was found while in seedling layer 4 families were recorded and the Fabaceae was dominant family. Under shrub species,family Apocynaceae was found as dominant among the 7 families of shrubs. Herb layerreflects total 11familiesand the Asteraceae was the dominating family. Fabaceae family was dominant among the entire stratum in all the direction. Similarly, Pandey and Kumar (2018) also reported Fabaceae is dominant family among the 28 families listed.

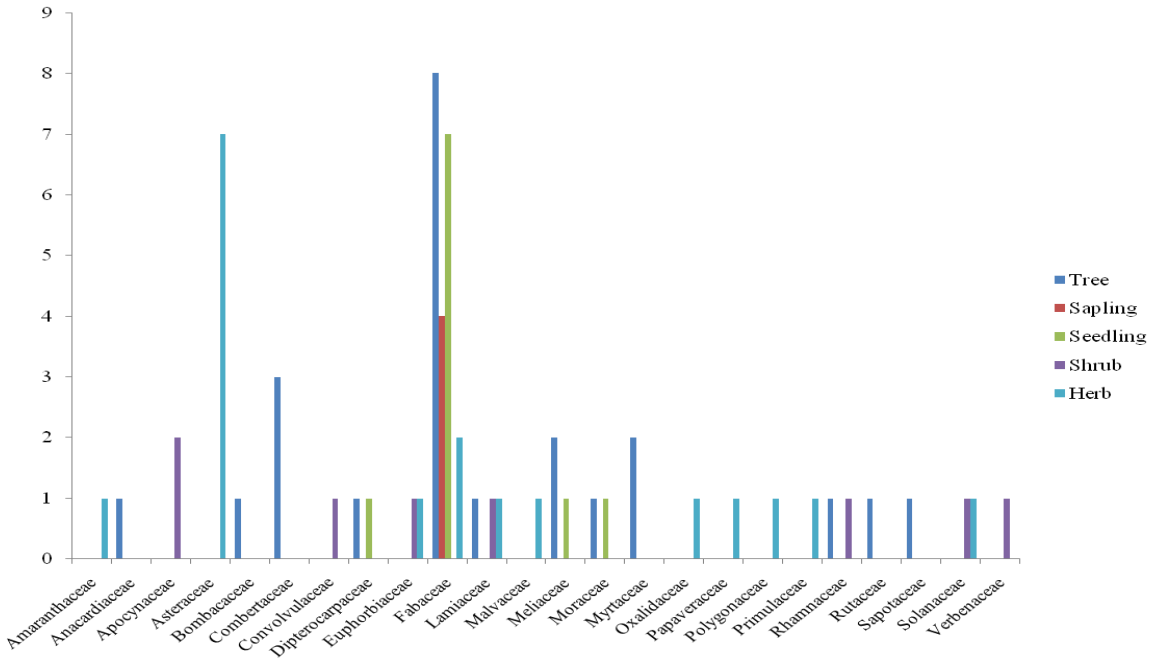


Figure 6. Family-wise distribution of vegetation in an urban setup of Sarguja

Species Regeneration Status

It reflected from the study that towards east direction only single species (*Delonix regia*) showed good regeneration potential among all the species. At west site good regeneration was revealed by *Cassia siamea*, *Cassia fistula*, *Delonix regia* and *Dalbergia sissoo*. North direction reflected that *Butea monosperma*, *Cassia fistula*, *Cassia siamea*, *Ficus racemosa* and *Pongamia pinnata* have good regeneration potential over the site. At south direction *Cassia siamea* and *Saraca indica* showed good regeneration potential among all the recorded species. The regeneration potential (in percentage) of various species found in the urban setup of Sarguja is shown in Figure 7. Towards east direction nearly 75% species were not regenerative, in west direction 60% species was not regenerative, in north direction about one third of the species not regenerative while in south the value of not regenerative species was higher (> 80%) than the other direction. The overall good regeneration of urban vegetation was recorded toward west direction followed by north, south and least in east direction. The present findings were corroborates with the earlier findings of Sarkar and

Devi (2014) and Jhariya and Oraon (2012). Similarly,Yadav and Jhariya (2017) reported that in natural forest nearly 11.77% species had good regeneration potential, 29.41% species fair regeneration while 35.29% species were not regenerating. In the plantation site 50% species showed good regeneration and remaining 50% species have fair regeneration status which supports the present findings.

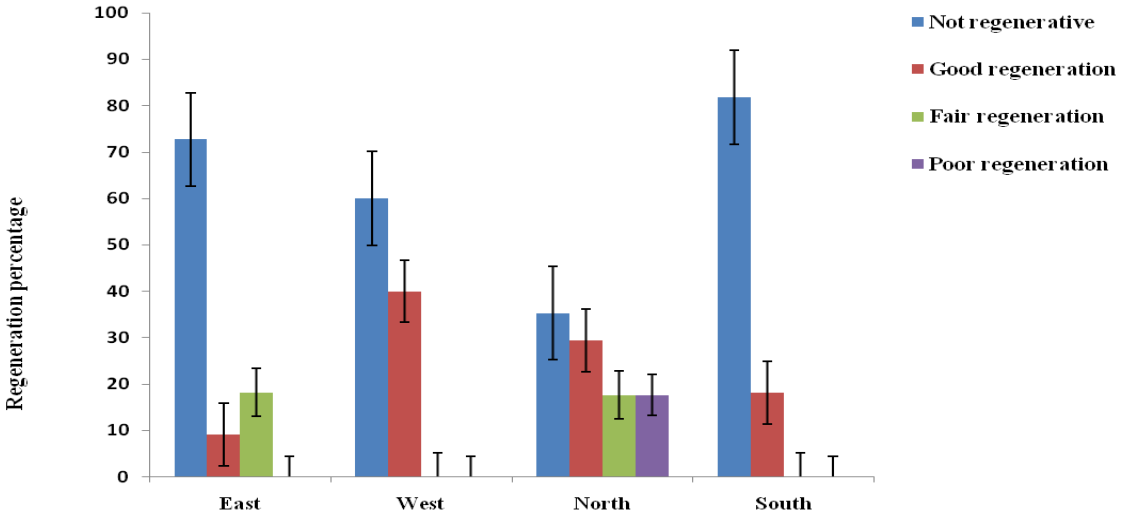


Figure7. Regenerationstatus (%) of urban vegetation in Sarguja

CONCLUSION

The vegetation in the urban setup is important from development, planning and towards sustainable cities. The vegetation in the present investigation shows substantial diversity and distribution across the sites in an urban setup. The species occurrence in terms of frequency revealed that most of the species showed rare category, while other classes were least in occurrence. These call for protection and proper management of rare species for the development of the future stands. The conservation implication should be implied on priority basis to promote the urban greening and urban forestry perspectives. Therefore, efforts should be more for ensuring health and diversity of urban vegetation in order to improve urban environment and to maintained environmental integrity and prosperity.

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RESEARCH ARTICLE

EFFECT OF PEG INDUCED WATER DEFICIT STRESS ON PHYSIO-BIOCHEMICAL CHARACTERISTICS OF DIFFERENT PEARL MILLET VARIETIES

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Abstract: The present study aimed to scrutinize six pearl millet varieties, differing in their drought sensitivity to evaluate drought tolerance through physio-biochemical parameters. The main purpose of this work was to screen the highly tolerant and susceptible genotypes under PEG-6000 induced water deficit stress (WDS). WDS was induced in seedling on 10th and 20 day of germination by exposing them to different stress levels *i.e.* T1 (Control); T2 (5% PEG) and T3 (10% PEG). Significant reductions in parameters viz. shoot length, root length, seedling vigour index I, seedling vigour index II and Membrane stability index was observed. The antioxidant enzyme activity (Catalase and Superoxide Dismutase) was assayed for these varieties under water stress. There was a profound decrease in the Catalase activity whereas the SOD activity was increased in the varieties selected for the study.The water stress induced by supplementing 5% PEG in soil was tolerable by the plants as compared to 10% PEG. The results obtained were useful in screening drought tolerant Pearl Millet genotype.

Keywords: WDS, PEG, Drought, Pearl millet, Enzyme activity

INTRODUCTION

Pearl millet (*Pennisetumglaucum*L.) is the most widely grown minor cereal crop world-wide among the millets. It is recognized as an important food and forage crop in many countries of Asia and Africa. As pearl millet can withstand drought and high temperature stress during either the vegetative or reproductive phases of its growth, hence is mostly preferred for arid and semiarid regions which experience frequent periods of dry weather. Currently, drought is one of the most important limiting factors for crop production and becoming an increasingly severe problem in many regions of the world (Aslam*et al.,* 2006). Leaf rolling, stomata closure, deeper penetration of roots, higher relative water content and better osmotic adjustment are some of the mechanism that plants employ to overcome water stress.

Since physiological responses of plants to drought stress may vary at different developmental stages, it is considered that different indicators should be used for the phenotyping of drought tolerance (Tuberosa, 2012).Various methods have been employed from time to time to identify drought tolerant genotypes and efforts have been made in the past to screen tomato varieties which differed in drought tolerance (George *et al.,* 2013).

The most popular approach for induction of drought stress is to use high molecular weight osmotic substances, like polyethylene glycol (PEG). It have been used often as abiotic stress inducer in many studies to screen drought tolerant germplasm (Jatoi *et al.,* 2014).It is a polymer and considered as better chemical to induce water stress artificially. PEG induced osmotic stress is inductee to decrease cell water potential. PEG is a non-penetrable and nontoxic osmotic substance which is used to lower the water potential and it has been used to simulate drought stress. It is commonly establishedplants that adapt PEG show high level of tolerance to drought stress as compared to cell lines that fall short under the induced stress condition.

Rajasthan is the major state with an area of 4.77 m. ha under pearl millet production. Since moisture loss from sandy soils of western Rajasthan is fast, sowing is completed within 2-3 days of the rainfall.Farmers of this region therefore tend to undertake risk in sowing even under sub-optimal conditions results in poor plant stand. Therefore developments of cultivars that germinate and produce vigorous stand under limited soil moisture are expected to contribute to successful pearl millet production in the arid regions.

In past few decades, research for identification of drought tolerant genotypes has taken an impetus with special reference to stress management. But pearl millet, although an important crop of Rajasthan has not been exploited in this regard further, the aim of this study was to investigate the effects of osmotic stress generated by different levels of PEG-6000 on seedling stage of pearl millet genotypes. The primary objective of the present study was to screen out the most tolerant and most sensitive pearl millet genotypes under artificially induced PEG drought stress.

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MATERIALS AND METHODS

Experimental materials

Seeds of six pearl millet genotypes with different sensitivity to water-deficit stress i.e. (MH 1996, MH 1998, MH 1993, MH 2024, RHB 177 and KBH108.) were obtained from Pearl millet Research Station, RARI, Durgapura, Jaipur.

Experimental details

The experiment was laid out in randomized complete block design with two factors (genotypes and water stress level) and three replications. Fifty seeds of each genotype were grown in plastic pots filled with coco peat. To achieve a uniform emergence of seedlings, pots were filled with potting mixture Seedling vigour index I at was measured by the formula given by Singh and Kakralya (1995).

SV I = Germination % × (mean of seedling length (root + shoot)/100)

Seedling vigour index was measured by the formula given by Singh and Kakralya (1995).

SV II =germination % × Seedling dry weight

Membrane Stability Index

The pearl millet membrane stability index (MSI) was determined according to the method of Premchand*et al.,* (1990) as modified by Sairam (1994). Shoot portion (0.1 g) of different treatments and control were thoroughly washed in running tap water and double distilled water and thereafter placed in10 ml of double distilled water at 40°C for 30 minutes. After the end of this period their electrical conductivity was recorded by EC meter (C1). Subsequently the same samples were placed on boiling water bath (100° C) for 10 min and their electrical conductivity was recorded as above (C2). The membrane stability index (MSI) was calculated as:

$$MSI = [1-(C1/C2)] \times 100$$

Catalase (CAT) enzyme activity:

It was assayed by measuring the disappearance of H_2O_2 according to Teranishi*et al.,* (1974). 1gm fresh plant material was taken and homogenized it in 50mM chilled phosphate buffer (pH 7.0) and then it was centrifuged. For CAT activity, the reaction mixture will contain 2.7 ml 50mM phosphate buffer (pH7.0), 0.1ml enzyme extract, 0.2 ml 200mM H_2O_2 solution. Decrease in absorbance will be recorded at 410 nm for 3 minutes.

leaving the top 5 cm empty, sowing and then covered with 2 cm of potting mixture. Plants were allowed to germinate and irrigated regularly. There were three treatments comprising T1: (control i.e. no PEG); T2 (5% PEG); T3: (10% PEG). The germination was calculated by incubating 10 seeds of each variety under three treatments T1, T2 and T3. The shoot and root length was measured with the help of meter scale and thread.Various physio-biochemical indices have been monitored at two stages of early growth, i.e. at 10 days after sowing and 20 days after sowing. The growth parameters- shoot length, root length, seedling vigour index I and II, membrane stability index, catalase and superoxide dismutase enzyme activity were recorded.

Seedling Vigour Index

Superoxide dismutase (SOD) enzyme activity:

1gm of plant material was homogenized in 50mM phosphate buffer+ 0.25 ml triton+ 1% PVPP and then centrifuged at 10000 rpm. For SOD activity the reaction mixture contain 1.5 ml reaction buffer, 0.2 ml methionine, 0.1 ml enzyme extract, 0.1 ml Na_2CO_3, 0.1 ml NBT , 0.1 ml EDTA and 0.1 ml Riboflavin. Three test tubes were taken.One contain all things and was kept in dark (Blank A), second test tube contain all things except enzyme extract and was kept in light (Blank B), third test tube contain all things and was kept in light (Blank C). Absorbance was recorded at 560 nm.One unit of SOD activity was defined as amount of enzyme which causes 50% inhibition of photochemical reaction of NBT.

Statistical Analysis

Data were subjected to analysis of variance (ANOVA) using SAS 9.1 software. The least significant differences (LSD at p = 0.05%) test was used to define significant differences among treatment means.

RESULTS

Effect of PEG on Germination

The various concentrations of PEG had a significant effect on the germination of the seeds. Germination was significantly affected by the osmotic potential, by cultivars and their interaction (Table 1). An increase in PEG stress markedly decreased the germination percentage of all cultivars compared to their relative controls. Genotype KBH108 and MH2024 showed 100% germination in 5% PEG induced water stress while MH1996 showed least germination in both 5% and 10% PEG induced stress.

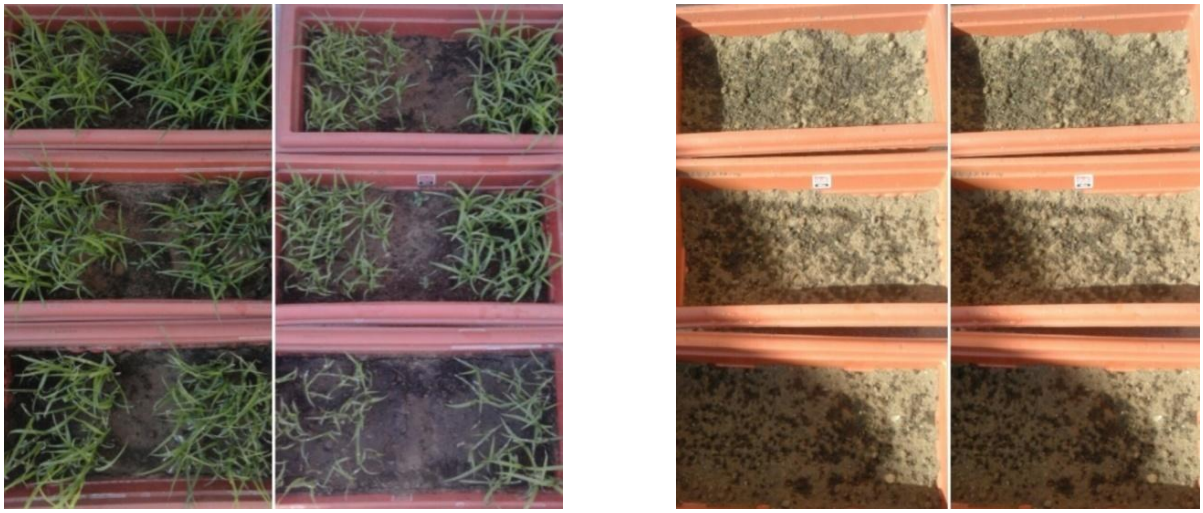

Figure 1: a) Pots before Germination b)Pots after Germination

Table 1. Effect of PEG treatments on germination of pearl millet hybrids:

S. No	Hybrids	T1	T2	T3
1	MH1996	80	70	70
2	MH1998	90	90	80
3	MH1993	100	80	80
4	MH2024	100	100	90
5	RHB177	100	90	70
6	KBH108	100	100	90

Effect of PEG on shoot and root length

For all genotypes, the shoot lengths of seedlings decreased with an increase in water stress (Table 2). 5% PEG treatment decreases the length of the coleoptile by 11.16 to 16.50% while 10%PEG reduces this dimension by 11.31 to 16.53% in all genotypes.GenotypeKBH 108 produced the longest shoot (17.4 cm) at control condition while showed highest reduction 16.50% and 16.53% at 5% and 10% PEG treatment respectively. While MH1996 showed lowest reduction 11.16% and 11.31% at 5 % and 10% PEG treatment respectively. PEG treatments decrease the root length of all the genotypes (Table2). In contrast, 10% PEG treatment increase the root length in all genotypes compared to 5% PEG.

Table 2. Effect of treatments on root length and shoot lengthof pearl millet hybrids:

	Shoot length			**Root length**		
Hybrids	**T1**	**T2**	**T3**	**T1**	**T2**	**T3**
MH1996	12.1	11.4	9.5	10.5	9.3	8.8
MH1998	15.7	15.2	14.4	11.6	10.8	9.9
MH1993	13.1	11.8	11.2	11.2	10.9	9.9
MH2024	13.3	12.9	11.9	14.9	11.4	10.9
RHB177	16.9	15	13.8	12.7	11.9	10.9
KBH108	17.4	15.7	15.2	14.6	13.1	12

Effect of PEG on Seedling vigor index I and Seedling vigor index II

Among the hybrids, MH 1996 was affected the least by drought stress because it gave the lowest reduction rate for seed vigour (Table 3). Seed vigour decreased with increase in concentration of PEG solution (Table 3). There were significant differences among hybrids for seed vigour in all drought levels. Among six pearl milletgenotypeKBH 108 produced the highest seed vigour and there were significant differences among hybrids for seed vigour in 5% and 10% PEG treatment.

Table 3. Effect on PEG on Seedling vigor index I and Seedling vigor index II of pearl millet hybrids

	Seedling vigour index I			**Seedling vigour index II**		
Hybrid	**T1**	**T2**	**T3**	**T1**	**T2**	**T3**
MH1996	18.08	16.56	12.81	2.78	2.48	1.61
MH1998	22.77	23.4	19.44	3.87	2.79	2.15
MH1993	24.3	22.7	18.99	3.81	2.36	1.44
MH2024	28.2	24.3	20.62	5.07	4.09	3.28
RHB177	29.6	24.21	17.29	4.74	4.14	2.29
KBH108	32	25.92	24.48	5.86	4.9	4

Figure 2: Effect of PEG induced water stress on Seedling length of Pearl millet hybrid

Effect of PEG on Membrane Stability Index:

As shown in table 4, the values for stability of cellular membranes in the leaf tissues studied revealed that there was a significant decline in MSI of stressed plants in all hybrids at both treatment level. A major impact of plant environmental stress is cellular membrane modification, which results in its perturbed function or total dysfunction. The cellular membrane dysfunction due to stress is well expressed in increased permeability and leakage of ions out, which can be readily measured by the efflux of electrolytes. The variety which shows maximum membrane stability index is KBH108 and minimum membrane stability index was observed in MH1996(Table 4).

Table 4. Effect of treatments on MSI of pearl millet hybrids

Hybrid	**T1**	**T2**	**T3**
MH 1996	67.33	60	53.67
MH1998	76	71	64.33
MH1993	77.67	74	65
MH2024	76.67	71	64.67
RHB177	74.67	68	62.33
KBH108	79.65	75	67.33

Effect of PEG on Catalase and Superoxide dismutase activity

In the present investigation as shown in figure 2, there was observed decrease in the CATactivity with the increase in the concentration of PEG creating water stress. In the present study, the CAT activity was found decreasing with the increasing PEG concentration as well as duration of drought induction. The variety which shows maximum antioxidant catalase activity is MH1996 and minimum catalase activity was observed in KBH 108.

In the present investigation as shown in figure 3, there was observed increase in the SOD activity with

the increase in the concentration of PEG creating water stress. SOD is a major scavenger of O_2 and its enzymatic action results in the formation of H_2O_2 and O_2. The SOD activity of leaves increases at both mild and severe stress condition as compare to the control plant leaves. The enhancement of SOD activity under stress condition shows a well organised defence system against ROS under stress condition. The variety which shows maximum SOD activity is KBH 108 and minimum SOD activity was observed in MH 1996.The readings wastaken in Umg^{-1} $prot^{-1}$.

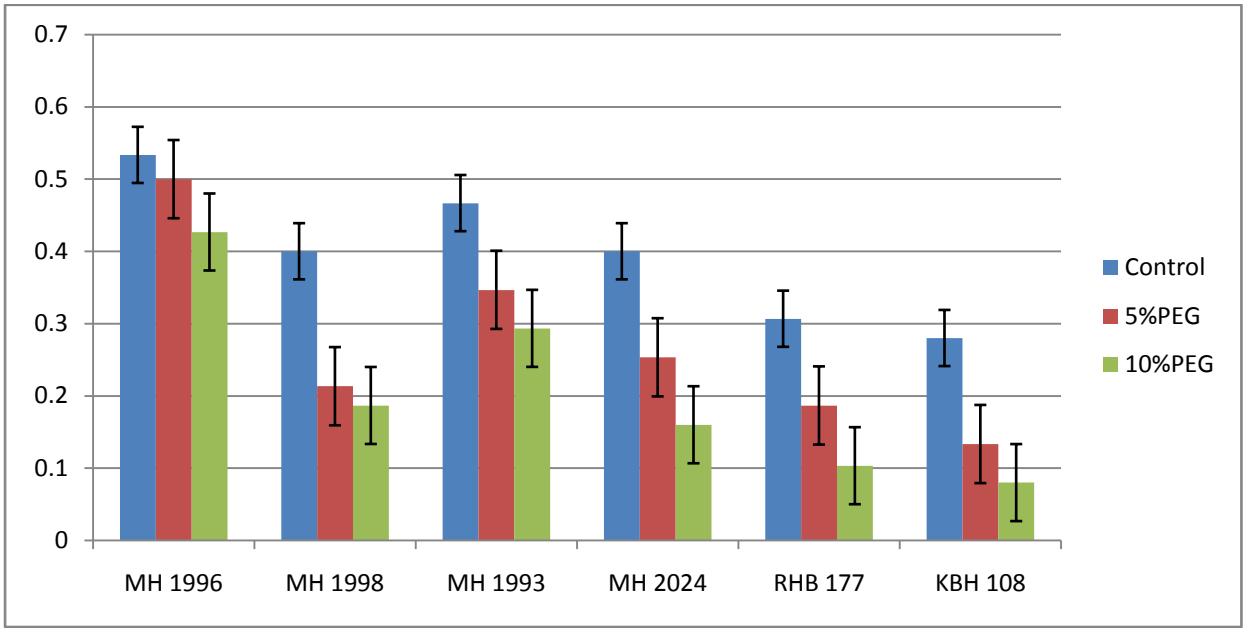

Figure3: Effect of PEG on Catalase activity of pearl millet hybrids.The bars indicates standard error (± SE) of mean (n = 3). All means are significantly different at $p \leq 0.05$.

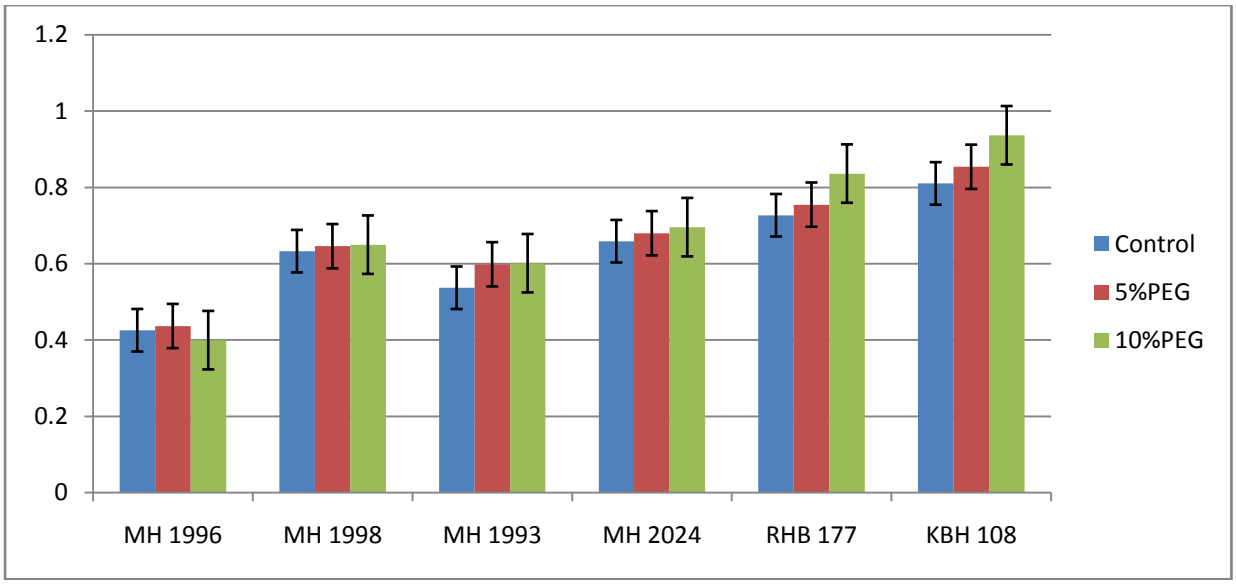

Figure4: Effect of PEG on Superoxide Dismutase activity of pearl millet hybrids. The bars indicates standard error (± SE) of mean (n = 3). All means are significantly different at $p \leq 0.05$.

DISCUSSION

Water stress due to drought is one of the most significant abiotic factors that limitthe seed germination, seeding growth, plants growth and yield (Hartmann *et al.*, 2005, Van den Berg and Zeng, 2006). Several methods have been developed to screen drought tolerant germplasm in plant species. Based on the literature available, PEG is considered as a superior chemical to induce water stress (Kaur *et al.*, 1998). Polyethylene glycol (PEG) molecules are inert, non-ionic, virtually impermeable chains and have been used frequently to induce water stress in crop plants (Carpita*et al.*, 1979; Landjeva*et al.*, 2008).Among investigated germplasm, KBH108 and MH2024 showed 100% germination in 5% PEG

induced water stress while MH1996 showed least germination in both 5% and 10% PEG induced stress than the other genotypes.The higher germination rates of the genotype may be due to theircapability to absorb water even under PEG induced water stress. Hearty, (1997) and Turk *et al.,* (2004) reported that water stress at germination stage delayed or reduced or hinder germination completely. However, once the grain attains a critical level of hydration it will lead to full seed germination. If, the physiological changes happen below the critical level it lead to complete inhibition of seed germination. Dodd and Donavon, (1999) stated that PEG induced reduction in germination percentage was because of reduction in the water potential gradient between seeds and their surroundings. Several reports on wheat varieties suggest that germination rate was affected by various abiotic stresses (Bayoumi*et al.,* 2008; Jajarmi, 2009; Alaei *et al.,* 2010). A higher level of germination under stress condition was observed in *Vignaaconitifolia*, however this finding may not be applicable to all cases and it depends on the germplasm used in screening (Soni *et al.,* 2011). Similar results like reduction in germination rate with the increase PEG were noted in chick pea also (Kaur *et al.,* 1998).

Strong negative correlation coefficient was noted between root length and PEG concentration. Roots are the primarily effected plant part under drought conditions than any other parts (Ghafoor, 2013). Root trait of all varieties provided useful information against different levels of PEG and this is very important attribute to study the drought stress. The germplasm which has better growth under stressed environment may have drought tolerance mechanism in it and these plants may have capability of holding a homeostasis under stressed conditions (Saxena and Toole, 2002). With few exceptions, the response of tomato varieties for root length was more or less similar against thedifferent levels of PEG. A gradual reduction in root length with an increasing concentration of PEG was the common tendency observed among all varieties. The reduction rate in root length is different in the varieties investigated. The root length at control varied in between 10.5 to 14.6 cm in the pearl millet genotype with mean root length 12.58 cm. At the highest concentration of PEG (10%) a drastic reduction in root length in all pearlmillet varieties was noted. It is well known fact that root architecture influences the yield and other agronomic traits, particularly under stress conditions (Ludlow and Muchow, 1990; Dorlodot*et al.,* 2007). Remarkable decrease in root length has been observed with increasing PEG concentrations was reported by Jajarmi*et al.,* (2009) and similar results like reduction in root length with increasing osmotic stress was identified in pea plants (Whalley*et al.,* 1998). Kulkarni and Deshpande, (2007) reported that early and rapid elongation of roots is a key trait of drought tolerance. A strong negative correlation between shoot length and PEG concentration has been observed and a positive correlation between shoot length and root length was identified and it clearly indicated that increase in root length helps in increase of shoot length. All the varieties showed common trend i.e. reduction rate in shoot length with increasing concentration of PEG. The decline in shoot length traits in response to induced osmatic stress is a commonly observed phenomenon which is depends on the tolerance capacity of the plant. Decreasing in growth rate with increasing osmatic stress was reported in several studies (Waseem *et al.,* 2006; Abdel- Raheem *et al.,* 2007; Aazami*et al.,* 2010).Higher MSI can be in generally considered as drought tolerant.Our results are in agreement with the findings of Sairam and Shrivastava, (2001) who reported that during stress there was a decrease in MSI irrespective of the genotypes. Geravandi*et al.,* (2011) demonstrated that drought tolerant genotypes contained higher MSI as compared to drought sensitive genotypes.Thegermplasm which is showing better performance can be considered as drought tolerant. Hence, germplasm with the capability of early vigour under stress conditions may be beneficial by increasing seedling competitiveness against weeds (Lemerle*et al.,* 2001). The early vigour of seedling with good development can be used as a trait of interest for the selection of tolerant germplasm (Richards, 2000; Botwright*et al.,* 2002). Root system with the ability of better growth under (Abdel-Raheem *et al.,* 2007). Siddique *et al.,* (1990) explained that plants with better early vigour can increase the crop water use efficiency. Contradictory results have been reported for activities of antioxidant enzymes in number of different plant species. These variations in antioxidant enzymes induced by stress not only depend on severity and duration of the stress treatment and also depend on species and age of the plant (Carvalho,2008). In our report, we observed that the activities of Catalasedecreased by increasing the PEG concentration, whereas increase in SOD activity were observed in both progressive stresses induced by PEG as compare to the control. An increase in SOD activity and decrease in CAT activity was also reported during drought stress in Liquorice (Pan *et al.,* 2006). It was reported that SOD as well as CAT activities increases in response to PEG induced drought stress in gerbera and Sesame (Lai *et al.,* 2007;Fazeli*et al.,* 2007). Deceased activity of SOD and CAT was reported in wheat subjected to long term field drought as well as PEG induced water deficit in wheat (Simova-Stoilova,*et al.,* 2007, Abdul *et al.,* 2017). Plants are well endowed with antioxidant molecules and scavenging systems which establish a link between tolerance to water stress and rise in antioxidant enzyme concentration in photosynthetic plants.

CONCLUSION

Drought is a foremost stress which decreases the production of crops worldwide (Iqbal *et al.*, 1999; Yang *et al.*, 2004). The problem is particularly very serious in arid and semi-arid regions (Ashraf *et al.*, 1995), where many developing and under developed countries are located. Pearl millet is the fourth most important cereal crop in India, after rice, wheat and sorghum, where it is widely grown in the states of Rajasthan, Maharashtra, Gujarat and Haryana where the food security of the poorest population depends vastly on pearl millet production. As in Rajasthan drought condition are very prevalent, so by our studies on "Effect of PEG induced water stress on different varieties of Pearl millet" we find out that which variety can grow best in such an drought condition.KBH 108 is found to be most stable crop that can withstand drought condition and will be best suited to be grown in Rajasthan followed by RHB 177 and MH 2024.

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RESEARCH ARTICLE

VEGETATION INTER-RELATIONSHIP AND REGENERATION STATUS IN TROPICAL FOREST STANDS OF CENTRAL INDIA

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Abstract: The regeneration status of the vegetation reflects the health of forest ecosystem. In this context, the present study was done in order to assess the rarity or commonness of the species along with regeneration status and species inter-relationship in tropical forest ecosystem. The study site of present investigation includes Achanakmar-Amarkantak Biosphere Reserve. Four forests stand *viz*., ddense, regenerating, medium and degraded forest having diverse vegetation attributes and representative of the region's vegetation were marked for the study. The permanent plot techniques were opted for enumeration through stratified random sampling method. The tropical forest studied reflects high rarity of the species in different sites under various stratified vegetation. Total 24 species distributed into 17 families were recorded in the study sites. The species commonness or high occurrence of the species is found to be negligible due dominance of species over the area. Regeneration status of different species showed drastic scenario in different forest stands. The good regeneration in different sites varied from 9.09-30.77% being lowest in degraded forest site and maximum in regenerated forest stand. The results revealed significant differences in species abundance, occurrence, regeneration status and inter-relationship in various forests stands. The screening of the species on the basis of abundance, regeneration status and positive or close inter-relationship between different species at site level can be utilized as effective tools for the scientific management, conservation and sustainable development of forest stands.

Keywords: Abundance, Inter-relationship, Rarity, Regeneration status, Vegetation stands

INTRODUCTION

The tropical forests are major natural resource having diverse variation in terms of floral and faunal biodiversity. These forests are subjected to alter due to various biotic and abiotic factors which accelerate the process of degradation of these landscapes throughout the world. In Indian context, these situations area more alarming due to population rise, urbanization, industrialization, resource dependency, land use change, changing climates, forest fragmentation and forest fire events (Kagezi et al. 2016;Yadav et al. 2017; Yadav and Jhariya, 2017; Jhariya and Yadav, 2018; Oraon et al. 2018; Jhariya et al. 2019).

The tropical forests are reported to have the diversity in vegetational life forms and great inherent self-recruitment ability to sustain these terrestrial ecosystems (Singh and Chaturvedi, 2017; Oraon and Jhariya, 2018). The natural forest stand development depends upon site condition, seed factors and regeneration status of the species under specific environmental conditions (Jhariya and Oraon, 2012). The renewal of forest is the natural events in which seedlings undergone through various processes towards establishing as a tree. The regeneration potential and its status is a detrimental for sustainable forestry towards management and conservation of these valuable forests resources.

The systematic and precise information on species status, abundance, occurrence, species inter-relationship in different vegetal layer at species and site level is insufficient and lacking. The depletion of species in a given area due to site specific disturbance causes improper natural recovery which declines the population dynamics in a stand and leads towards alterations in the ecosystems integrity (Singh et al. 2009; Singh and Chaturvedi, 2017; Jhariya et al. 2019). These problems of natural sustenance of vegetation need to be properly addressed and managed for healthy ecosystems. In this connection the present investigation deals with the species inter-relationship and regeneration status of different forest stands in a tropical ecosystem of Chhattisgarh, India.

MATERIALS AND METHODS

The present investigation was carried out at tropical forest of Chhattisgarh, India. The study site includes the Achanakmar-Amarkantak Biosphere Reserve. Four forests stand *viz*., dense, medium, regenerating and degraded forest stand having diverse vegetation attributes and representative of the region's vegetation were marked for the study. The area situated at 22º 15' - 22º 58' N latitude and 81º 25' - 82º 5' E longitude. The study region reflects tropical climate with average temperature of 17.2^{o}C (January) to 31.8^{o}C (May). The total average annual precipitation is about 1400 mm. The laterite, black and alluvial soil are characteristic features of the entire region (Yadav, 2016, 2018, 2019). The forest

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are mostly tropical (i.e., dry tropical and moist deciduous) over the entire region (Champion and Seth, 1968).

The stratified random sampling was done within the 1 hectare permanent plot in various forests stands. The trees were measured within 10m × 10m sized quadrats and girth at breast height (GBH) of individuals was measured at species level. For measuring saplings and seedlings, a quadrat of 2m × 2m within 10m × 10m quadrats at the center point was laid. The GBH of trees and saplings were recorded at 1.37 m from ground level. The seedling was measured above 10 cm from the ground level. The field data were subjected to various analyses *viz.*, frequency, density, abundance, and importance value index (IVI) through standard methods (Curtis and McIntosh, 1950; Phillips, 1959).

The species rarity or commonness of the species was calculated as the frequency class of the species (Raunkiaer, 1934; Hewit and Kellman, 2002). As per frequency classes, the species were categorized as A, B, C, D, and E, where A represents rare (0–20%), B represents low frequency (20–40%), C represents intermediate frequency (40–60%), D represents moderately high frequency (60–80%), and E represents high frequency or common (80–100%). The regeneration potential of urban vegetation was determined as per the Khan et al. (1987). The statistical interpretation (multivariate analysis) was done through two-way dendrogram to assess the inter-relationship of the different tree species in various forests stand by using MINITAB presents version 15.0 statistical computer software (Kumar et al. 2017; Jhariya 2017).

RESULTS AND DISCUSSION

Floristic status of vegetation

The family-wise distribution of the species across the sites was presented in Figure 1. A sum of 17 families includes 24 species were recorded over entire region of study sites. The distribution of species as per family reflected that the dominant family was Combretaceae (3 species) followed by Anacardeaceae, Caesalpiniaceae, Euphorbiaceae, Leguminaceae, Myrtaceae (2 species each). The families (11) having single species were Annonaceae, Bombicaceae, Burseraceae, Dipterocarpaceae, Ebnaceae, Lythraceae, Phyllanthaceae, Rhamnaceae, Samydaceae, Sapindaceae and Sapotaceae, respectively.

The present finding is supported by Rahman et al. (2010) they mentioned 22 species representing 17 families in the study sites. Total 12 species were reported by Gutierrez et al. (2004) during the investigation. Further they reported the disturbance regimes alter the stand scenario in term of species and plant family presence. Similarly, Pawar et al. (2014) reported the number of tree species in different sites ranged from 6-12.

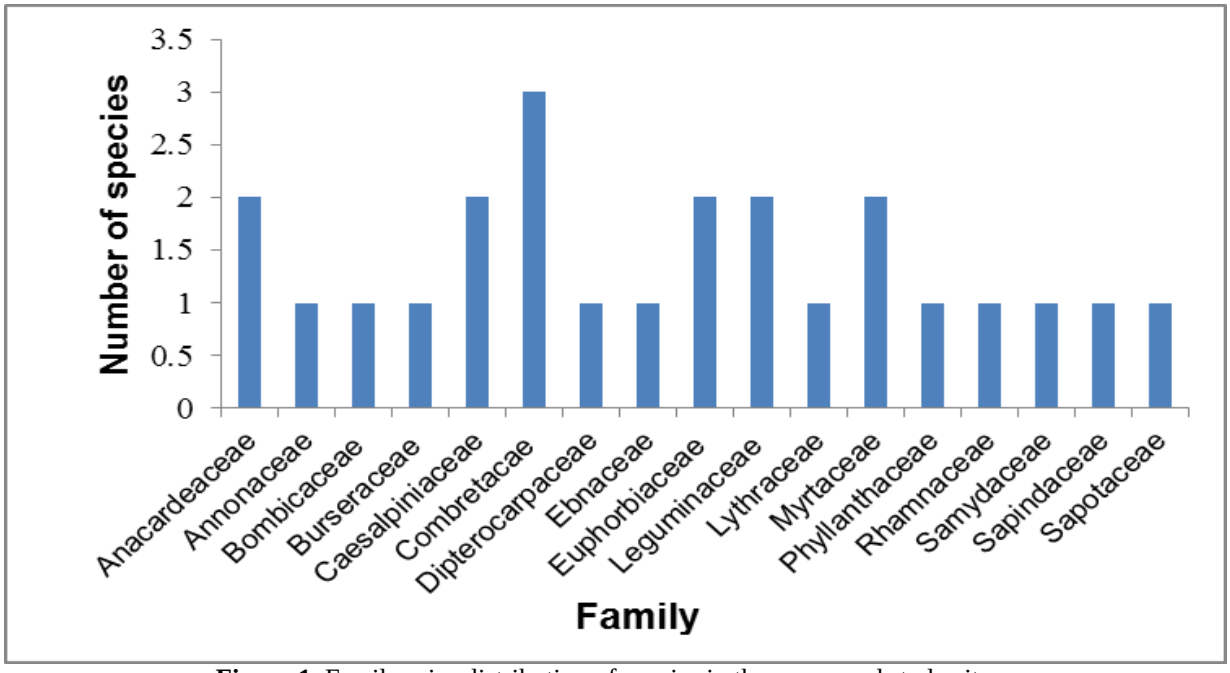

Figure 1. Family-wise distribution of species in the concerned study sites

Vegetation Abundance

The species occurrence and abundance of species under different vegetation stratum in various forest stand of tropical forest studied are given in Table 1 & Table 2.

Dense forest site reflected total 8 species under seedlings, out of which 5 species (62.5%) were rare in the studied site while remaining 37.5% (3 species) where showed low frequency class distribution. There is no species which reflects common, intermediate and moderately high frequency class in seedling

layer. Similar trend in sapling layer was recorded as 66.67% reflected rarity over that area while remaining showed low frequency class. The tree layer reflected 50% species were showed rarity over the area 37.50% species revealed low frequency class, 6.25% species reflected intermediate frequency and 6.25% species have common distribution over the area. At regenerated forest site total 5 seedling species were recorded, of which 40% species were rare, 40% species showed low frequency class and 20% species recorded intermediate frequency class. The sapling layer showed almost similar trends as in case of seedling. Tree layer revealed that 83.33% species showed rarity over the area while remaining 16.67% species were showed common occurrence. The medium forest site, in seedling stratum cent percent species showed rarity over the study site, whereas in case of saplings 66.67% species showed rarity while remaining species reported the low frequency in the medium forest site. In tree layer 66.67% reported rarity in the site, 22.22% species showed low frequency class and 11.11% reported under species commonness. At degraded forest site 3 out of 4 species showed rarity under the seedling layer, cent percent species reported rarity under sapling layer while in tree layer 4 out of 7 species showed rarity. In this site not a single species under seedling, sapling and trees were recorded under the category of moderately high to common in occurrence.

The abundance of the species depends on the site condition, prevailing environmental factors as well as various natural and anthropogenic factors in a given localities. It is reported that in various ecosystems most of the species under different plant life forms showed more occurrence of species individually. The similar trend was also reflected during the present investigation that the most of the species showed rarity over the area under different forest stands (Raunkiaer, 1934; Hewit and Kellman, 2002; Kumar *et al*. 2017; Jhariya, 2017; Oraon and Jhariya, 2018).

Vegetational inter-relationship in different forest stand

Figure 2 shows the inter-relationship of tree species between various phytosociological parameter (density, IVI and basal area) of different species found in dense forest stands. It reflects variable pattern of clustering which revealed some species have smaller cluster while some distant clustering pattern. The smaller clustering between species and various parameters showed their positive inter-relationship and association between them. In dense forest stand the smaller clustering were framed between *Diospyros melanoxylon* and *Buchanania lanzan*, *Lannea grandis* and *Miliusa tomentosa*, *Lagerstroemia parviflora* and *Anogeissus latifolia*.

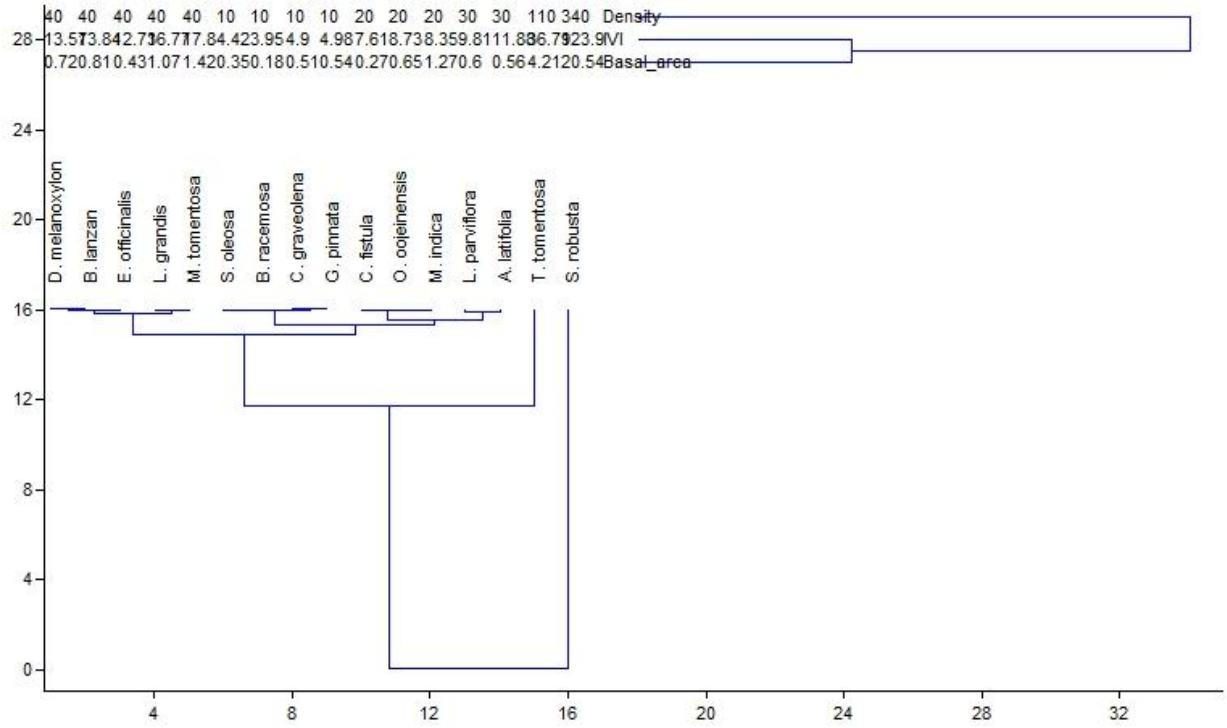

Figure 2 Clustering of various phytosociological parameter of tree layer of dense forest site

Table 1. Occurrence of the species as per the rarity and commonness scheme at tropical forest of Chhattisgarh

Species	Dense Forest Site			Regenerated Forest Site			Medium Forest Site			Degraded Forest Site		
	Seedlings	Saplings	Trees	Seedlings	Saplings	Trees	Seedlings	Saplings	Trees	Seedlings	Saplings	Trees
Anogeissus latifolia Wall. ex Bedd	10 (A)	0	30 (B)	0	0	10 (A)	0	0	10 (A)	0	0	10 (A)
Bauhinia racemosa Lam.	0	0	10 (A)	--	--	--	--	--	--	--	--	--
Bombax malabaricum Linn.	--	--	--	--	--	--	--	--	--	0	0	20 (A)
*Buchanania lanzan S*preng,	10 (A)	0	30 (B)	0	0	10 (A)	--	--	--	0	0	30 (B)
Careya arborea Roxb.	--	--	--	--	--	--	0	0	20 (A)	--	--	--
Casearia graveolens	30 (B)	10 (A)	10 (A)	30 (B)	40 (B)	10 (A)	10 (A)	20 (A)	0	30 (B)	10 (A)	0
Cassia fistula Linn.	0	0	20 (A)	--	--	--	--	--	--	--	--	--
Cleistenthus collinus (Roxb) Benth & Hook.	--	--	--	0	0	10 (A)	--	--	--	--	--	--
Diospyros melanoxylon Roxb.	30 (B)	0	30 (B)	50 (C)	60 (C)	10 (A)	10 (A)	10 (A)	10 (A)	10 (A)	0	40 (B)
Emblica officinalis Gaerth,	0	0	30 (B)	--	--	--	--	--	--	--	--	--
Garuga pinnata Roxb.	0	0	10 (A)	--	--	--	--	--	--	--	--	--
Lagerstroemia parviflora Roxb.	0	0	20 (A)	--	--	--	10 (A)	0	10 (A)	--	--	--
Lannea grandis Engl.	0	0	40 (B)	0	0	20 (A)	0	0	10 (A)	--	--	--
Madhuca indica J.F. Gmel.	0	0	10 (A)	--	--	--	--	--	--	0	0	20 (A)
Mallotus philipensis	10 (A)	30 (B)	0	20 (A)	10 (A)	0	--	--	--	--	--	--
Miliusa tomentosa (Roxb.) J.Sinclair	0	20 (A)	40 (B)	--	--	--	--	--	--	--	--	--
Ougeinia oojeinensis (Roxb.) Hochr.	0	0	20 (A)	0	0	20 (A)	--	--	--	--	--	--
Pterocarpus marsupium Roxb	--	--	--	0	0	10 (A)	--	--	--	0	0	20 (A)
Schleichera oleosa (Lour) Oken.	0	0	10 (A)	--	--	--	--	--	--	--	--	--
Shorea robusta Gaertn F.	40 (B)	0	100 (E)	40 (B)	10 (A)	100 (E)	20 (A)	0	100 (E)	10 (A)	0	0
Syzygium cumini	10 (A)	0	0	0	0	10 (A)	10 (A)	40 (B)	10 (A)	0	20 (A)	0
Terminalia chebula Retz.	--	--	--	0	0	10 (A)	--	--	--	--	--	--
Terminalia tomentosa Wt & Ang.	10 (A)	0	50 (C)	10 (A)	0	90 (E)	0	0	30 (B)	0	0	50 (C)
Zizyphus xylopyra Willd	--	--	--	--	--	--	0	0	30 (B)	10 (A)	0	0

Table 2. Species distribution as per frequency class in a tropical forest of Chhattisgarh

Study site	Vegetation Layer	Class A (Rare)	Class B (Low)	Class C (Intermediate)	Class D (Moderately High)	Class E (Common)
Dense Forest Site	Seedling	5	3	0	0	0
	Sapling	2	1	0	0	0
	Tree	8	6	1	0	1
Regenerated Forest Site	Seedling	2	2	1	0	0
	Sapling	2	1	1	0	0
	Tree	10	0	0	0	2
Medium Forest Site	Seedling	5	0	0	0	0
	Sapling	2	1	0	0	0
	Tree	6	2	0	0	1
Degraded Forest Site	Seedling	3	1	0	0	0
	Sapling	2	0	0	0	0
	Tree	4	2	1	0	0

Table 3. Regeneration status of species at tropical forest of Chhattisgarh

Species	Dense Forest Site				Regenerated Forest Site				Medium Forest Site				Degraded Forest Site			
	Seedlings	Saplings	Trees	Regeneration Status	Seedlings	Saplings	Trees	Regeneration Status	Seedlings	Saplings	Trees	Regeneration Status	Seedlings	Saplings	Trees	Regeneration Status
Anogeissus latifolia Wall. ex Bedd	2500	0	30	FR	0	0	10	NR	0	0	10	NR	0	0	10	NR
Bauhinia racemosa Lam.	0	0	10	NR	--	--	--	--	--	--	--	--	--	--	--	--
Bombax malabaricum Linn.	--	--	--	--	--	--	--	--	--	--	--	--	0	0	20	NR
*Buchanania lanzan S*preng,	5000	0	40	FR	0	0	10	NR	--	--	--	--	0	0	40	NR
Careya arborea Roxb.	--	--	--	--	--	--	--	--	0	0	20	NR	--	--	--	--
Casearia graveolens	7500	2500	10	GR	10000	12500	10	GR	10000	7500	0	GR	17500	2500	0	GR
Cassia fistula Linn.	0	0	20	NR	--	--	--	--	--	--	--	--	--	--	--	--
Cleistenthus collinus (Roxb) Benth & Hook.	--	--	--	--	0	0	10	NR	--	--	--	--	--	--	--	--
Diospyros melanoxylon Roxb.	10000	0	40	FR	27500	15000	10	GR	2500	2500	10	GR	2500	0	70	FR
Emblica officinalis Gaerth,	0	0	40	NR	--	--	--	--	--	--	--	--	--	--	--	--
Garuga pinnata Roxb.	0	0	10	NR	--	--	--	--	--	--	--	--	--	--	--	--
Lagerstroemia parviflora Roxb.	0	0	30	NR	--	--	--	--	2500	0	10	FR	--	--	--	--
Lannea grandis Engl.	0	0	40	NR	0	0	20	NR	0	0	10	NR	--	--	--	--
Madhuca indica J.F. Gmel.	0	0	20	NR	--	--	--	--	--	--	--		0	0	50	NR
Mallotus philipensis	2500	20000	0	GR	10000	5000	0	GR	--	--	--	--	--	--	--	--
Miliusa tomentosa (Roxb.) J.Sinclair	0	5000	40	PR	--	--	--	--	--	--	--	--	--	--	--	--
Ougeinia oojeinensis (Roxb.) Hochr.	0	0	20	NR	0	0	40	NR	--	--	--	--	--	--	--	--
Pterocarpus marsupium Roxb	--	--	--	--	0	0	10	NR	--	--	--	--	0	0	20	NR
Schleichera oleosa (Lour) Oken.	0	0	10	NR	--	--	--	--	--	--	--	--	--	--	--	--
Shorea robusta Gaertn F.	32500	0	340	FR	40000	2500	500	GR	15000	0	470	FR	2500	0	0	FR
Syzygium cumini	5000	0	0	FR	0	0	10	NR	2500	10000	10	GR	0	5000	0	PR
Terminalia chebula Retz.	--	--	--	--	0	0	10	NR	--	--	--	--	--	--	--	--
Terminalia tomentosa Wt & Ang.	2500	0	110	FR	5000	0	120	FR	0	0	40	NR	0	0	50	NR
Zizyphus xylopyra Willd	--	--	--	--	--	--	--	--	0	0	30	NR	2500	0	0	FR

Table 4. Regeneration categorization of species (%) in different sites in tropical forest of Chhattisgarh

Status	Dense Forest Site	Regenerated Forest Site	Medium Forest Site	Degraded Forest Site
Good Regeneration	11.11	30.77	30.0	9.09
Fair Regeneration	33.33	7.69	20.0	27.27
Poor Regeneration	5.56	0.0	0.0	9.09
Not Regenerating	50.0	61.54	50.0	54.55

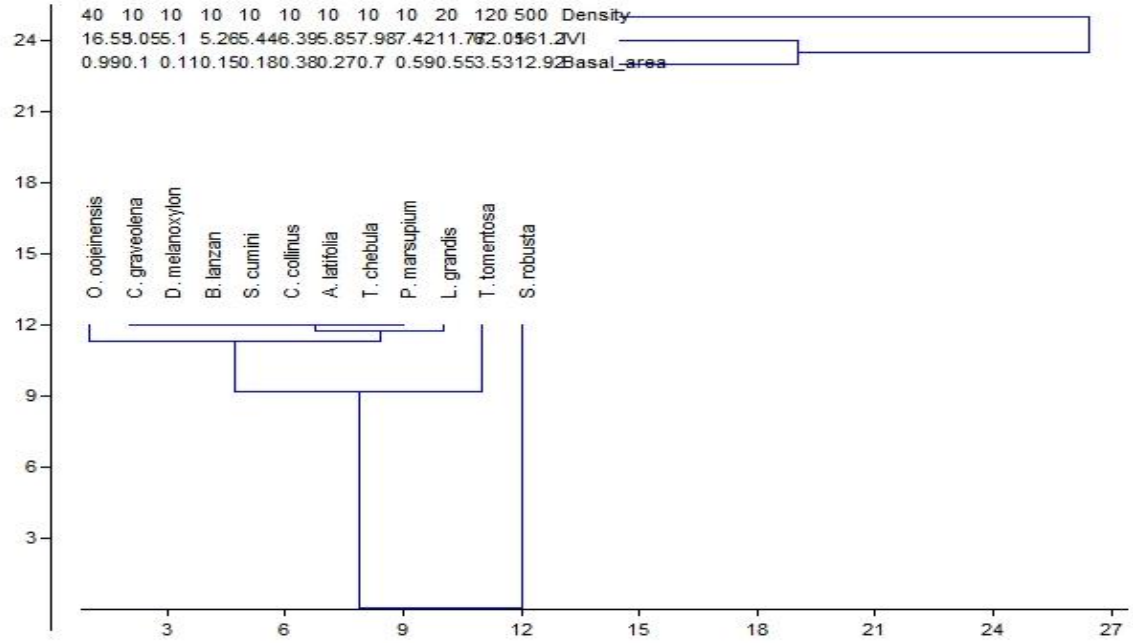

Figure 3 Clustering of various phytosociological parameter of tree layer of regenerated forest site

Figure 3 showed various clumpy clustering between various species, while the *Shorea robusta* revealed distant position among all the species recorded in regenerated forests stand. The cluster analysis for various phytosociological parameters in medium forest stand (Figure 4) revealed similar trend in case of the *Shorea robusta* while the small clustering were framed between *Terminalia tomentosa* and *Zizyphus xylopyra*.

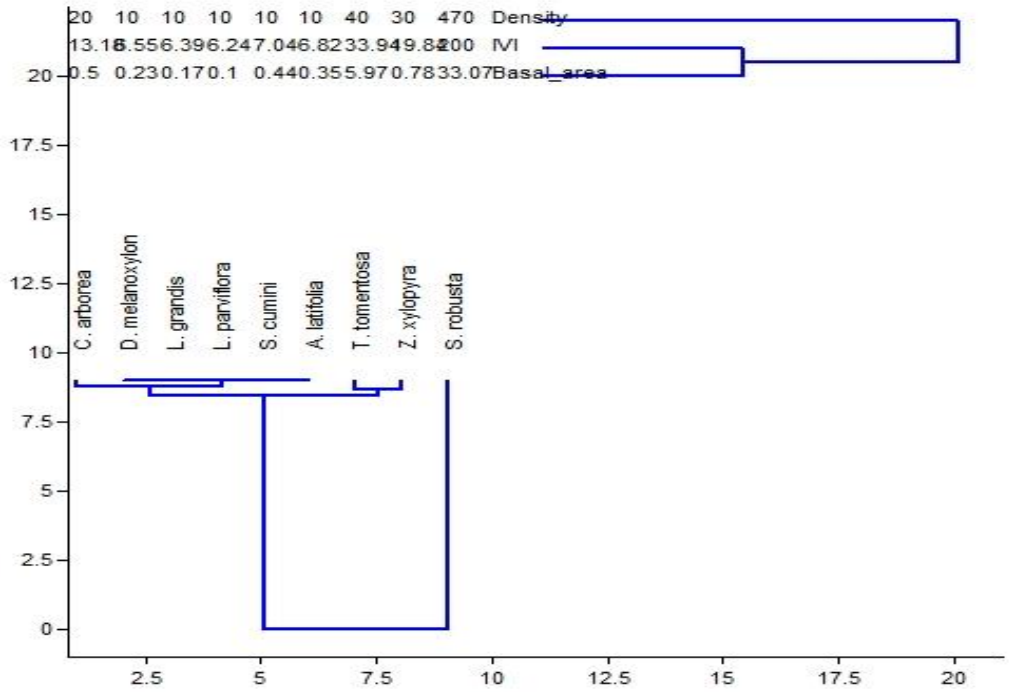

Figure 4 Clustering of various phytosociological parameter of tree layer of medium forest site

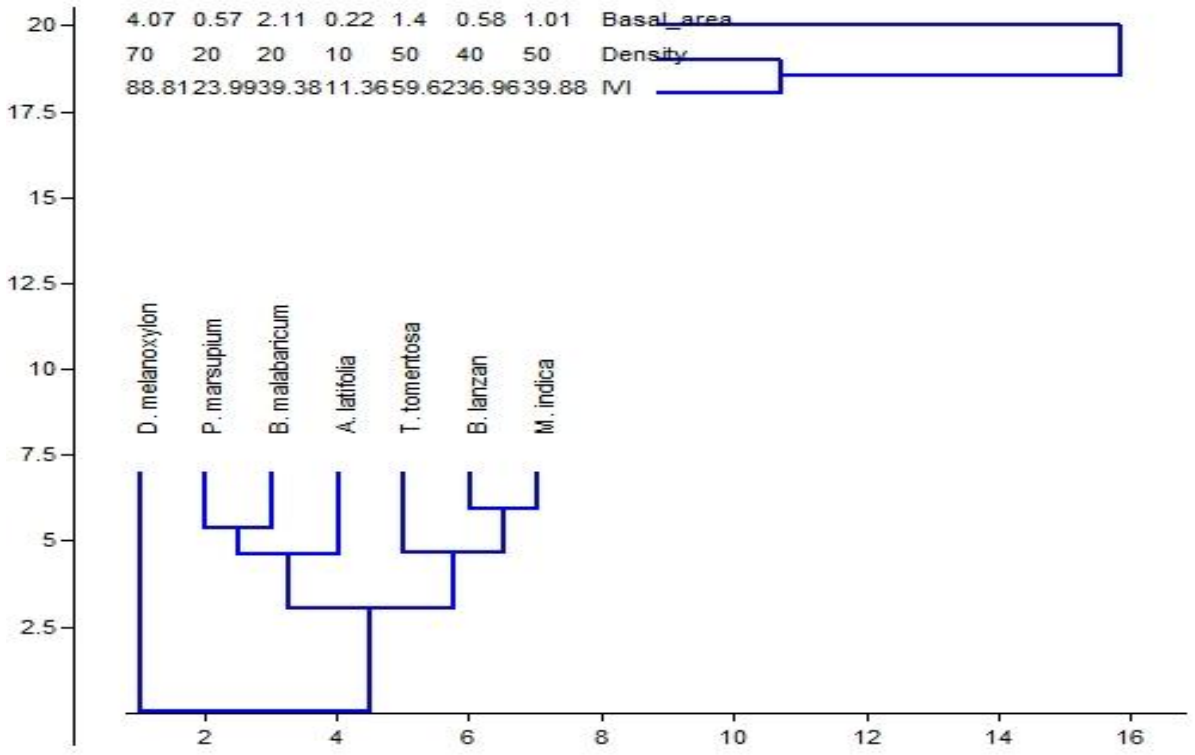

Figure 5 Clustering of various phytosociological parameter of tree layer of degraded forest site

Figure 5 showed small clustering between *Pterocarpus marsupium* and *Bombax malabaricum*, *Buchanania lanzan* and *Madhuca indica*, whereas *Diospyros melanoxylon* reflected distant position among all the species.

Clustering of various phytosociological parameters of tree layer in different forest stands showed significant level of variation among them. Similarly, Eni et al. (2012) mentioned utility of such analysis for establishing the inter-relationship of forest vegetation in Nigeria. Further such analysis work as fact finding tools and reduces the data complexity to find out the key species, parameters or attributes which have significant contribution and inter-relationship between them (Kumar et al. 2017; Jhariya, 2017).

Vegetation regeneration status

The regeneration of species and its overall scenario in different forest stands under tropical condition is reported in Table 3 & 4. In dense forest site 6 species showed fair regeneration 2 species good regeneration, 1 species poor regeneration and 9 species not regenerating. At regenerated forest stand 1 species reflected fair regeneration, 4 species good regeneration and 8 species were not regenerating. In case of medium forest stands 2 species showed fair regeneration, 3 species good regeneration and 5 species were not regenerating. Degraded forest stands showed 3 species under fair regeneration, 1 species each in good regeneration and poor regeneration as well as 6 species under not regenerating condition. The regeneration scenario revealed that the regenerated forest have highest regeneration potential (30.77%) followed by medium forest stand (30.0%), dense forest stand (11.11%) and least at degraded forest stands (9.09%). It evident from the study that in all the forest stands nearly half or more than half of the species were not regenerating.

Regeneration of various species in a given forest stands is crucial factors determining the future stand development, stand stocking and quality, and health of the sustained forest in terms of stand volume, biomass and productivity (Pawar et al. 2012; Jhariya, 2014). The transformation rate of species into different vegetal layers viz., seedlings into saplings, and saplings into trees are drastically reduced in different forests stands as confirmed by the more than half of the species are not regenerating under specific environmental conditions. Similar trend were also mentioned by various workers under similar types of forest stands (Good and Good, 1972; Pawar et al. 2012; Jhariya and Oraon, 2012; Oraon and Jhariya, 2018).

CONCLUSION

Enormous diversity of vegetation in different forest stands were recorded in terms of species and family distributed over the area. The degraded forest revealed lesser diversity and abundance of species, family, density, basal area and regeneration status in relation to other forests stand. Moreover, the pooled data reflected that nearly 65.38% species were rare in occurrence followed by 24.36% species have low occurrence, 5.13% species each showed intermediate and commonness over the area while moderately high frequency class reflects no presence of an species

across the sites under different vegetal layers. This therefore, highlights the conservation priority of the rare species otherwise it may be collapsed due to improper regeneration and other ecological constraints. Besides this the species having good regeneration (*Casearia graveolens*, *Diospyros melanoxylon*, *Mallotus philipensis*, *Shorea robusta* and *Syzygium cumini*) can be selected for the further reforestation and afforestation plans due to its greater adaptability and wider ecological amplitude which accelerate the sustainable development these forest stands.

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RESEARCH ARTICLE

DYE YIELDING PLANTS OF BARWANI DISTRICT, MADHYA PRADESH

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Abstract: Natural dyes are colours obtained from plants, invertebrates or minerals. Vegetable dyes from angiosperm plant sources comprise major part of natural dyes. Further, other biological sources such as fungi and lichens also contribute in the production of natural dyes. Due to the discovery of synthetic dyes in nineteenth century a marked decline in the use of natural dyes was experienced. Now-a-days extraction and use of natural dyes is confined as traditional knowledge (TK) among the rural people of few villages only. Barwani district in Madhya Pradesh has few areas where this TK is still in practice among the villagers. District is situated on the south-west part of Madhya Pradesh and lies between 21°37'N-74°27'E and 22°22'N-75°30'E. In the present communication TK available with the villagers in Barwani district is documented using semi-structured questionnaire. During field survey in the study area, 11 plant species are recorded which are used as a source of natural dyes was experienced. Now-a-days extraction and use of natural dyes in confined as traditional knowledge (TK) among the rural people of few villages only.

Keywords: Natural dyes plant, Indigenous knowledge, Medicinal uses, Angiosperm

INTRODUCTION

Natural dyes are obtained from animal or plant material without any chemical treatment. Natural dyes have several advantages over the synthetic dyes like their biodegradable and non-toxic nature, environment friendly and aesthetically appealing properties. Easy extraction of colour by boiling the plants, berries, leaves, bark or flower heads in water increases the acceptability of natural dyes. The Madhya Pradesh state in the heart of India, is very rich in biodiversity as well as in the forest cover. Barwani district in Madhya Pradesh has such few areas where this TK is still available among the villager's. District is situated on the South – West part of Madhya Pradesh. In the human civilization plants are used not only as the basic needs of life such as food, fiber, fuel, cloths and shelter but also as sources of natural dyes for dying cloths, designs and painting. The present study was undertaken to assess the diversity of dye-yielding plants of Barwani district. Now-a-days extraction and use of natural dyes is confined as traditional knowledge and will also be helpful in understanding the social and cultural life of tribes of this region.

Available literature shows that several studies were carried out on dye yielding plants in the recent past (Rashmi et al., 2004, Debajit and Tiwari 2005, Shiva 2007, Purohit et al., 2007, Gour 2008 and Garg et al., 2010). However, dye yielding plants are not properly studied with reference to Madhya Pradesh (Tiwari and Bharat 2008; Choudhary and Upadhyay 2011).

Study area

The name of Badwani originated from the forest of 'Bad' (*Ficus bengalensis*) which has surrounded the city in old times, 'wani' is the old word for the garden, therefore the city got the name 'Badwani' which means 'Gardens of Bads'. Badwani is situated on the South-West corner of Madhya Pradesh. The district lies between 21°37/ and 22/ North latitudes and 74º 27/ and 75º 30/ East longitudes. The district is triangular in shape with the highest point in the West. The total geographical area of the district is 3665 Sq.km. out of which forests occupy 1875.88 Sq. km. (51%) of area. The present study was conducted in Barwani District from March 2011 to Dec. 2012 to gather information on plants used by locals in traditional way.

MATERIALS AND METHODS

Extensive field surveys were undertaken in various localities of Badwani district and information was recorded on various aspects of dye-yielding resources, along with their other ethnobotanical information. The detailed information of dye-yielding plants has been recorded through observation and personal interviews with old and elderly people of the study area. Around 30 informants, belonging to diverse fields were interviewed. Besides personal interviews, relevant literature was also consulted. Photographs of plant specimens have been taken. Various relevant floras were consulted for identification of plant specimens.

RESULTS

The plants, with their parts used for dyeing purpose and specificity in application are enumerated in Table 1. Specimens of all these plants were collected and deposited in the herbarium Department of Botany, P.M.B. Gujrati Science College, University of Devi Ahilya University, Indore. It has been

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observed that the people of few areas like Toranmal, Nagalwadi, Varla and Dhavali of Badwani District still use plants for obtaining natural dyes. In the studied area, about 11 dye-yielding arboreal species belonging to 8 families have been identified which are generally used by the people to obtain dyes (Fig. 3). Each species is provided with correct local name, botanical name, family, dye-yielding parts and colour obtained (Table-1). Fabaceae and Euphorbiaceae are the two angiosperm families which contributed maximum plants as dye yielding species. Among the collected species, bark of two species, flowers of 04 species, fruits of 01 species, leaves of 03 species and bark as well as flowers of 01 species are used to get natural dyes.

Table 1. Important natural dye yielding plants with application specificity

S. No.	**Plants Species / Family / Vernacular Name**	**Plant parts used**	**Colour/hue of dye**	**Application specificity**
1.	*Ampelocissus latifolia* (Roxb.) Planch. Family : Vitaceae Vern. name: Amrola	Leaves	Green	Dye obtained from this plant species is used to dyeing textiles clothes etc.
2.	*Ziziphus xylopyrus* (Retz.) Willd. Family : Rhamnaceae Vern. Ghatbor	Bark	Pink	Bark is used as mordant in silk dyeing.
3.	*Butea monosperma* (Lam.) Taub. Family : Fabaceae Vern. Palasa	Bark, Flowers	Yellow, Orange	Flowers yield deep yellowish orange dye used for colouring clothes and other decorative purposes.
4.	*Indigofera tinctoria* L. Family : Fabaceae Ver. Neel	Leaves	Blue	The dye obtained is used to dye cotton clothes.
5.	*Acacia catechu* (L.f.) Willd. Family : Mimosaceae Vern. Katha	Heartwood	Reddish Black/ Brown Yellow	A fast reddish black dye is obtained from the heartwood by boiling in water for about one hour. The dye is used in the preparation of local ink and dyeing cotton fabrics.
6.	*Nyctanthes arbor-tristis* L. Family : Oleaceae Vern. Harsingar	Flowers	Yellow	Flowers tube contains on orange coolouring matter 'hydathin' which is used in colouring of silk. It is also useful in printing purposes.
7.	*Wrightia tinctoria* R.Br. Family : Apocynaceae Vern. Dhudi	Flowers	Pink	An adjuvant in dyeing.
8.	*Justicia adhatoda* L. Family : Acanthaceae Vern. Dudi	Flowers	Yellow	The flowers are used to get a yellow dye for fabrics however; alum is used as mordant in the process.
9.	*Woodflordia fruticosa* (L.) Kurz Family : Lythraceae Vern. Dhawai	Flowers	Pink / Red	Flowers yield a brick red dye which is used for colouring rope materials, cloths etc.
10.	*Mallotus philippensis* (Lam.) Mull.Arg. Family : Euphorbiaceae Vern. Sindhuri	Fruits	Red	Fruits are used to get a dye for silk.
11.	*Phyllanthus emblica* L. Family : Euphorbiaceae Vern. Aonwla	Leaves	Black / Grey	A black dye is obtained from the bark or fruits which are soaked in water for 4 to 5 days or boiled. The dye is used for dying fishing nets.

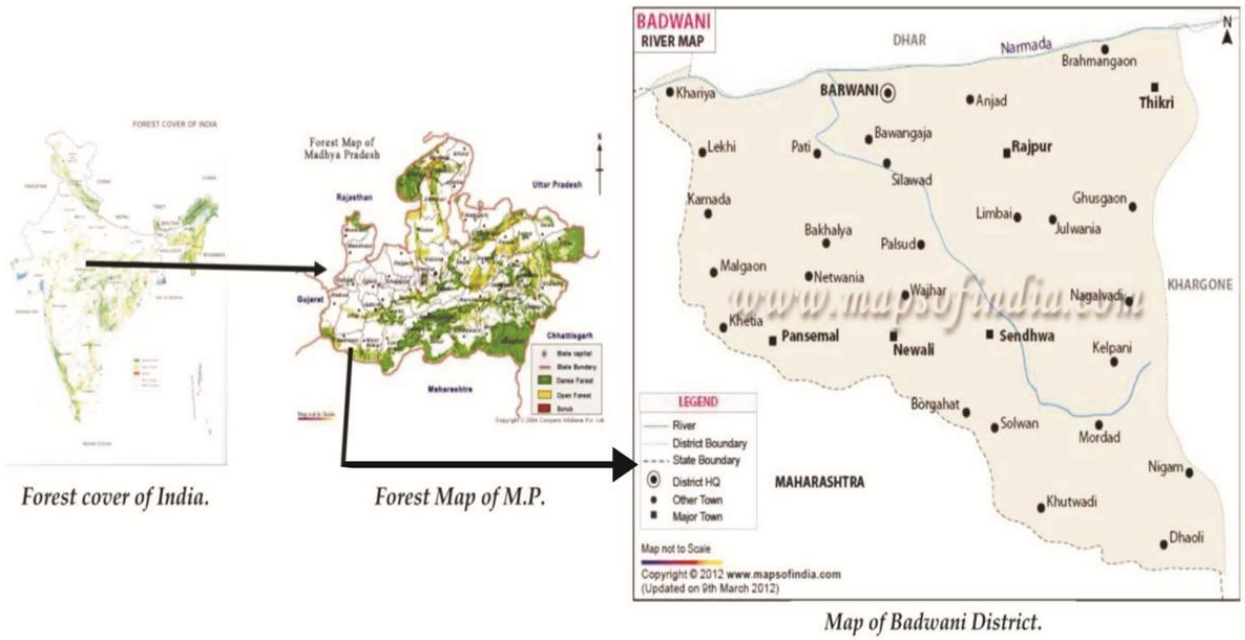

Fig. 1: Map of Badwani District in India.

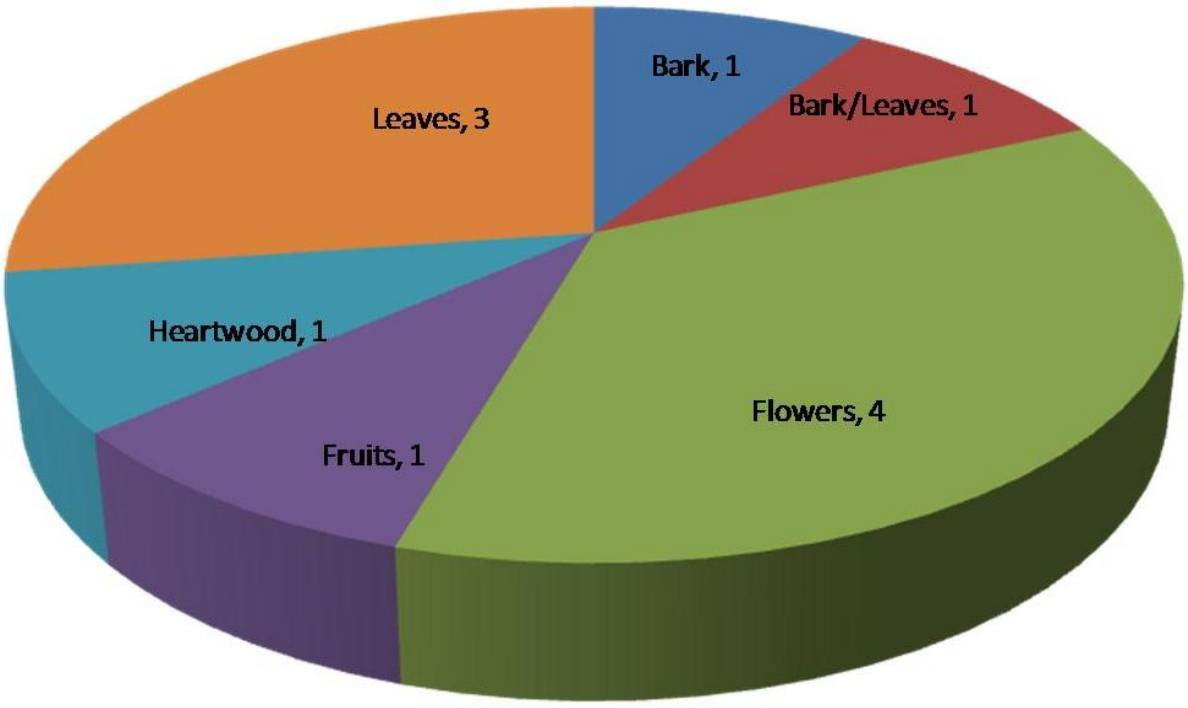

Fig. 2 : Number wise presentation of Dye-Yielding Plants

Fig. 3: Dye Yielding Plants in the study area (a) *Ampelocissus latifolia* (Roxb.) Planch. (b) *Ziziphus xylopyrus* (Retz.) Willd.,(c) *Butea monosperma* (Lam.) Taub., (d) *Indigofera tinctoria* L., (e) *Acacia catechu* (L.f.) Willd.(f) *Nyctanthes arbor-tristis* L.(g) *Wrightia tinctoria* R.Br.(h) *Justicia adhatoda* L.(i) *Woodflordia fruticosa* (L.) Kurz (j) *Mallotus philippensis* (Lam.) Mull.Arg. (k) *Phyllanthus emblica* L.

In terms of plant parts utilized for dye extraction it was found that flowers of 04% has been utilized for dye extraction followed by leaves (03%), bark (03%), fruit (01%) (Fig.2).

CONCLUSIONS

During the survey 11 species are identified as the dye yielding species form Barwani District, MP. These species are used to get the different type of dyes by the local people. Two members of Fabaceae and Euphorbiaceae each yield natural dye, however, other families like Vitaceae, Rhamnaceae, Mimosaceae, Oleaceae, Apocynaceae, Acanthaceae and Lythraceae contribute 01 species each as dye yielding plants from the area.

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RESEARCH ARTICLE

PHYSICAL PROPERTIES ANALYSIS IN KIDNEY BEANS (*PHASEOLUS VULGARIS* L.)

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Abstract: Physical properties of Rajma (*Phaseolus vulgaris* L.) seeds were investigated and their application was also discussed. Four varieties of the kidney beans were locally collected from Garhwal region of Uttarakhand for determination of physical properties. Sample of one thousand seeds of each varieties of Rajma was taken for conducting the study. The physical parameters viz moisture content, dimensions (length, width, thickness), diameter, sphericity, surface area, volume, shape factor, bulk density, true density, porosity(%) and angle of response for different germplasm of Rajma *i.e.* PRG-4, PRG-20, PRG-2, PRG-5 and PRG-1 was taken for conducting the study. Our results showed that the germplasm PRG-5 had maximum moisture, dimensions, surface area and volume, maximum spericity and bulk density for PRG-2, maximum shape factor for PRG-4, maximum porosity (%) for PRG-1 and angle of response PRG-1.

Keywords: Rajma, Physical properties, Garhwal region, Uttarakhand.

INTRODUCTION

Rajma (*Phaseolus vulgaris* L.) or kidney beans are very popular all over the world because of its health benefits as well as their savory texture. Rajma is termed as "King of Nutrition". Brazil and Mexico is the top producer of the rajma (Broughton *et al.*, 2003).

The importance of bean to diets in the developing world is reflected in the fact that for developing countries only 13% of production is exported. The two major exporters are China and Myanmar (Gepts *et al.*, 2008). In north India red kidney beans are popularly known as "Rajma". Mostly it is grown in the northern and southern part of India. Kidney beans are also called as "Common bean", "Haircot bean", "Navy bean" or "Snap bean" (www.agrifarming.in). In comparison with other food crops, rajma has one of the widest ranges of variation in growth habits, seed characteristics (size, shape, and colour), maturation times and adaptation (Jones 1999). Common bean is a rich source of dietary proteins, complex carbohydrate, dietary fibers and minerals, such as iron and zinc, and certain vitamins. Bean production is more than twice that of chickpea, which is the second most important grain legume. When developmental status is considered further, it is seen that developing countries produce 86% of worldwide production of beans. Rajma has high content of lysine which is a good complement other than cereal crops like rice or corn which are deficient in this amino acid. It contains some antinutritional factors such as phytates, protease and amylase inhibitors, lectins and polyphenols (tannins), reduce the activity of some enzymes and the absorption of metabolites (Diaz-Batalla *et al.*, 2006; Paul Gepts, *et al.*, 2001; Batista *et al.*, 2010).

The objective of this study is to determine the engineering (physical and mechanical) properties of five locally available varieties of Rajma grown in Tehri Garhwal District of Uttarakhand, to establish a convenient reference data for their mechanization and processing. The knowledge of the engineering properties is useful for both engineers and food scientists; plant and animal breeders and it is also important in data collection in the design of machines, structures, processes and controls; and in determining the efficiency of a machine or an operation.

MATERIALS AND METHOD

The investigation was carried out at the Department of Agricultural Engineering, College of Forestry Ranichauri, Veer Chandra Singh Garhwali Uttarakhand University of Horticulture and Forestry, Tehri Garhwal (India). The present study was conducted to evaluate the some physical properties of kidney bean (rajma). The genotypes were collected from Dargi and Ranichauri villages of Tehri Garhwal district of Uttarakhand. Hundred seeds of each sample of five local varieties (germplasm) were used in this study. The seeds were cleaned up manually to remove all foreign matters such as chaff, dust and stones *etc*. These germplasm were stored in dry and cool place in ambient condition until further study. The procedures for determination of physical properties of kidney bean are discussed below:

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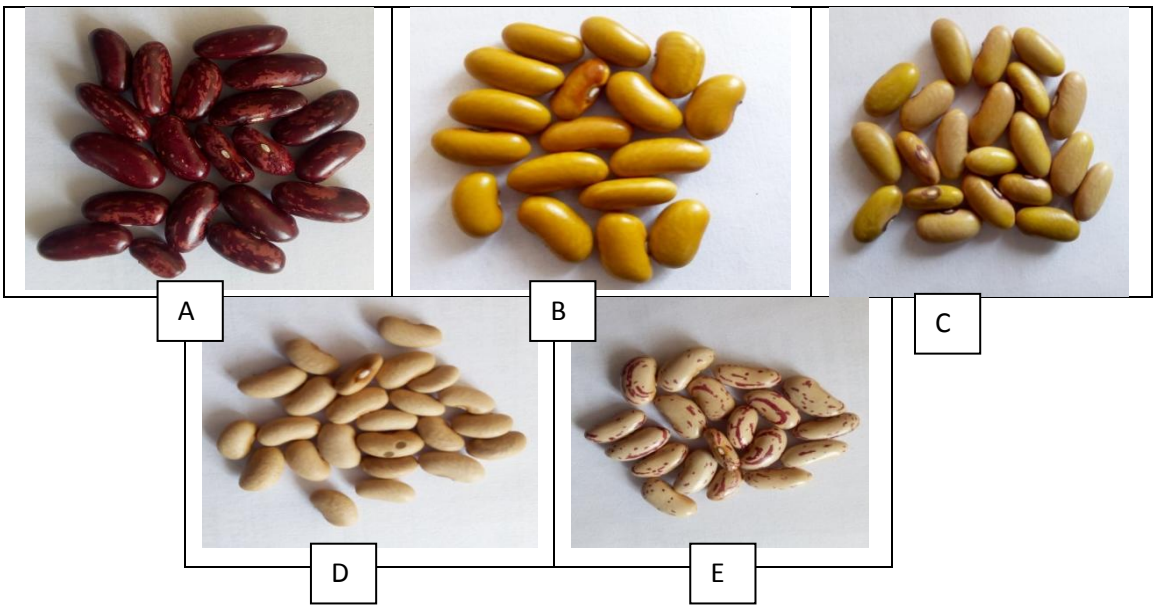

Figure 1: PRG-4, PRG-20, PRG-2, PRG-5 and PRG-1

Physical Properties

Size of the seed

For estimating the size, 50 random seeds of each species were collected. The size of rajma seeds was determined in the terms of Arithmetic mean diameter (AMD), Geometric mean diameter (GMD), Square mean diameter (SMD), Equivalent diameter (EQD) of the given kidney beans are calculated with the help of vernier caliper having a least count of 0.02 mm. the size is determined by the relationship given by (Mohsenin, 1978)

$$AMD = L+W+T \quad \ldots(1)$$

$$GMD = (LWT)^{\frac{1}{3}} \quad \ldots(2)$$

$$SMD = LW + WT + TL)^{\frac{1}{2}} \quad \ldots(3)$$

$$EQD = \frac{AMD + GMD + SMD}{3} \quad \ldots(4)$$

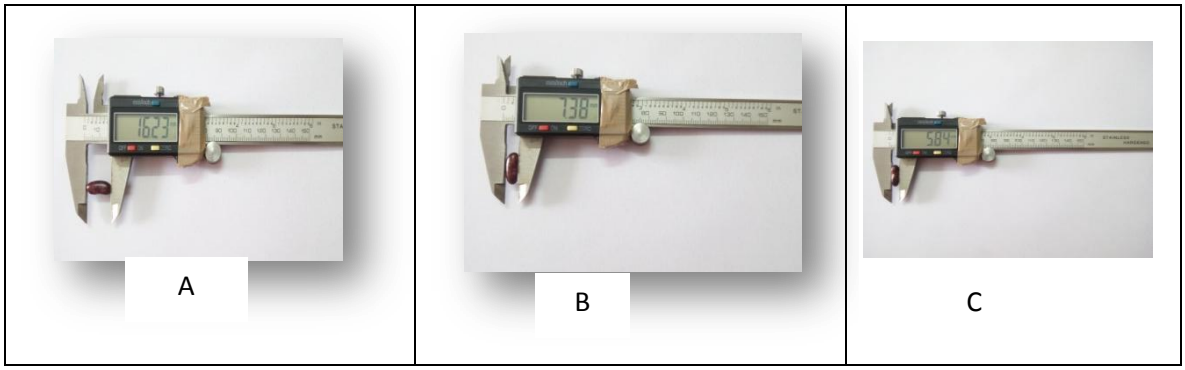

Fig 2. Measurements (a) Length, (b) width and (c) thickness

Moisture content

Moisture content of the corm was determined by oven dry method. Three samples of corm were taken and kept in oven at 105° for 24 hours. Thereafter, samples were taken out and their dry weight was determined using electronic balance. The moisture content was calculated by the following relationship (**Singh and Singh, 2015 and Singh and Chandra 2014**).

$$Mc = \frac{W_{w-W_d}}{W_d} \times 100 \quad \ldots(5)$$

Where,

M_c = Moisture content (dry basis).

W_w = Weight of materials before oven drying.

W_d = Weight of material after oven drying.

Thousand seed weight

Three samples, each germplasm comprising of one hundred corms were taken. The weight of each seed was determined using electronic balance having a least count of 0.1g. The average of all three samples was taken to determine the average weight of rajma seeds.

Coefficient of static friction (μ)

The coefficient of static friction was measured for all the five different kidney beans species by inclined

plane method. The kidney beans have been kept on a horizontal surface and the slope is being increased gradually. The angle at which kidney beans started sliding was recorded, and the coefficient of static friction will be computed by using the following relationship as given below,

$$\mu = \tan(Ø) \quad \ldots (6)$$

Bulk density, true density and Porosity

The Bulk density, true density and Porosity were measured by the toluene displacement method. The procedure was replicated three times and the average bulk density of the seed was calculated the equation given below **(Mohsenin, 1970.)**

$$\text{B.D. } (\rho) \frac{w}{v} \quad \ldots (7)$$

Where,

ρ = Bulk density, g/cc

W = weight of the corm, g

V = Volume of the sample, cc

$$\text{Porosity } (\varepsilon) = \frac{1-\rho}{\rho} \times 100 \quad \ldots (8)$$

Results and Conclusion: Table 1 given below showed the mean and standard errors of phyisical parameters viz moisture content, dimensions (length, width, thickness), diameter, sphericity, surface area, volume, shape factor, bulk density, true density, porosity(%) and angle of response for different verities of Rajma i.e. PRG-4, PRG-20, PRG-2, PRG-5 and PRG-1. In this study 100 samples for each germplasm have been taken for conducting this study. The length, width and thickness had been measured in mm, surface area in mm^2, volume in mm^3, density in gm/mm^3 and angle of response was measured in degree.

Table 1. Physical parameters for different verities (PRG-4, PRG-20, PRG-2, PRG-5 and PRG-1) of Rajma

ITEMS→ MEASUREMENT↓	***PRG-4***	***PRG-20***	***PRG-2***	***PRG-5***	***PRG-1***
MOISTURE CONTENT	8.81±0.525	13.66±0.719	15.84±2.899	18.93±0.306	9.12±0.480
DIMENSIONS (Length)(mm)	1.216±0.545	0.841±0.173	1.044±0.465	1.318±0.694	1.109±0.788
(Width) (mm)	0.617±0.020	0.467±0.018	0.609±0.005	0.669±0.093	0.608±0.044
(Thickness) (mm)	0.466±0.065	0.320±0.059	0.478±0.085	0.488±0.059	0.428±0.087
DIAMETER (GMD) (mm)	0.704±0.056	0.501±0.092	0.672±0.045	0.754±0.008	0.660±0.012
SPHERICITY	0.578±0.054	0.595±0.089	0.643±0.076	0.572±0.036	0.595±0.065
SURFACE AREA(mm^2)	1.892±0.073	0.942±0.008	1.625±0.032	2.186±0.011	1.636±0.064
VOLUME(mm^3)	0.222±0.009	0.078±0.008	0.182±0.084	0.274±0.099	0.237±0.068
SHAPE FACTOR	0.190±0.094	0.177±0.086	0.183±0.099	0.187±0.090	0.189±0.099
BULK DENSITY	0.205	0.1667	0.23	0.1992	0.091
TRUE DENSITY	1.345	1.358	1.276	1.444	1.229
POROSITY (%)	84.75	87.72	81.97	86.20	92.59
ANGLE OF REPOSE	23.215±2.589	23.019±2.24	23.109±1.459	23.25±1.67	23.8±1.2

The present result showed that PRG-5 has the maximum moisture content *i.e.* (18.93 + 0.306) whereas PRG-4 has minimum (8.81±0.525). Altuntas and Demirtola, 2007 have experimented on three different moisture percent (8.21, 11.83 and 18.01) on rajam. The present result had much higher moisture content may be better fertility of soil and atmospheric condition of hilly areas of rainfed conditions of Garhwal region of Uttarakhand. The germplasm PRG-5 had maximum length, width and thickness whereas minimum length, width and thickness had PRG-20. The Maximum sphericity was found in PRG-2 (0.643 + 0.076) and minimum sphericity for PRG-5 (0.572±0.036). The germplasm of Rajma PRG-5 (2.186 ± 0.011) has maximum surface area and minimum surface area for PRG-20(0.942±0.008). The maximum volume was found for PRG-1(0.437±0.068) and minimum volume for PRG-20(0.078±0.008). The PRG-4 had maximum shape factor and minimum shape factor for PRG-20. The maximum bulk density had been found for PRG-2 and minimum bulk density for PRG-20. The germplasm PRG-5 had maximum true density and minimum true density for PRG-2. The maximum porosity (%) was found for PRG-1 and minimum porosity for PRG-2 whereas maximum angle of response was found for PGR-1 and minimum for PRG-20. In the higher moisture content rajma that is 18.01 in rajam the length (16.766±0.31), width (8.992±0.12), sphericity (61.31±0.04) and porosity (61.114±1.9) were estimated (Altuntas and Demirtola, 2007). The Physical and functional properties of Rajma (*Phaseolus vulgaris*) was studied and found that the diameter 7.97 mm, average sphericity 57.22%, Porosity 19%, compressibility index 24.90, Hausner's ratio 1.238 (Gani, *et al.*, 2015). Therefore, this study suggests that the physical property of Rajma was better in Garhwal region of Uttarakhand, India.

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RESEARCH ARTICLE

INFLUENCE OF ORGANIC, INORGANIC AND INTEGRATED NUTRIENT MANAGEMENT ON BIOMASS YIELD AND QUALITY OF BRAHMI

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Abstract: *Bacopa monnieri* (L.) Pennel, commonly known as Brahmi is an important medicinal crop which is in high popularity because of its high market value. Brahmi belongs to the family Plantaginaceae and is extensively being used in Indian system of medicine as a memmory booster. Brahmi is used to treat insomnia, insanity, depression, psychosis, stress, cardiac, respiratory problems etc. The therapeutic effect is mainly based on bacosides (saponins). Bacoside A (a saponin glycoside) is the major active ingredient. Nowadays organic farming or integrated nutrient farming in crop production is gaining much boom because of our increasing health consciousness. In this context an experiment was conducted to study the effect of organic and inorganic sources of nutrients on the quality of brahmi at All India Coordinated Research Project on Medicinal, Aromatic Plants & Betelvine, College of Horticulture, Kerala Agricultural University, Thrissur during 2018 - 2019. The experimental design was RBD with six different treatments .The results of the study revealed that plants which received integrated nutrient management (NPK @ 100:60:60 kg/ha along with Farm yard manure @ 10 t/ha) recorded higher biomass yield (6672 kg/ha)and Bacoside A content (0.94%). Integrated Nutrient Management was thus found more effective to boost up the production of the active constituent Bacoside A compared to purely organic or purely inorganic management practices in Brahmi cultivation. Calcium, Magnesium and iron content were also found higher in INM.

Keywords: *Bacopa monnieri* , Bacoside A, Integrated nutrient management (INM)

INTRODUCTION

Bacopa monnieri (L.) Pennell is a perennial trailing herb which belongs to the family Plantaginaceae. This medicinal plant famous as brahmi in Hindi and Malayalam is a very effective memory booster and brain tonic used in Ayurveda. It is effective in the treatment of epilepsy, asthma, ulcers, tumors, enlarged spleen, inflammations, leprosy, anemia and gastroenteritis. The whole plant is medicinal containing saponin bacosides as major active ingredients. This bacoside saponin can be taken as marker compound for authentication of true brahmi samples. Brahmi is the major essential ingredient in many ayurvedic formulations like Brahmeegritham, Brahmi oil and Saraswatharishtam (Nair and Sashtri, 1990; Nambiar *et al*.,2000;Ved and G.S.,2007). Since the nutrient management can greatly influence the yield as well as quality parameters of plants, an experiment was conducted to study the effect of organic, inorganic and integrated nutrient management on the biomass yield and quality of Brahmi (*Bacopa monnieri* L.)

MATERIALS AND METHODS

Field experiment was conducted at AICRP on MAP&B, College of Horticulture, Kerala Agricultural University, Vellanikkara during 2018. The site lied between 13^0 32'N latitude and 76^0 26'E longitude at an elevation of about 40 m from MSL and had typical humid tropical climate. The soil was latterite sandy loam of oxisol group. The experimental design was RBD with six treatments in four replications. (T_1 - control,T_2 -NPK (100:60:60 kg/ha) **,** T_3- FYM (10 t/ha), T_4- FYM (5 t/ha) ,T_5- FYM (10 t/ha)+ NPK (100:60:60 kg/ha) ,T_6- FYM (5 t/ha)+ NPK (100:60:60 kg/ha)).

The experimental area was ploughed and leveled thoroughly. Plot size taken was 6 m^2. Fresh noded cuttings of about 10 cm were planted at a spacing of 20cm X 20cm. Harvest was done at the 5 th month after planting. The fresh biomass yield was observed at the time of harvest. Fresh leaves were subjected to bacoside A analysis by HPTLC (High performance thin layer chromatography) method using bacoside standard purchased from Reddys Lab ,Mumbai (Powar and Jadav, 2015). For HPTLC the solid phase used was Silica gel 60 $F_{254,}$ mobile phase as Toluene: Ethyl acetate: methanol: Acetic acid (3:4:3:1). Methanol extract of Brahmi was used. The spray reagent used was anisaldehyde followed by heating of the plate at 100^0C for 2 minutes.

Heavy metal and mineral content analysis were also carried out in the Brahmi samples by ICP -OES (Inductively coupled plasma - optical emission spectrometry) method (Powar and Jadav, 2015) . The data was subjected to statistical analysis using MSTAT – C package (Freed, 2006).

RESULTS AND DISCUSSION

It was observed that the plants in T_5 which received integrated nutrient management FYM (@10 t/ha) + NPK (@100:60:60 kg/ha) significantly recorded higher biomass /fresh herbage yield (667 kg/ha)

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followed by the plants (T6) treated with FYM @ 5 t/ha+ NPK @100:60:60 kg/ha with a yield of 6216 kg/ha. Lowest yield (3335 kg/ha) was recorded for absolute control plots.

The same trend was observed for Bacoside A content also. It was significantly higher for INM treatments T5 with 0.94% bacoside A content .Treatment T6 was on par with a bacoside content 0.93 %. Lowest bacoside content was observed in absolute control (T_1, 0.66%) as detailed in Table.1 and Figure 1. Calcium, Magnesium and iron content were also found higher in INM treated plants (Table 2). Effect of nutrients on the heavy metal load of the plant material when tested revealed absence of arsenic, cadmium, led and chromium. Similar results are reported from other scientific studies also. Agronomic manipulation studies carried out in Brahmi in Assam plains by Aparna *et al* in 2014 showed that 2t/ha enriched compost application gave a hike in the yield (144 g/m^2). Singh *et al* from Pantnagar (2007) revealed that INM application with 75 kg N + 5 t/ha FYM/ha gave maximum yield compared to all other treatments and control in their soils for Brahmi.

Table 1. Effect of nutrients on herbage yield and Bacoside A content of Brahmi

Nutrient Management	**Treatment code**	**Treatment details**	**Bacoside AA (%)**	**Herbage yield kg/ha kg/ha**
Absolute	T1	No manures and fertilizers	0.655[e]	3335[f]
Inorganic	T2	NPK (100:60:60 kg/ha)	0.845[c]	5189[c]
Organic	T3	FYM (10 t/ha)	0.870[b]	4169[d]
	T4	FYM (5 t/ha)	0.733[d]	3629[e]
Integrated	T5	**FYM (10 t/ha) + NPK (100:60:60**	**0.940[a]**	**6672[a]**
	T6	**FYM (5 t/ha) + NPK (100:60:60**	**0.928[a]**	**6216[b]**
	CD(0.05)		0.021	47.323

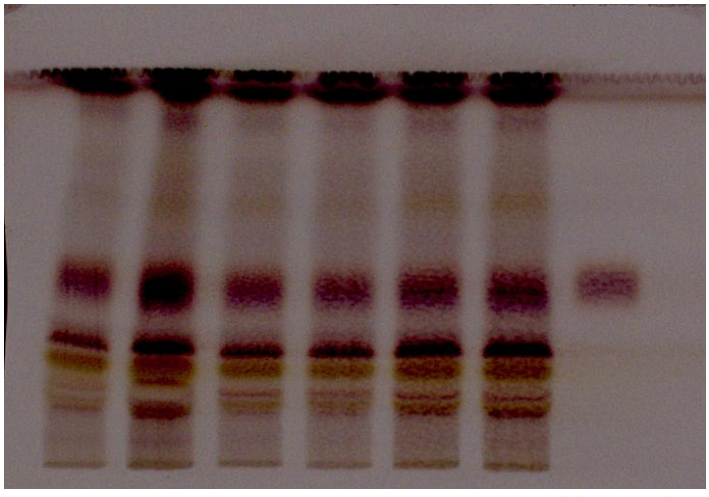


Figure 1. HPTLC of Brahmi – Bacoside A estimation

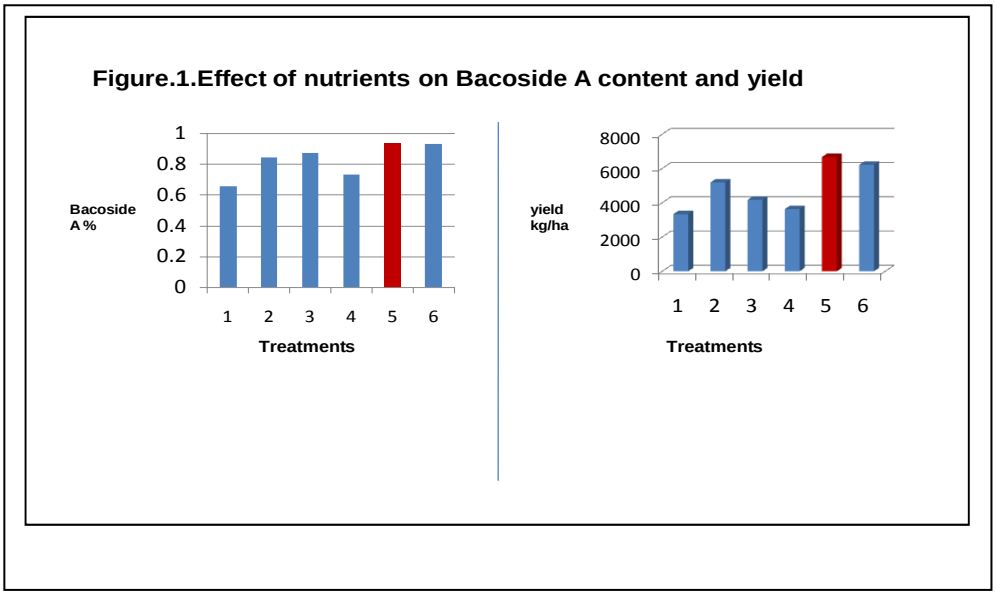
Table 2. Effect of nutrients on mineral content in brahmi

Treatments	Mn %	Zn%	Ca %	Mg%	Fe **%**
T1	0.050	0.013	0.93	1.16	0.046
T2	0.062	0.015	0.94	1.20	0.069
T3	0.068	0.021	1.18	1.35	0.077
T4	0.063	0.018	1.05	1.35	0.066
T5	**0.069**	**0.210**	**1.20**	**1.38**	**0.118**
T6	0.067	0.200	1.18	1.35	0.111

Table 3. Effect of nutrients on heavy metal content in brahmi

Treatments	As **%**	Cr **%**	Cd **%**	Pb **%**
T1	BDL	BDL	BDL	BDL
T2	BDL	BDL	BDL	BDL
T3	BDL	BDL	BDL	BDL
T4	BDL	BDL	BDL	BDL
T5	BDL	BDL	BDL	BDL
T6	BDL	BDL	BDL	BDL

BDL= below detectable level



CONCLUSION

Based on the present investigation it was concluded that INM is better compared to pure organic or pure inorganic nutrient management in Brahmi (*Bacopa monnieri*) cultivation with respect to Bacoside A content as well as herbage yield.

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SHORT COMMUNICATION

COLORED PAN TRAPS FOR INSECT POLLINATORS/VISITORS DIVERSITY IN MUSTARD ECOSYSTEM IN AMBIKAPUR OF CHHATTISGARH

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Abstract: A field experiment was undertaken at Rajmohini Devi College of Agriculture and Research Station, Ambikapur Surguja (Chhattisgarh) during 2017-18 to study the insect pollinators diversity in mustard ecosystem using different fluorescent colored pan trap *i.e.* White, blue and yellow at onset of bloom, full bloom and end of bloom of mustard crop in two different habitats. In habitat-I various insect visitors i.e. *Apis dorsata, A. indica, A. florea*, syrphid fly, house fly, black pants and small ants were recorded. In the colored pan traps maximum population was noticed in yellow pan trap (14.31 insect/tarp) followed by blue trap (13.99 insect/trap) and minimum in white (6.3 insect/trap) at onset of bloom. However, at the full bloom highest population was recorded in yellow pan trap (24.65 insect/trap) followed by blue pan trap (20.98 insects/trap) and lowest in white trap (20.65 insects/trap). Similarly at the end of bloom higher population was recorded in yellow pan trap (14.32 insects/trap) followed by blue pan trap (13.99 insects/trap) and the lowest in white pan trap (5.99 insects/trap) .Habitat-II During the onset of bloom higher population was recorded in yellow pan trap (21.66 insect/trap) followed by in blue trap (21.65 insect/trap) and lower population was recorded in white pan trap (8.32 insect/trap). At the full bloom period maximum population was noticed in blue pan trap (18.65 insect/trap) followed by in yellow pan trap (16.65 insect/trap) however the lowest population was recorded in white colored pan trap (9.98 insect /trap).Similarly, at the end of bloom higher population was recorded in yellow pan trap (24.99 insect/trap) followed by blue pan trap (24.98 insect/trap) and the lower population was noticed in white colored pan trap (8.65 insect/trap).

Keywords: Diversity, Colored pan trap, Habitat, Pollinators, Mustard ecosystem

INTRODUCTION

Insects are considered as a pollinators they are an important visitors of different flowers. They transfer the pollen from one flower to another and benefit the farming community by means of pollination process. Pollination is depends on various orders of insects i.e. Diptera, Hymenoptera, Coleoptera, Thysanoptera and Neuropterans etc. Due to pollination process yield is enhanced about 15-17 per cent in cross pollinated crop. Honeybees, beetles, bugs, birds and winds are as pollinator's agents. Among the pollinators honey Bees are the most efficient pollinators which pollinate the crop for obtain the good yield.

MATERIALS AND METHODS

A field experiment was undertaken at Rajmohini Devi College of Agriculture and Research Station, Ambikapur Surguja (Chhattisgarh) during 2017-18 to study the insect pollinator's diversity in mustard ecosystem using different fluorescent colored pan traps *i.e.* White, blue and yellow were placed at onset of bloom, full bloom and end of bloom on mustard crop. Diversity of insect visitors was recorded during the flowering period of mustard (October to November). During the study various fluorescent colored pan traps *viz* white, yellow and blue were used and were placed before 0900h in the morning and picked up after 1500h in the afternoon and the population of insect pollinators/visitors in each traps were recorded at three stages of flowering period i.e. onset of bloom, full bloom and at the end of bloom at the GPS location of mustard ecosystem N 23.141752^{0} and ^{E} 83.1804720.

RESULT AND DISCUSSION

The result shows in table 1a of habitat-I and habitat-II table 1b, the average population of insect visitors/pollinators were recorded maximum population in yellow colored pan trap at full bloom period in mustard crop however the low population of insect visitors/pollinators was recorded at onset of bloom and end of bloom at Ambikapur, Surguja of Chhattisgarh, India.

Table 1. Population of insect pollinators/visitors in Mustard ecosystem (average of three observations)

Visitors	Onset of Bloom			Full Bloom			End of Bloom		
	White	Yellow	Blue	White	Yellow	Blue	White	Yellow	Blue
Habitat -I									

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Apis dorsata	0.66	1.66	2.00	7.33	6.33	5.33	2.00	3.66	2.66
Apis indica	1.33	3.33	2.33	4.00	5.00	6.66	1.66	6.33	4.33
Apis florea	0.33	1.66	1.00	3.66	5.66	3.00	1.33	2.33	3.00
Syrphid fly	0.66	1.33	1.00	1.33	2.00	2.33	1.00	2.00	4.00
House fly	0.33	2.00	1.33	4.33	5.66	3.66	0.00	0.00	0.00
Black ant	1.66	2.33	3.00	0.00	0.00	0.00	0.00	0.00	0.00
Small ants	1.33	2.00	3.33	0.00	0.00	0.00	0.00	0.00	0.00
Total	**6.3**	**14.31**	**13.99**	**20.65**	**24.65**	**20.98**	**5.99**	**14.32**	**13.99**

Table 2. Population of insect pollinators/visitors in Mustard ecosystem (average of three observations)

Visitors	**Onset of Bloom**			**Full Bloom**			**End of Bloom**		
	White	Yellow	Blue	White	Yellow	Blue	White	Yellow	Blue
Habitat -II									
Apis dorsata	2.33	6.00	3.33	2.33	4.33	2.66	0.66	2.33	3.00
Apis indica	2.00	5.00	4.33	1.66	3.33	5.00	1.00	3.66	2.33
Apis florea	1.33	3.66	5.66	3.66	4.00	5.33	1.33	3.00	4.00
Syrphid fly	1.66	4.00	6.00	1.33	3.66	3.33	2.00	5.00	4.66
House fly	1.00	3.00	2.33	1.00	1.33	2.33	1.33	4.00	4.33
Black ant	00	00	00	00	00	00	2.33	5.00	3.66
Small ants	00	00	00	00	00	00	1.66	2.00	3.00
Total	**8.32**	**21.66**	**21.65**	**9.98**	**16.65**	**18.65**	**8.65**	**24.99**	**24.98**

Result obtained from the study of different fluorescent colored pan traps were used i.e. white, yellow and blue for attract the different insect pollinators during onset, full bloom and end of bloom in mustard ecosystem. Various colored pan traps i.e. white yellow and blue were installed at onset of bloom, full bloom and end of bloom. Insect pollinators i.e. *Apis dorsata, A. indica, A. florea*, syrphid fly, house fly, black ant and small ant were recorded. Above insect pollinators were recorded higher population in yellow (14.31 insect/trap) fluorescent colored pan trap followed by in blue fluorescent colored pan trap (13.99 insect/trap) . During the full bloom period maximum insect pollinators/visitors were trapped on yellow fluorescent colored pan trap (24.65 insect/trap) followed by in blue pan trap (20.98 insect/trap) and minimum population was recorded in white colored pan trap(20.65 insect/trap). Similarly at the end of bloom maximum population was recorded in yellow fluorescent colored pan trap (14.32 insect/trap) followed by blue fluorescent pan trap (13.99 insect/trap) and lowest in white colored pan trap (5.99 insect/trap).

Similarly in habitat-II same traps were installed to trap the various insect pollinators were trapped. During the onset of bloom higher population was recorded in yellow fluorescent colored pan trap (21.66 insect/trap) followed by in blue trap (21.65 insect/trap) and lower population was recorded in white pan trap (8.32 insect/trap). During the full bloom period maximum population was noticed in blue pan trap (18.65 insect/trap) followed by in yellow pan trap (16.65 insect/trap) however the lowest population was recorded in white colored pan trap (9.98 insect /trap). At the end of bloom higher population was recorded in yellow pan trap (24.99 insect/trap) followed by blue pan trap (24.98 insect/trap) however the lower population was found in white colored pan trap (8.65 insect/trap) Table 1b. Earlier workers Joshua and Hanula (2007) who worked out the efficiency of Malaise traps and colored pan traps for collecting flower visiting insects. Nuttman *et al.* (2011) has been reported the utility of Aerial Pan-Trapping for Assessing insect pollinators, Vrdoljak and Samways (2012) has also reported the optimising coloured pan traps to survey flower visiting insects and Devi *et al*. (2016) impact of Habitat on Insect Pollinator Diversity on Coriander Bloom, Devi *et al*. (2017) has also reported the diversity of Insect Pollinators in Mustard. Painkra (2018) has observed the pollinators diversity in bitter gourd ecosystem using the colored pan trap ie yellow, white and blue for trapping the various insects.

CONCLUSION

Obtained results of this research showed that maximum population of insect visitors were observed during the full bloom period in mustard ecosystem in yellow colored pan trap followed by blue and white colored pan traps.

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