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Research Article

Studies on the influence of different organic manures on soil microbial activity, growth and yield performance of Blackgram (*Vigna mungo* L.)

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ABSTRACT

The core principle of organic farming revolves around utilizing naturally occurring resources, such as organic waste, along with natural processes like decomposition, biological nitrogen fixation, and resistance, to fulfill the requirements of crop production. By employing organic manures to promote crop growth, the organic matter content of the soil is enhanced. Organic manures not only provide nutrients but also serve as a food source for microorganisms, fostering their population growth. This, in turn, enhances nutrient mineralization in the soil, improving soil fertility and productivity. Experiment was conducted to explore the impact of different organic manures on soil microbial activity, as well as the growth and yield of black gram (*Vigna mungo* L.). The study consisted of seven treatments and three replications, following a Completely Randomized Design (CRD). The treatment involving a combination of FYM (25%), Vermicompost (25%), Cow dung (25%), and Bone meal (25%) exhibited the highest abundance of various microorganisms, including Bacteria, Fungi, Actinomycetes, PO_4-solubilizing microorganisms, and *Rhizobium*, whereas the control group had the lowest abundance. Several plant parameters, such as plant height, number of branches per plant, number of leaves per plant, dry matter production, test weight and plant yield, were measured. Application of recommended dose of fertilizer (RDF) (100%) resulted in significantly higher values for these parameters, whereas the control group yielded the lowest results.

KEY WORDS: Blackgram; *Rhizobium*; Biofertilizers; *Vigna mungo*; Rhizosphere

1. Introduction

The increasing awareness of environmental conservation, health risks associated with agrochemicals and consumers' preference for safe and hazard-free food are significant factors driving the growing interest in organic agriculture. The fundamental objective of organic farming is to achieve sustainable agricultural production while preserving natural resources and ensuring the production of high-quality agricultural goods. The advent of high-tech modern agriculture, heavily reliant on fertilizers and chemicals since the mid-1960s, helped bridge the gap between food demand and supply for a rapidly growing population. However, by the late 1980s, signs of soil exhaustion and declining overall productivity per unit area began to emerge, despite the use of modern technologies. This trend has persisted until today.

In agriculture, organic manures play a crucial role by adding essential organic and mineral matter to the soil. Organic systems prioritize the management of organic matter to enhance soil fertility and productivity. Organic matter exerts a significant influence on nearly all soil properties, and it is considered a valuable repository of nutrients. The presence of soil fauna and microorganisms is indispensable for the

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mineralization of organic matter. The soil is home to a dynamic population of microorganisms, arthropods, and other soil biota.

Black gram (*Vigna mungo* L.) holds the position of being the third most significant pulse crop in India. This annual pulse crop originated in central Asia and is also extensively cultivated in regions such as West Indies, Japan, and other tropical/subtropical countries. The seeds of black gram are highly nutritious, containing a substantial amount of protein (24-26%) as well as potassium, phosphorus, and calcium, with good sodium content. Additionally, it is known for its richness in vitamins A, B_1, B_3, along with nutritionally valuable proteins, essential minerals, and vitamins. Black gram is associated with medicinal properties, including the treatment of diabetes, sexual dysfunction, nervous disorders, hair disorders, digestive system disorders, and rheumatic afflictions (Anonymous, 2010). As a summer pulse crop, it has a relatively short duration of 90-120 days (Delic *et al.*, 2009). Notably, black gram is valued for its high digestibility and the absence of flatulence effects (Fary, 2002). In order to reduce production costs associated with mineral fertilization and promote environmental protection, application organic manures can be employed to increase pulse production. With this objective experiment was conducted to assess the different organic manures on the growth and yield of blackgram.

2. Materials and Methods

The treatments used in the experiment were as follows: T_1 represented the control group, T_2 received the recommended dose of fertilizer (RDF) at 100%, T_3 was treated with 100% Farmyard Manure (FYM), T_4 received 100% Vermicompost, T_5 was treated with 100% Cow dung, T_6 received 100% Bone meal, and T_7 was a combination of FYM (25%) + Vermicompost (25%) + Cow dung (25%) + Bone meal (25%).

For the experiment, recommended dose of organic manures per ha was taken into consideration and calculated as per the kg of soils required for the pot experiment, each treatment was replicated three times to ensure reliable results. The variety of blackgram used in the pot experiment was LBG 17. Data collected on various soil microbial populations, blackgram growth, and yield parameters were subjected to statistical analysis using Fisher's method of analysis of variance, with the interpretation guidelines outlined by Gomez and Gomez (1984).

To enumerate the microbial populations, soil samples were collected from the pot experiment site. Bacteria, fungi, actinomycetes, as well as physiological groups such as *Rhizobium* and Phosphorus Solubilizing Microorganisms (PSM), were assessed using the serial dilution agar plate method. During different stages of blackgram growth 30 days after sowing (DAS) and At harvest, random plant selections were made from each treatment to record observations on various growth and yield parameters. These observations were integral to assessing the impact of the different treatments. Overall, this experiment aimed to investigate the effects of various organic manures on soil microbial activity, blackgram growth, and yield parameters in order to gain insights into sustainable agricultural practices.

3. Results and Discussion

3.1 Assessment of Microbial Population in the Study Site

At 30 days after sowing (DAS), the combined application of FYM (25%) + vermicompost (25%)

+ cow dung (25%) + bone meal (25%) resulted in a significantly higher bacterial population (cfu × 10^7 g^{-1} of dry soil) compared to other treatments. The next best treatments were cow dung (100%), vermicompost (100%), FYM (100%), and RDF (100%). The control treatment exhibited a significantly lower number of bacterial colonies. Similar trend was also noticed at harvest stage for bacterial population.

Regarding fungal population (cfu × 10^4 g^{-1} of dry soil), at 30 DAS, the combined application of FYM (25%) + vermicompost (25%) + cow dung (25%) + bone meal (25%) showed a significantly higher fungal population compared to other treatments. The control treatment exhibited a significantly lower number of fungal colonies, which was comparable to the other treatments except vermicompost (100%), FYM (100%), RDF (100%), and cow dung. At harvest, the combined application of FYM (25%) + vermicompost (25%) + cow dung (25%) + bone meal (25%) resulted in a significantly higher fungal population compared to all other treatments. The control treatment exhibited a significantly lower number of fungal colonies, which was comparable to the other treatments except FYM and RDF. Similar results were also obtained by Santoshagowda *et al.*, 2015 and recorded combined application of organic manures showed the significant increase the growth and yield attributes of blackgram.

Actinomycetes population significantly influenced by the application of different combination of organic manures. At 30 days after sowing (DAS), the application of FYM (25%) + vermicompost (25%) + cow dung (25%) + bone meal (25%) resulted in a significantly higher actinomycetes population (cfu × 10^3 g^{-1} of dry soil) compared to other treatments. The control treatment exhibited a significantly lower number of actinomycetes colonies, which was comparable to the other treatments except vermicompost (100%), FYM (100%), RDF (100%), and cow dung. At harvest, the combined application of FYM (25%) + vermicompost (25%) + cow dung (25%) + bone meal (25%) showed a higher actinomycetes population compared to all other treatments. The control treatment exhibited a significantly lower number of actinomycetes colonies, which was comparable to the other treatments except RDF, cow dung, and bone meal. These findings highlight the positive impact of specific treatments, particularly the combined application of organic amendments, on the actinomycetes population in the soil. The presence of higher actinomycetes populations can contribute to enhanced soil health and nutrient cycling, thus promoting overall crop productivity and sustainability.

With respect to Phosphate Solubilizing Microbial (PSM) population, At 30 days after sowing (DAS), the combined application of FYM (25%) + vermicompost (25%) + cow dung (25%) + bone meal (25%) resulted in a significantly higher population of phosphate solubilizing microbes (PSM) (cfu × 10^3 g^{-1} of dry soil) compared to all treatments. The control treatment exhibited a significantly lower number of PSM colonies, which was comparable to the other treatments. At harvest, the combined application of FYM (25%) + vermicompost (25%) + cow dung (25%) + bone meal (25%) showed a significantly higher population of phosphate solubilizing microbes compared to all other treatment. The control treatment exhibited a significantly lower number of PSM colonies. These results indicate that the application of specific treatments, particularly the combined use of organic amendments, can significantly enhance the population of phosphate solubilizing microbes in the soil (Ananth *et al.*, 2015). This is beneficial for nutrient availability and uptake by plants, ultimately contributing to improved crop growth and productivity.

The population of nitrogen-fixing microbes, specifically *Rhizobium*, was evaluated at 30 days after sowing (DAS) and At harvest stage in blackgram under different treatments. Among the treatments, the application of FYM (25%) + vermicompost (25%) + cow dung (25%) + bone meal (25%) resulted in a significantly higher population of nitrogen-fixing microbes at 30 DAS compared to all treatments. The control treatment exhibited a significantly lower number of nitrogen-fixing microbial colonies. Similarly, At harvest, the combined application of FYM (25%) + vermicompost (25%) + cow dung (25%) + bone meal (25%) showed a significantly higher population of nitrogen-fixing microbes compared to all other treatments. The control treatment exhibited a significantly lower number of nitrogen-fixing microbial colonies, while the remaining treatments showed similar results to each other. These findings emphasize the positive impact of incorporating organic amendments, particularly the combined use of FYM, vermicompost, cow dung, and bone meal, in promoting the population of nitrogen-fixing microbes like *Rhizobium*, which can enhance nitrogen fixation in the soil and contribute to improved blackgram growth and development.

In the current study, a noticeable enhancement in the population of various soil microorganisms, including bacteria, fungi, actinomycetes, *Rhizobium*, and phosphate solubilizers, was observed at different stages of blackgram growth (30 DAS and At harvest). This increase can be attributed to the availability of easily metabolizable compounds early in the growth stage, as well as the active growth phase of the crop, resulting in higher root exudate production and providing favorable conditions for diverse microbial communities. The combined application of different organic manures, such as FYM, cow dung and bone meal, resulted in a significant increase in microbial population, particularly with the treatment of FYM (25%) + vermicompost (25%) + cow dung (25%) + bone meal (25%). In contrast, the control treatment showed lower microbial population, indicating minimal impact on the soil microbial community. These findings highlight the positive influence of organic manure additions on the abundance of beneficial soil microorganisms. Consistent with previous studies conducted by Sreenivasa (2007), Deshpande *et al*. (2010), and Dhok and Ghodpage (2011), the present results also demonstrate that the combined application of organic substances leads to higher soil microbial populations compared to individual organic inputs or the control treatment. These findings further support the notion that organic amendments have a positive impact on soil microorganisms. Moreover, the effects of these microbial populations on plant growth parameters were also investigated (Table 1).

At 30 days after sowing (DAS), the application of recommended dose of fertilizer (RDF) resulted in significantly higher plant height of blackgram (29.00 cm) compared to all other treatments. The control treatment exhibited significantly lower plant height (13.33 cm), the combined application of FYM (25%) + vermicompost (25%) + cow dung (25%) + bone meal (25%) recorded (26.33 cm), FYM (100%) (24.67 cm), cow dung (100%) (22.33 cm) and vermicompost (100%) (21.00 cm),. At harvest, the application of RDF (100%) continued to yield significantly taller plants (38.33 cm) compared to all other treatments. The treatments with combined application of FYM

(25%) + vermicompost (25%) + cow dung (25%) + bone meal (25%) (54.00 cm) and vermicompost (100%) alone (49.67 cm) followed as the next best options. The control treatment displayed significantly lower plant height (38.33 cm), which was similar to the remaining treatments.

At harvest, the application of recommended dose of fertilizer (RDF) resulted in a significantly higher number of branches per plant of blackgram (8.67), which was followed combined application of FYM (25%) + vermicompost (25%) + cow dung (25%) + bone meal (25%) (7.33) and vermicompost (100%) (5.0). The control treatment exhibited a significantly lower number of branches per plant (3.00).

The number of leaves per blackgram plant was assessed at two different stages of growth. At 30 days after sowing (DAS), the application of RDF resulted in a significantly higher number of leaves per plant (5.33) followed by the treatment which received combination FYM (25%) + vermicompost (25%) + cow dung (25%) + bone meal (25%) where (4.00) leaves were observed. The control treatment exhibited a significantly lower number of leaves per plant (1.67). Moving At harvest stage, the application of RDF (100%) showed a significant increase in the number of leaves per plant (13.67) compared to all other treatments, the combined application of FYM (25%) + vermicompost (25%) + cow dung (25%) + bone meal (25%) (11.33), FYM (100%) (10.67), cow dung (100%) (9.00) and vermicompost (100%) (8.33). Once again, the control treatment displayed a significantly lower number of leaves per plant (5.00).

Significant variations were observed in the dry matter production per plant of blackgram at different stages of growth. At 30 DAS, the application of RDF (100%) demonstrated a significantly higher dry matter production per plant (3.67 g plant^{-1}) compared to other treatments, the next best treatment was FYM (25%) + vermicompost (25%) + cow dung (25%) + bone meal (25%) (2.33 g plant^{-1}). Conversely, the control treatment exhibited a significantly lower dry matter production per plant (0.93 g plant^{-1}). Moving forward to At harvest stage, RDF (100%) continued to exhibit a significant impact on dry matter production per plant, recording the highest value (9.33 g plant^{-1}) among all treatments. In contrast, the control treatment resulted in a significantly lower dry matter production per plant (3.67 g plant^{-1}). The treatment which received combination of FYM (25%) + vermicompost (25%) + cow dung (25%) + bone meal (25%) recorded (6.67 g plant^{-1}) which was followed farmyard manure (100%) recorded (6.00 g plant^{-1}), cow dung (100%) (5.67 g plant^{-1}) and vermicompost (100%) (5.00 g plant^{-1}). Furthermore, blackgram plants treated with RDF (100%) showcased a substantial increase in grain yield (952 kg/ha) compared to all other treatments, while the control treatment yielded significantly lower results (387 kg/ha).

The present study revealed that plant height, number of branches, number of leaves, dry matter production, and yield exhibited an increasing trend as the plants matured. The application of inorganic fertilizer resulted in higher plant yield, as it is readily absorbed by plants, while organic manures require microbial decomposition, leading to a slower release of nutrients. The growth and yield of blackgram were significantly higher when treated with recommended dose of fertilizer (RDF) throughout the growth stages, in contrast to the significantly lower plant growth and yield observed in the control group without organic manure supplementation (compared to RDF) (Table 2). Previous studies have also reported enhanced crop growth and yields with the combined application of organic manures (Thomas and Lal, 2003; Patil *et al*., 2012; Dakshayini *et al*., 2020).

4. Conclusion

These findings highlight the positive impact of specific treatments, particularly the combined application of organic amendments, on the bacterial and fungal populations in the soil. Such treatments can play a crucial role in enhancing soil microbiota and promoting beneficial microbial activity for improved agricultural outcomes.

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Table 1: Represents the population of bacteria, fungi, actinomycetes, phosphate solubilizing microbes (PSM), and *Rhizobium* at various growth stages of blackgram, influenced by nutrient management practices using organic materials in pot experiments.

Treatments	Bacteria (CFU × 10^7)		Fungi (CFU × 10^4)		Actinomycetes (CFU × 10^3)		PSM (CFU × 10^3)		*Rhizobium* (CFU × 10^3)	
	30 DAS	At harvest	30 DAS	At harvest	30 DAS	At harvest	30 DAS	At harvest	30 DAS	At harvest
T_1 - Control	25.00	58.00	14.67	26.00	18.33	30.67	22.33	31.00	29.33	54.33
T_2 - Recommended Dose of Fertilizer (RDF)-100%	31.33	75.33	18.00	32.67	22.67	35.33	27.00	37.67	39.00	61.33
T_3 - Farmyard Manure (FYM) - 100%	38.00	87.67	28.00	41.33	32.67	44.00	33.67	45.00	52.33	77.67
T_4 - Vermicompost - 100%	36.33	81.00	22.00	34.00	28.67	39.67	29.67	40.33	45.00	71.00
T_5 - Cow dung - 100%	37.00	84.00	27.33	38.67	31.00	41.33	30.00	42.00	48.67	73.00
T_6 - Bone meal - 100%	27.67	72.67	17.00	29.00	20.33	34.67	32.33	43.67	41.33	68.67
T_7 - T_3 (25%) + T_4 (25%) + T_5 (25%) + T_6 (25%)	42.67	93.33	31.33	45.67	38.00	49.67	37.00	48.33	58.00	81.33
S.Em	0.94	1.36	1.02	1.23	1.48	1.29	1.13	1.21	1.76	1.07
CD (p=0.05%)	2.80	4.08	3.04	3.66	4.42	3.85	3.38	3.63	5.28	3.20

Note: Values are mean of three replications

Table 2: Represents the growth and yield parameters of blackgram at various growth stages, influenced by nutrient management practices using organic materials in pot experiments.

Treatments	Plant height (cm)		No. of Branches/plant		No. of Leaves/plant		Plant Dry Weight (gm)		Yield (kg/ha)
	30 DAS	At harvest	30 DAS	At harvest	30 DAS	At harvest	30 DAS	At harvest	
T_1 - Control	13.33	38.33	1.33	3.00	1.67	5.00	0.93	3.67	387
T_2 - Recommended Dose of Fertilizer (RDF)- 100%	29.00	58.67	6.33	8.67	5.33	13.67	3.67	9.33	952
T_3 - Farmyard Manure (FYM) - 100%	24.67	51.33	4.67	6.00	3.67	10.67	2.00	6.00	746
T_4 - Vermicompost - 100%	21.00	49.67	3.67	5.00	3.00	8.33	1.00	5.00	715
T_5 - Cow dung - 100%	22.33	50.00	4.00	5.33	3.00	9.00	1.67	5.67	723
T_6 - Bone meal - 100%	17.00	48.67	3.00	4.67	2.33	7.00	1.00	4.00	654
T_7 - T_3 (25%) + T_4 (25%) + T_5 (25%) + T_6 (25%)	26.33	54.00	5.00	7.33	4.00	11.33	2.33	6.67	897
S.Em	0.81	1.04	0.36	0.31	0.22	0.64	0.28	0.32	8.76
CD (p=0.05%)	2.43	3.12	1.07	0.92	0.65	1.92	0.83	0.95	26.28

Note: Values are mean of three replications

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Research Article

Assessment of field efficacy & economic viability of selected entomopathogenic fungal isolates against *Chilo partellus* (Crambidae : Lepidoptera)

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ABSTRACT

The present study aimed to evaluate the field efficacy of four promising isolates (Bb-5a, Bb-23, Bb-45, and Ma-35) of entomopathogenic fungi (EPF) against maize stem borer, *Chilo partellus*, during winter seasons over a two-year period at ICAR-National Bureau of Agricultural Insect Resources (NBAIR), Bengaluru, Karnataka, India. All tested EPF isolates demonstrated superior efficacy compared to the control in suppressing maize stem borer damage across both seasons. Pooled data analysis revealed that the incidences of dead hearts (DH), stem tunnelling (ST), galleries (per plant), and exit holes (per plant) ranged from 8.7-18.3%, 5.06-7.97 cm/plant, 1.69-2.42 per plant, and 2.72-5.12 per plant, respectively, compared to untreated control values of 21.6%, 8.93 cm/plant, 3.23 per plant, and 6.72 per plant, respectively. Among all treatments, Bb-5a exhibited significantly superior efficacy in reducing the incidence of dead hearts (8.7%), stem tunnelling (5.06 cm/plant), galleries (1.69 per plant), and exit holes (2.72 per plant). Furthermore, Bb-5a demonstrated a higher yield of 68.8 t/ha, representing a 17.65% increase over the control. Bb-5a isolate also recorded higher gross returns of ₹ 82660/ha, resulting in a maximum net profit of ₹ 56660/ha with a cost-benefit ratio of 1:3.18. These findings highlight the potential of Bb-5a as an effective biocontrol agent against maize stem borer, offering economic benefits to farmers.

KEY WORDS: *Chilo partellus*; *Beauveria bassiana*; *Metarhizium anisopliae*; Maize stem borer

1. Introduction

The maize stem borer, *Chilo partellus* Swinhoe (Crambidae: Lepidoptera), poses a significant constraint to maize (*Zea mays* L.) production globally, with resultant crop losses ranging from 24 to 83 per cent (Sarup *et al*., 1987). Maize holds a pivotal position in India's agricultural landscape, serving as a primary cereal crop for both human consumption and animal feed, covering approximately 37 per cent of cultivated land and yielding around 225 lakh tonnes annually (Anon. 2016). India's maize cultivation faces substantial threats from over 20 insect pests, among which *C. partellus* is a particularly dominant, infesting maize crop throughout their growth stages and causing substantial yield losses of 90-95% (Jalali and Singh, 2002).

C. partellus emerges as a prominent maize pest across various regions of Africa and southern Asia. The pest's infestation cycle typically commences with egg deposition on maize leaves, followed by larval emergence and migration into leaf whorls where feeding and subsequent lesion formation occur. Larvae progress to late third or

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early fourth instars, subsequently boring into the stem to feed on tissues and create tunnels. The larvae's feeding activity, particularly within leaf whorls or stem regions, disrupts meristematic tissues, leading to the characteristic symptom of "dead heart" and eventual plant mortality (Tadele and Pringle, 2007).

Despite considerable research efforts focused on *C. partellus* management in India, there remains a gap in understanding the pest's dynamics within the Karnataka region. The transition from conventional synthetic insecticides to biological control agents has gained momentum due to environmental concerns and challenges associated with insecticide resistance (Lewis *et al.*, 1996). Consequently, entomopathogenic fungi have emerged as promising, environmentally benign insect control agents (Miranpuri and Kachatourian, 1993; Renuka *et al*., 2015). This study endeavors to assess the efficacy of select isolates of entomopathogenic fungi in mitigating maize stem borer damage inflicted by *C. partellus* during winter seasons, while also examining the economic implications of their implementation.

2. Materials and Methods

The field trial targeting maize stem borer, *Chilo partellus*, was conducted at the ICAR-NBAIR, Bengaluru, Karnataka, India, over two consecutive winter seasons. The trial employed four promising isolates of entomopathogenic fungi, namely *Beauveria bassiana* strains Bb-5a, Bb-23, Bb-45, and *Metarhizium anisopliae* strain Ma-35. Employing a completely randomized block design, the experiment encompassed five treatments, each replicated four times. The selected maize hybrid for assessment was Nityashree (NAH-2049), known for its susceptibility to stem borer infestation. Maize seeds were manually dibbled into experimental plots measuring 5×6 m for each replication, maintaining a spacing of 60×30 cm. Standard agronomic practices were adhered to, including recommended fertilizer doses, following the package of practices outlined by the University of Agricultural Sciences, Bengaluru, Karnataka. Application of the oil formulation of the entomopathogenic fungal isolates, diluted in water at a rate of 5ml per liter, was administered twice using a hand sprayer. Treatments, with a spore dose of 1×10^8 cfu/ml, were applied 15 and 30 days after germination during both years.

Subsequently, laboratory-reared *C. partellus* larvae at the second instar stage, at a rate of eight larvae per plant, were carefully introduced into the inner leaf whorl using a camel hair brush, one week after the second spray. Observations on parameters such as the number of dead hearts (DH), extent of stem tunneling (cm/plant) (ST), number of galleries per plant, number of exit holes per plant (EH), and yield metrics were recorded at harvest by longitudinally splitting plants from top to base. Statistical analysis was performed using SPSS v16 software, with the analyzed data subjected to Duncan's Multiple Range Test (DMRT). Treatment-wise cob, grain, and straw yields per plant were recorded and converted to tonnes/quintals per hectare.

Meteorological data for the experimental period over two years included T_{max} (29.12 °C), T_{min} (17.72 °C), RH-I (89.2%), RH-II (47.8%), and RF (191.2 mm) for the first year, and T_{max} (29.54 °C), T_{min} (15.96 °C), RH-I (84.98%), RH-II (39.9%), and RF (74.2 mm) for the second year. These data, obtained from the meteorological observatory at the University of Agricultural Sciences, Bangalore, Karnataka, were utilized for experimental purposes.

Table 1: Pooled effect of different isolates of EPF on maize stem borer, *Chilo partellus*

Isolate	Average No. of dead hearts	Dead heart (%)	No. of galleries/ plant	No. of Exit holes/plant	Stem tunneling/ plant (cm)	Cob yield/ 10 plants (Kg)	Cob yield (t/ha)
Bb-5a	2.62[a] (1.63)	8.7	1.69[a] (1.48)	2.72[a] (1.66)	5.06[a] (2.36)	12.4[a] (3.59)	68.8
Bb-23	4.55[bc] (2.14)	15.2	2.18[a] (1.64)	5.12[bc] (2.27)	7.97[b] (2.91)	9.6[b] (3.18)	53.1
Bb-45	5.50[c] (2.36)	18.3	2.42[ab] (1.71)	5.08[bc] (2.26)	7.17[ab] (2.77)	10.1[ab] (3.26)	56.1
Ma-35	3.69[ab] (1.93)	12.3	2.02[a] (1.59)	3.29[ab] (1.83)	5.43[a] (2.43)	11.7[ab] (3.49)	64.3
Control	6.48[c] (2.56)	21.6	3.23[b] (1.93)	6.72[c] (2.60)	8.93[b] (3.07)	9.5[b] (3.16)	52.5
CD @ 0.05	1.15	-	0.87	2.24	2.53	2.5	-

Note: Means followed by the similar letters in the columns are not significantly different at 5% by DMRT

3. Results and Discussion

The field trial investigating the efficacy of entomopathogenic fungi (EPF) isolates against maize stem borer, *Chilo partellus*, during winter seasons yielded significant results over two consecutive years. Pooled data analysis revealed notable suppression of maize stem borer damage across various parameters compared to the untreated control.

3.1 Dead Hearts

Analysis of dead heart percentages (Table 1) showed that both Bb-5a (8.7 %) and Ma-35 (12.3 %) isolates demonstrated statistically significant reductions in dead heart incidence compared to the untreated control (21.6 %). The observed decrease in dead hearts could be attributed to the endophytic activity of *B. bassiana* and *M. anisopliae* isolates, as reported in previous studies. Endophytic colonization of maize plants by these isolates is known to result in reduced dead heart occurrences.

3.2 Number of Galleries

The pooled data exhibited a reduction in the number of galleries per plant, ranging from 1.69 to 2.42, compared to the untreated control (3.23 per plant). Bb-5a displayed the lowest number of galleries (1.69 per plant), followed by Ma-35 and Bb-23, with similar levels of efficacy. The decrease in gallery formation is likely attributable to the reduced survival of *C. partellus* larvae due to the endophytic activity of the EPF isolates (Table 1).

3.3 Number of Exit Holes

Similarly, the number of exit holes per plant ranged from 2.72 to 3.29 in treated plots, compared to 6.72 in the untreated control (Table 1). Bb-5a exhibited the lowest number of exit holes (2.72 per plant), followed by Ma-35. This reduction in exit holes aligns with the decreased survival of *C. partellus* larvae resulting from the endophytic activity of *B. bassiana* and *M. anisopliae* isolates.

3.4 Stem Tunneling

Stem tunneling measurements ranged from 5.06 to 7.97 cm per plant in treated plots, contrasting with 8.93 cm in the untreated control. Both Bb-5a and Ma-35 isolates showed the least damage, with Bb-5a demonstrating the lowest stem tunneling (5.06 cm per plant). The diminished stem tunneling observed can be attributed to the systematic activity of the EPF isolates within the maize plant system (Table 1).

These findings underscore the potential of *B. bassiana* and *M. anisopliae* isolates in mitigating maize stem borer damage during winter seasons. The observed reductions in various damage parameters corroborate previous research highlighting the efficacy of endophytic EPF isolates in insect pest management.

3.5 Yield

The pooled cob yield data from the experimental trials demonstrated the superior performance of all four tested entomopathogenic fungi (EPF) isolates, with cob yields ranging from 68.8 to 64.3 tonnes per hectare (t/ha). Notably, Bb-5a (68.8 t/ha) and Ma-35 (64.3 t/ha) treatments exhibited the highest yields, significantly surpassing the untreated control (52.5 t/ha), as detailed in Table 1. Regarding grain yield, pooled data analysis revealed significantly higher yields of 52.80 and 51.40 quintals per hectare (q/ha) for Bb-5a and Ma-35 isolates, respectively, representing a 20.27% and 18.09% increase over the control. These results were statistically comparable between the two isolates, as shown in Table 2. Additionally, Bb-45 and Bb-23 isolates recorded grain yields of 50.0 and 48.50 q/ha, respectively, with 15.80% and 13.20% increases over the control. Conversely, the untreated control exhibited the lowest grain yield.

Similarly, the pooled data on maize straw yield highlighted significantly higher yields of 70.10 and 69.17 q/ha for Bb-5a and Ma-35 isolates, respectively, representing increases of 17.65% and 16.54% over the control. These yields were statistically similar between the two isolates

Table 2: Pooled data of cost economics of different isolates of EPF on *Chilo partellus*

Isolates	Grain yield (q/ha)	Per cent increase over control	Straw yield (q/ha)	Per cent increase over control	Gross income (`/ha)	Cost of cultivation (`/ha)	Net profit (`/ha)	Net gain over control (`/ha)	B:C ratio
Bb-5a	52.80[a] (7.30)	20.27	70.1[a] (8.40)	17.65	82660	26000	56660	15384	3.18
Bb-23	48.50[c] (7.00)	13.20	64.05[c] (8.03)	9.87	75860	26000	49860	8584	2.92
Bb-45	50.00[b] (7.11)	15.80	66.13[b] (8.16)	12.70	78226	26000	52226	10950	3.01
Ma-35	51.40[a] (7.20)	18.09	69.17[a] (8.35)	16.54	80654	26000	54654	13378	3.10
Control	42.10[d] (6.53)	00.00	57.73[d] (7.63)	00.00	66276	25000	41276	00000	2.65
CD @ 0.05	1.90	-	2.52	-	-	-	-	-	-

Note: Means followed by the similar letters in the columns are not significantly different at 5% by DMRT

(Table 2). Moreover, Bb-45 and Bb-23 isolates recorded straw yields of 66.13 and 64.05 q/ha, respectively, with increases of 12.70% and 9.87% over the control. The untreated control exhibited the lowest straw yield. The enhanced yields associated with these endophytic isolates may be attributed to their effective suppression of maize stem borer. Previous research by Reddy *et al*. (2009) reported significantly higher yields in sorghum plots treated with endophytic *B. bassiana* compared to untreated controls under artificial *C. partellus* infestation.

3.6 Cost Economics

The cost economics of EPF application against maize stem borer, *C. partellus*, during winter seasons were analyzed and presented in Table 2. Among all the promising EPF isolates, Bb-5a and Ma-35 recorded higher gross returns of ₹ 82660/ha and ₹ 80654/ha, respectively, resulting in maximum net profits of ₹ 56660/ha and ₹ 54654/ha, respectively, which were statistically comparable. Following closely were Bb-45 and Bb-23, with gross returns of ₹ 78226/ha and ₹ 75860/ha, respectively (net returns of ₹ 52226/ha and ₹ 49860/ha, respectively). The lowest profits were recorded in the untreated control, with ₹ 41276/ha.

Regarding the cost-benefit ratio, the highest ratio was observed in Bb-5a (1:3.18), followed by Ma-35 (1:3.10). Conversely, Bb-45 and Bb-23 exhibited lower ratios of 1:3.01 and 1:2.92, respectively, while the untreated control recorded the lowest ratio of 1:2.65 (Table 2). It's worth noting that the cost economics analysis, along with the cost-benefit ratio, represents novel contributions to the field, with no existing comparable data available.

4. Conclusion

In conclusion, the field trials conducted during winter seasons showcased the superior efficacy of Bb-5a and Ma-35 isolates in suppressing maize stem borer, *C. partellus*. These isolates offer promising alternatives to chemical insecticides for pest management, exhibiting eco-friendly attributes with no discernible adverse effects on the environment, human health, or animal health. Consequently, we recommend the adoption of Bb-5a and Ma-35 isolates as viable strategies for stem borer control among farmers, contributing to sustainable agricultural practices and reducing reliance on conventional chemical insecticides. Further research and field trials are warranted to explore their broader applicability and optimize their integration into pest management strategies for maize cultivation.

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Research Article

Comprehensive study on understanding the incidence of Yellow Leaf Disease in Dakshina Kannada districts of Karnataka

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ABSTRACT
Yellow Leaf Disease (YLD) poses a significant threat to arecanut cultivation, particularly in regions like Dakshina Kannada district, Karnataka, India. This study presents a comprehensive survey conducted across three taluks - Puttur, Bantwal, and Sullia to assess the incidence and intensity of YLD. In Puttur Taluk, disease incidence ranged widely from 0 to 100 percent, with Bajathoor and Savanur exhibiting the highest rates. Similar patterns were observed in disease intensity. Conversely, certain villages displayed no YLD intensity, suggesting potential areas for further investigation into disease resistance mechanisms. In Bantwal Taluk, varying levels of incidence and intensity were observed, emphasizing the localized nature of disease dynamics. In Sullia Taluk, a broad spectrum of vulnerability was noted, with Sampaje recording the highest incidence and intensity. However, several villages showed no observable disease incidence, indicating areas of resilience or effective management practices. Overall, the survey underscores the need for targeted interventions to mitigate YLD's impact on arecanut cultivation. These interventions should encompass cultural practices, chemical control measures, breeding for disease resistance, and integrated pest management. Additionally, capacity-building initiatives and farmer education programs are essential for enhancing awareness and adoption of best practices. Addressing the challenges posed by YLD requires a collaborative effort from all stakeholders, guided by scientific research and tailored to local farming communities' specific needs. Through collective action, the resilience of arecanut cultivation in Dakshina Kannada district and beyond can be ensured.

KEY WORDS: Arecanut; Yellow Leaf Disease; Survey; Dakshina Kannada; Incidence; Severity

1. Introduction

Arecanut plays a vital role in cultural and social customs, especially in South and Southeast Asian countries like India, Bangladesh, Myanmar, Thailand and Indonesia. It is commonly offered as a gesture of hospitality, friendship, or respect during social gatherings, ceremonies, and religious rituals. It is often chewed along with betel leaf and slaked lime as a traditional stimulant, particularly in cultural and social contexts (Raghavan and Baruah, 1958). This practice, known as betel chewing, is deeply ingrained in the cultural fabric of many communities and is believed to have stimulant, digestive, and medicinal properties.

Arecanut cultivation is a significant source of livelihood for millions of farmers in regions where it is grown. The crop contributes substantially to the agricultural economy of countries like India, Indonesia, Sri Lanka, and Papua New Guinea. It provides employment opportunities not only in cultivation but also in processing, transportation, and marketing sectors. It is rich in nutrients and bioactive compounds. It contains carbohydrates, proteins, fats, vitamins (particularly vitamin E),

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and minerals like calcium, phosphorus, and iron. However, excessive consumption of arecanut, especially when chewed with other additives like tobacco, can lead to adverse health effects. However, its use in traditional medicine should be approached with caution due to potential health risks associated with excessive consumption. Industrial Uses: Apart from its traditional and cultural significance, arecanut finds applications in various industries. Its extracts are used in the manufacture of dyes, tannins and chemicals. Additionally, husks are utilized for making handicrafts, biofuels, and animal feed.

India is the largest producer and consumer of arecanut in the world, with a total area of 730.82 thousand ha and production of 1208.93 thousand tonnes and productivity of 1654 kg/ha. In Karnataka, 500 thousand ha of area is under arecanut cultivation and production of 950 thousand tonnes with productivity of 1900 kg/ha of nuts were produced during 2020-21 (DASD, 2021). Arecanut, being a major commercial crop in several tropical regions, is susceptible to various diseases that can adversely affect its growth, yield, and quality. Some of the common diseases of arecanut are Yellow Leaf Disease (Phytoplasma); Leaf spot and Inflorescence die back (*Colletotrichum gleosporoides*); Fruit Rot (*Phytophthora arecae*); Leaf Blight (*Pestalotiopsis palmarum*); Bud Rot (*Phytophthora palmivora*) and Foot rot (*Ganoderma lucidum*)

Yellow Leaf Disease (YLD) of arecanut is a significant concern in regions where arecanut cultivation is prominent, such as parts of India, including the Dakshina Kannada district in Karnataka (Hiremata *et al*., 2020). This disease was first reported by Nambiar, 1949, caused by various primarily by phytoplasma, manifests as yellowing of the leaves and leading to considerable yield losses if not managed effectively. Symptoms includes yellowing of leaves, often starts from the lower leaves and gradually progresses upwards. Infected leaves may exhibit yellowing along the margins or throughout the leaf surface, with veins retaining green color initially (Chowdappa *et al*., 1995). As the disease advances, affected leaves may dry up and drop prematurely, leading to defoliation and reduced crown region.

YLD is caused by phytoplasma (a type of bacteria-like organism). Transmission of the disease is primarily by insect vectors, plant hoppers (*Proutista moesta*). YLD significantly reduce the yield and quality of arecanut. Severe infections can lead to stunted growth, reduced nut size and decreased nut production, ultimately affecting the economic viability of arecanut cultivation. Managing Yellow Leaf Disease requires an integrated approach that combines cultural, chemical and biological control measures. These may include the use of disease-resistant varieties, regular monitoring and early detection of symptoms, pruning and removal of infected plant parts and adoption of good agricultural practices to minimize disease spread. Ongoing research and extension efforts are focused on understanding the severity and spread of disease from endemic regions to non-traditional areas. Collaboration between researchers, extension agents and farmers are essential for disseminating information for implementing effective disease management strategies. Keeping all these points in view, the study has been carried out and implemented.

2. Materials and Methods

An intensive roving survey was carried out during 2021-22 to assess the incidence and intensity of yellow leaf disease in major arecanut growing regions of Karnataka *viz.*, Putturu, Bantwal and Sullia taluks of Chikkamagaluru district. At each location five fields were selected randomly and per cent disease incidence was calculated by observing twenty five plants per garden and the intensity was calculated by selecting five plants at each garden by following scoring system (Table 1a, 1b, 1c) given by George *et al.* (1980).

Table 1a: Intensity of yellow leaf disease of arecanut - Scoring for yellowing (Y)

Sl. No.	Features	Scoring
1	Healthy	0
2	Tip yellowing upto 25 % of leaflets	1
3	Tip yellowing upto 50 % of leaflets	2
4	Tip yellowing more than 50 % + marginal yellowing upto 25 %	3
5	Tip yellowing in full + marginal yellowing upto 50 % + Complete yellowing upto 25 %	4
6	Tip yellowing in full + marginal yellowing upto more than 50 % + Complete yellowing upto 50 %	5
7	Tip yellowing in full + marginal yellowing upto more than 50 % + Complete yellowing in full (mild)	6
8	Complete yellowing in full (severe)	7

Table 1b: Intensity of yellow leaf disease of arecanut - Scoring for Necrosis (N)

Sl. No.	Features	Scoring
1	Healthy	0
2	Necrosis upto 50 % of the leaflets	1
3	Necrosis in more than 50% of the leaflets	2

Table 1c: Intensity of yellow leaf disease of arecanut-Scoring for crown size reduction (R)

Sl. No.	Features	Scoring
1	Healthy	0
2	Reduction in size of younger leaves	0.5
3	Reduction in size of younger leaves and stem tapering	1

2.1 Estimation of percent disease incidence

The percent disease incidence was assessed by recording the number of plants showing disease symptoms, out of the total number of plants examined.

Symptoms: Rawther (1976) recorded the characteristic symptoms of the disease as

a. Inter-venial foliar yellowing starting from the tips of leaflets in two to three leaves of the outermost whorl (Y).
b. Necrosis of the leaflets and eventually dry up (N).
c. In advanced stage, leaves are reduced in size, become stiff and pointed, closely bunched and abnormally puckered (R).

Based on the standard symptoms observed during survey, percent disease incidence and intensity was calculated.

The percentage of disease incidence was calculated by using the following formula.

$$\text{Disease Incidence (DI \%)} = \frac{\text{Number of infected plants}}{\text{Total number of plants examined}} \times 100$$

2.2 Assessment for the intensity of yellow leaf disease of arecanut

Mean of twenty-five plants were used to determine the intensity scoring by using formula given below by George *et al*. (1980).

Scoring system to assess intensity in yellow leaf affected gardens

Intensity = $((Y+N)/L + R) \times 10$

Where,
Y = Total scoring for yellowing of the leaves (0-7)
N = Scoring for the necrosis of the leaves (0-2)
R=Scoring for reduction in size of the crown (0-1)
L = Half the number of leaves in the crown

3. Results and Discussion

3.1 Symptomology of the yellow leaf disease of arecanut

Symptom appearance and its presence on the plant can be seen across all the three seasons in a year. However, conspicuous symptoms of YLD were well pronounced immediately after the onset of South-West monsoon rains especially during August-November where, its cessation and maintaining of the green colour by the younger leaves is commonly observed in majority of gardens. Characteristics of yellowing starts from the tip of the leaflets of second or third fronds of the outer most whorls of the palm. Later on yellowing gradually extends to the middle of the lamina showing typical demarcation of yellow and green parallel bands on both sides of the midrib of the leaflets. The leaves become stiff and pointed, closely bunched and abnormally puckered. As the disease progresses, yellowing extends to the whole lamina and leaf tips become necrotic and dries up during summer leaving the leaf stalk green. Subsequently, symptoms appeared on both mature and immature nuts wherein, kernel size gets reduced, discoloration and rotting of kernels and non production of inflorescence and if produced, drying up of the entire inflorescence was also observed. At the advanced stage, crown size reduction, reduction in the inter nodal length and tapering of the stem followed by decapitation of the entire crown leaving a bare trunk were conspicuous. Affected plant roots also exhibit brown to black colour discoloration (Fig.1). By observing these symptoms, incidence and intensity of palms were calculated.

Fig. 1: Typical Symptoms of yellow leaf disease of Arecanut

3.2 Survey for the incidence and intensity of yellow leaf disease of arecanut in Chikkamagaluru district

Arecanut is a cornerstone of agriculture in Dakshina Kannada district, Karnataka, India. However, the cultivation of this economically significant crop faces substantial challenges due to various diseases, with Yellow Leaf Disease (YLD) being one of the most prevalent and detrimental.

Understanding the extent and severity of YLD across different taluks and districts is paramount for effective disease management and sustainable arecanut cultivation practices. This study presents the comprehensive findings of a survey conducted to assess YLD incidence and intensity in three taluks of Dakshina Kannada district was represented in Table 2.

In Puttur Taluk, the survey unveiled a wide spectrum of disease incidence, ranging from 0 to 100 percent, indicating varied vulnerability levels among different areas. Notably, Bajathoor exhibited the highest disease incidence at 100 percent, closely followed by Savanur at 95 percent. Concurrently, disease intensity exhibited a similar pattern, ranging from 0 to 97.50 percent, with Bajathoor recording the highest intensity at 97.50 percent, trailed closely by Savanur at 92.50 percent. Remarkably, certain villages such as Barike, Navoor, Nekkare, Kochhi, and Punchatharu displayed no intensity of YLD, signaling potential areas for further study on disease resistance mechanisms.

The survey in Bantwal Taluk uncovered disease incidence varying from 0 to 40.00 percent, with corresponding intensity levels ranging from 0 to 30.00 percent. Notably, Urimajalu emerged as the village with the highest disease incidence at 50.00 percent, closely followed by Ananthady at 40.00 percent. However, intensity levels displayed a different trend, with Urimajalu recording the highest intensity at 30.00 percent, followed by Ananthady at 27.50 percent. These findings underscore the localized nature of disease dynamics within the taluk and highlight the need for tailored management strategies.

In Sullia Taluk, disease incidence spanned from 0 to 100.00 percent, showcasing a broad spectrum of vulnerability across surveyed areas. Sampaje registered the highest disease incidence at 100.00 percent, accompanied by a significant intensity of 95.00 percent. Additionally, villages such as Panja, Alletti, and Guttigar exhibited a disease incidence of 95.00 percent, emphasizing the widespread nature of YLD within the taluk. Conversely, several villages including Yenekallu, Parla, Nadugallu, Balladi, Panjipalla, and Sannara displayed no observable disease incidence, suggesting potential areas of resilience or effective disease management practices.

4. Conclusion

The findings of this survey shed light on the significant challenges posed by Yellow Leaf Disease (YLD) in arecanut cultivation across the taluks of Puttur, Bantwal, and Sullia in Dakshina Kannada district, Karnataka. The survey revealed a diverse landscape of YLD incidence and intensity across different areas within the surveyed taluks.

In Puttur Taluk, a wide spectrum of disease incidence ranging from 0 to 100 percent was observed, with Bajathoor and Savanur emerging as the areas with the highest incidence rates. Similarly, disease intensity exhibited a similar pattern, with Bajathoor and Savanur recording the highest intensities. Notably, certain villages displayed no intensity of YLD, indicating potential areas for further investigation into disease resistance mechanisms.

In Bantwal Taluk, the survey uncovered varying levels of disease incidence and intensity, highlighting the localized nature of disease dynamics within the taluk. Urimajalu and Ananthady emerged as the villages with the highest disease incidence, with intensity levels reflecting a different trend. These findings underscore the need for tailored management strategies to address the localized nature of YLD outbreaks.

Table 2: Prevalence and severity of yellow leaf disease across different areca nut plantations in Dakshina Kannada district

District	Taluk	Village	Latitude	Longitude	Variety	Age of the crop	Area cultivated (ha)	Other Pests, diseases	Per cent Disease Incidence	Intensity (%)
Daksihna Kannada	Puttur	Barike	12.7939	75.4157	Mangala	28	0.08	LS, RR	0.00	0.00
		Navoor	12.7064	75.3870	Mangala	20	0.45	LS, SI	0.00	0.00
		Idyadi	12.7593	75.3075	Mangala	50	1.29	ND	10.00	6.00
		Panemajalu	12.7497	75.3025	South Kanara	45	0.55	LS, ND	10.00	14.00
		Golitottu	12.8478	75.3591	Mangala	28	0.61	LS	10.00	0.00
		Kumla	12.8544	75.3569	Mangala	30	1.34	ND	50.00	52.50
		Kuchila	12.8629	75.3637	Mangala	35	0.08	LS, RR	10.00	12.00
		Nekkare	12.8511	75.3664	Mangala	45	0.40	LS	0.00	0.00
		Kochhi	12.8487	75.3744	Mangala	56	0.08	ND	0.00	0.00
		Haradi	12.7736	75.1963	South Kanara	34	0.49	LS, SI	65.00	55.00
		Bajathoor	12.8416	75.2957	South Kanara	30	0.49	LS, CC	100.00	97.50
		Madody	12.7129	75.3586	South Kanara	45	0.40	LS	85.00	87.50
		Savanur	12.7417	75.3083	South Kanara	38	0.49	SI	95.00	92.50
		Punchatharu	12.7096	75.3815	South Kanara	35	0.13	LS	0.00	0.00
		Mogaru	12.9619	74.9694	South Kanara	50	0.13	LS, RR	85.00	67.50
		Kodaka	12.8782	75.4191	Mangala	56	1.36	ND	10.00	12.00
		Kovadi	12.8786	75.4192	Mangala	35	1.69	RR	10.00	0.00
		Uruvalu	12.8667	75.2848	Mangala	34	1.23	LS, SI	10.00	25.00
		Ilanthila	12.8583	75.2769	Mangala	32	0.43	ND	16.00	28.00
		Marakkada	12.7318	75.3663	Mangala	35	0.12	SI	23.00	45.00
		Kaniyoor	12.7165	75.3678	Mangala	43	0.87	ND	14.00	35.00
		Munur palike	12.8765	75.3550	Mangala	51	0.77	RG	17.50	43.00
		Upparapalike	12.8557	75.3748	Mangala	28	0.34	SI	8.00	25.00
		Alanthya	12.8272	75.3602	Mangala	26	0.32	SI, RG	6.00	21.00
		Shirady	12.8270	75.5303	Mangala	41	0.12	ND	2.00	15.00

*CC-Crown chocking, QW-Quick wilt of pepper, LS-Leaf spot, RR-Root rot, RG-Root grub, BR-Bud rot, ND-Nut drop, KR-Kole roga, SI-Sucking insects

Contd...

Table 2: Prevalence and severity of yellow leaf disease across different areca nut plantations in Dakshina Kannada district (Contd..)

District	Taluk	Village	Latitude	Longitude	Variety	Age of the crop	Area cultivated (ha)	Other Pests, diseases	Per cent Disease Incidence	Intensity (%)
Daksihna Kannada	Bantwal	Mithur	12.7831	75.1400	Mangala	25	1.69	LS, RG	0.00	0.00
		Surya	12.7889	75.1304	Mangala	30	1.69	LS	0.00	0.00
		Netlamudnur	12.7927	75.1310	Mangala	35	0.12	KR, SI	0.00	0.00
		Karinka	12.7919	75.1172	Mangala	28	0.06	LS	10.00	6.00
		Nadumane	12.7939	75.1131	Mangala	39	0.69	LS, RG	0.00	0.00
		Irandoor	12.7897	75.1102	Mangala	45	0.48	LS	30.00	14.00
		Vittalpadanur	12.7791	75.1048	Inter Mangala	36	1.77	LS, SI	0.00	0.00
		Naithottu	12.7960	75.1070	Inter Mangala	45	1.77	ND, RR	10.00	6.00
		Ananthady	12.8022	75.1152	Inter Mangala	50	1.01	LS	40.00	27.50
		Sankesha	12.8035	75.1095	Inter Mangala	55	1.13	LS, RG	0.00	0.00
		Hancharike	13.4575	75.2484	Inter Mangala	45	0.40	ND	10.00	6.00
		Badakodi	12.7191	75.1272	Inter Mangala	65	0.81	SI, RG	0.00	0.00
		Kodimbala	12.7215	75.4752	Mangala	40	0.61	LS, RR	0.00	0.00
		Urimajalu	12.7842	75.1398	Mangala	34	0.81	RG	50.00	30.00
		Kesarukodige	13.4676	75.2533	Mangala	28	0.96	LS, SI	0.00	0.00
		Punja	12.6786	75.4724	Mangala	25	1.77	LS, RG	0.00	0.00
		Addabailu	12.6785	75.4919	Mangala	20	0.81	ND	0.00	0.00
		Kukkipadi	12.9754	75.0691	Mangala	25	0.21	LS, ND	0.00	0.00
		Kedila	12.8064	75.1619	Mangala	35	0.28	LS, RG	2.00	12.00
		Peraje	12.8208	75.1481	Mangala	34	0.19	ND	2.00	10.00
		Rayee	12.9464	75.0554	Mangala	45	1.12	ND	0.00	0.00
		Sarapady	12.8790	75.1276	Mangala	38	1.25	RG, ND	0.00	0.00
		Budoil	12.9483	75.0721	Mangala	28	0.76	ND	2.00	10.00
		Chennaithodi	12.9718	75.0995	Mangala	23	1.23	LS, ND	0.00	0.00
		Mudanadugodu	12.9090	75.0705	Mangala	43	1.40	RG	0.00	0.00

*CC-Crown chocking, QW-Quick wilt of pepper, LS-Leaf spot, RR-Root rot, RG-Root grub, BR-Bud rot, ND-Nut drop, KR-Kole roga, SI-Sucking insects

Contd…

Table 2: Prevalence and severity of yellow leaf disease across different areca nut plantations in Dakshina Kannada district (Contd..)

District	Taluk	Village	Latitude	Longitude	Variety	Age of the crop	Area cultivated (ha)	Other Pests, diseases	Per cent Disease Incidence	Intensity (%)
Daksihna Kannada	Sullia	Yenekallu	12.6738	75.5649	Mangala	25	0.81	RR	0.00	0.00
		MuthlajeBailu	12.6751	75.5655	Mangala	30	0.81	LS, RG	10.00	17.00
		Pundigadde	12.6614	75.5612	Mangala	38	0.89	ND	5.00	10.00
		Parla	12.6732	75.5547	Mangala	45	1.77	ND, RR	0.00	0.00
		Nadugallu	12.6455	75.5661	Inter Mangala	23	0.81	LS	0.00	0.00
		Balladi	12.6693	75.5509	Inter Mangala	45	0.49	LS	0.00	0.00
		Halemajalu	12.6431	75.5506	Inter Mangala	47	0.12	ND, KR	12.00	16.00
		Panjipalla	12.6381	75.5407	Mangala	50	0.15	LS	0.00	0.00
		Mettinadka	12.6190	75.5478	Inter Mangala	54	0.28	LS	20.00	26.00
		Salthadi	12.6163	75.5454	Mangala	34	0.15	LS, KR	10.00	12.00
		Santhinagar	12.6907	75.4080	Mangala	33	0.81	LS, RG	30.00	29.00
		Budengi	12.6889	75.5465	Mangala	45	0.51	ND	40.00	37.00
		Sannara	12.6954	75.5551	Mangala	27	0.18	LS, RR	0.00	0.00
		Sampaje	12.4938	75.5669	Mangala	65	1.23	SI	100.00	92.00
		Panja	12.6801	75.4742	Mangala	28	0.76	LS, RG	95.00	87.50
		Harihara pallathodka	12.6043	75.6108	Mangala	56	0.92	LS	27.00	32.00
		Adyadka	12.4863	75.4768	Mangala	34	1.17	LS, SI	79.00	52.50
		Aranthodu	12.5157	75.4636	Mangala	51	0.33	LS, ND	91.00	61.00
		Guttigar	12.6304	75.5261	Mangala	35	0.54	RG	95.00	63.00
		Kollamogru	12.5758	75.6109	Mangala	45	0.12	LS, RG	82.00	67.00
		Madapadi	12.5791	75.5381	Mangala	56	0.91	RG	87.00	63.00
		Murulya	12.6856	75.4187	Mangala	67	0.43	LS, SI	84.00	70.00
		Marakanja	12.5630	75.5059	Mangala	29	0.33	ND	89.00	80.00
		Alletti	12.5330	75.3747	Mangala	35	0.32	LS	95.00	80.00
		Morangallu H. C	12.5345	75.3843	Mangala	52	1.30	SI, RG	83.00	70.00

*CC-Crown chocking, QW-Quick wilt of pepper, LS-Leaf spot, RR-Root rot, RG-Root grub, BR-Bud rot, ND-Nut drop, KR-Kole roga, SI-Sucking insects

In Sullia Taluk, the survey showcased a broad spectrum of vulnerability to YLD across surveyed areas. Sampaje recorded the highest disease incidence and intensity, emphasizing the widespread nature of YLD within the taluk. However, several villages displayed no observable disease incidence, suggesting potential areas of resilience or effective disease management practices.

Overall, the comprehensive findings of this survey underscore the importance of understanding the extent and severity of YLD for effective disease management and sustainable arecanut cultivation practices. By identifying areas of high vulnerability and localized disease hotspots, this study provides valuable insights for targeted interventions aimed at mitigating the impact of YLD on arecanut cultivation in Dakshina Kannada district. These strategies should encompass a combination of cultural practices, chemical control measures, breeding for disease resistance, and integrated pest management approaches. Additionally, capacity-building initiatives and farmer education programs are crucial for enhancing awareness and adoption of best practices to mitigate the impact of YLD and ensure the resilience of arecanut cultivation in the region. In conclusion, addressing the challenges posed by YLD requires a concerted effort from all stakeholders, guided by scientific research and tailored to the specific needs of the local farming communities. Through collective action and sustained commitment, we can safeguard the future of arecanut cultivation in Dakshina Kannada district and beyond.

The results indicate the probable association of various soil and environmental factors contributing for highest disease incidence in these districts. As the arecanut crop is cultivated from centuries its extensive cultivation especially in hilly tracts which receives an average annual rainfall of 3573 mm during South-West monsoon hastens the intensity of the disease to be endemic in nature. These results were in accordance with the results obtained by Krishnamurthy and Vajranabhaiah (2000) who reported the 24.4 to 40.00 per cent disease incidence in Sringeri and coastal zones (Sampaje belt) of Karnataka.

5. Conflict of interest

The authors have no conflict of interest.

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Research Article

Consequences of Niger (*Guizotia abyssinica*) crop on diversity of entomofauna

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ABSTRACT

Niger is primarily farmed during rainy season on about 0.52 million acres of land in India. Over 80% of the acreage and output is contributed by Madhya Pradesh, Maharashtra, and Orissa. Numerous beneficial traits, including as high biomass potential, tolerance to marginal soils, and pollinator appeal, make Niger *Guizotia abyssinica* an ideal dicot species to coexist alongside perennial warm-season grasses. Study illustrates the variety of visitors to flowers. The information was gathered through direct observation on two days a week for three months, from 8 am to 4 pm. There were 1162 individuals in all, representing 45 species, in five orders and 27 families. The family Apidae has greater number of individuals compared to any other family, followed by family formicidae and family lycaenidae. There has to be a greater environmental knowledge today regarding practical habitat management that could contribute to an increase in insect pollinators. In order to preserve pollinators and to fully utilize the potential of crop pollination, exploratory study should also be conducted.

KEY WORDS: Agroecosystem; Biodiversity; Biomass; Ecosystem; Niger; Pollinators

1. Introduction

An oilseed crop known as Niger, or *Guizotia abyssinica*, is mostly grown as a marginal crop in India. It makes up 3% in India. It is grown in the Indian states of Andhra Pradesh, Madhya Pradesh, Orissa, Maharashtra, Bihar, Karnataka, Nagar Haveli, and West Bengal; Madhya Pradesh is the biggest of these. It is a dicotyledonous herb with a 2 m maximum height and moderate to good branching. The niger blossom's predominant colour is yellow with a touch of green. Ray florets in the heads range in size from 5 to 20 mm in length to 15 to 50 mm in diameter. Each of the two to three growing capitulate (heads) has a ray. The semi-spherical container has a diameter of 1-2 cm and a height of 0.5-0.8 cm. Involucral bracts are arranged in two rows on either side of the receptacle. The capitulum is composed of six to eight healthy female ray florets with narrowly elliptic, obovate ovules. The stigma is comprised of two 2 mm long coiled branches. The hermaphrodite disc florets, which normally number 40 to 60 per capitulum, are organised into three whorls. The disc florets have yellow to orange anthers, and the stigma is quite hairy.

Niger is typically grown in light, poor soils with a gritty texture. The Niger is mostly pollinated by insects, particularly bees, because it is a totally out-crossing plant with a self-incompatibility mechanism (Venkataramegowda *et al*., 2013). As a result of the mixing of one or more forage species, which may promote biodiversity and offer

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habitat for insects and thus improve the pollination services in agro ecosystems, ecosystem goods and services from biomass commodity production systems rose considerably (Patil and Jagdale, 2021) Insects that visit Niger flowers range widely, from Collembola to Hymenoptera (Kevan and Baker, 1984). Insects harvest nectar, pollen, floral tissues, shelter, partners, and oviposition locations from flowers.

Niger is a bioenergy crop that offers more environmental services, such as food and shelter for open-range species like birds and insects, and as a result might be a better option than huge biogas facilities because it is more advantageous to the environment. Plant and pollinator populations may drop in human-modified landscapes due to habitat loss and fragmentation, which may restrict pollination (Kremen, 2004). Threats to insect pollinators include loss of habitat, altered land use, excessive pesticide usage, and modern farming techniques.

The current study sought to examine the effects of the Niger on insect diversity because some plants can still support biodiversity. This work is significant because it has increased understanding of pollinators on the Niger in the Satara district, which will aid in the planning and execution of plant and pollinator conservation.

2. Materials and Methods

We investigated a seed-producing crop whose oil is generally extracted from the seeds. The investigation was conducted in the Karad, Khatav, and Patan tehsil of the Satara district's agricultural fields (Plate 2). Southern Maharashtra state of India. Karad Tehsil. The coordinates of Karad are 17.285°N 74.184°E. It measures 566 metres on average. The coordinates of Khatav are 17.6545°N 74.3614°E. It is 777 metres above sea level on average. The coordinates of Patan Tehsil are 17.37°N 73.9°E. It is 582 metres above sea level on average. Less rain fell on the agricultural fields, which had a 29°C temperature. The niger plant was grown in four distinct fields. There were roughly 2023.428 square metres of fields. From August through November 2020, prime flowering times, the investigation was carried out. For the pollinator survey, we set aside two 5 × 5 m plots at each location, one in each field.

Two times per week, during the three observation periods per day of 8 a.m. to 11 a.m. for the morning, 12 a.m. to 1 p.m., and 2 p.m. to 4 p.m. for the afternoon, pollinator visits to flowers were recorded. The following methods were used to count and gather data on the number of insects that visited the Niger flower reproductive whorls only.

Photos taken with a Canon EOS 200 camera were examined for evidence of flower visits.

1. Sweep net: Species were collected using a sweep net after being photographed and observed to determine how frequently they were visited. Sweeping insects were collected and put in plastic containers. The insects were collected, categorized, labeled, and preserved either dry-pinned or in 70% alcohol.

2. Collection by hand: Hand-collected insect pollinators were then placed in lethal bottles. The insects were prepared for pinning and kept in dry condition on a wooden box.

During their flowering season, the number of various insect pollinators of niger was investigated. Twenty flowers were chosen at random. A timer was used to time the number of different insects that visited all of the niger blooms in a square metre of space for two minutes each plant, every hour from 8 am to 4 pm. The information was saved for later analysis.

2.1 Identification

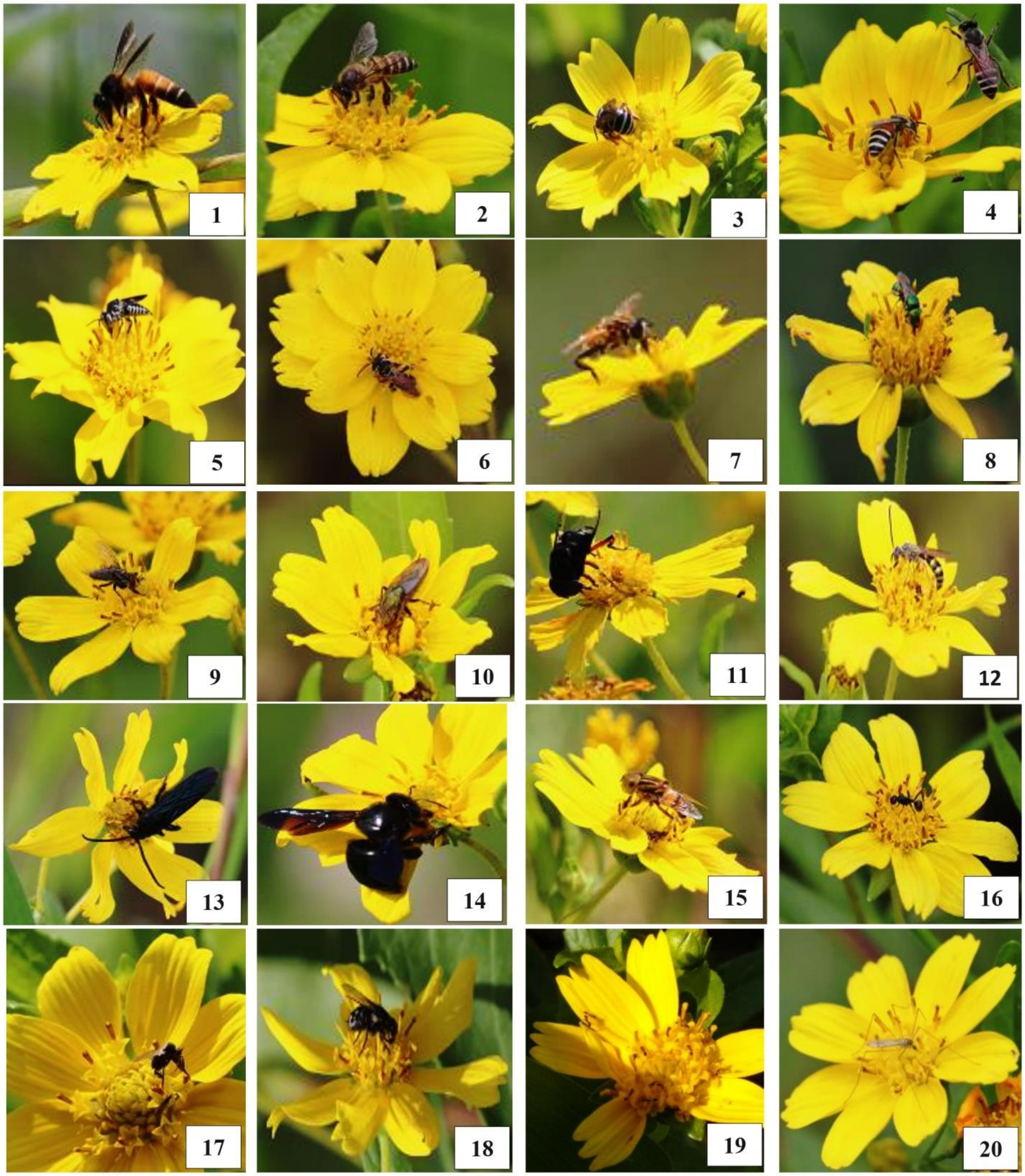
Collected insect pollinators were identified using standard manuals and keys found in Ananthkrishnan and David (2004).

3. Results and Discussion

In this study, 1162 individuals from 45 species (Plate 1a, 1b and 1c), representing five orders and 28 families, were found. These orders of insect visitors included the order Hymenoptera, (Apidae, Megachilidae, Halictidae, Formicidae, Thynnidae, Vespidae), Lepidoptera (Lycanidae, Pieridae, Nymphalidae, Crambidae, Choreutidae, Erebidae), Diptera (Muscidae, Syrphidae, Sphecidae, Tachinidae, Sarcophagidae, Rhiniidae, Limoniidae, Asilidae), Hemiptera (Pentatomidae, Coreidae,), Coleoptera (Scarabidae, Chrysomelidae, Phalacridae, Meloidae, Coccinellidae) (Table 1).

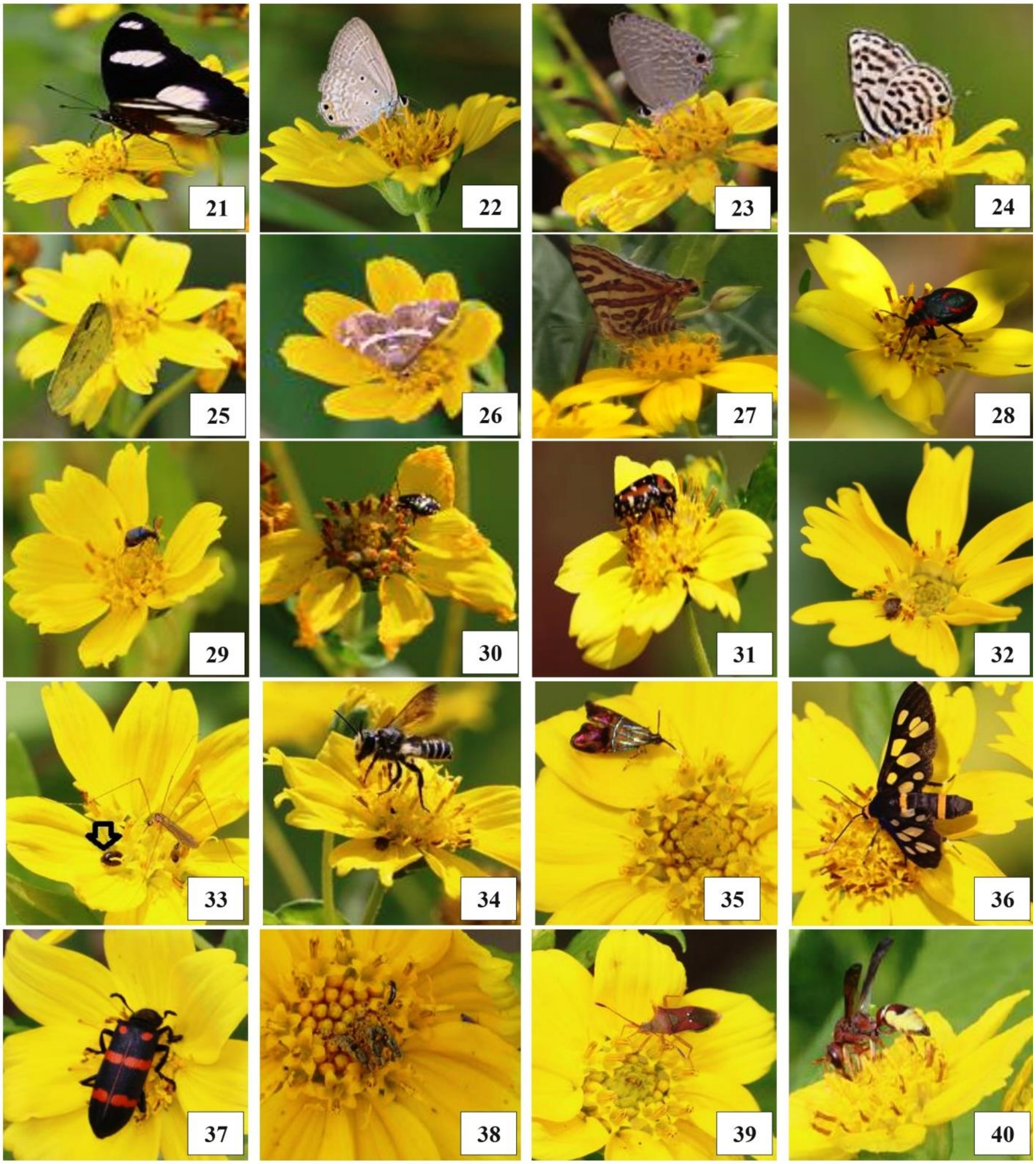
During the study, over different locations, all the insect pollinators observed were belonged to forty–five species, forty-one genera of twenty-eight families under five orders (Fig. 1). A maximum number of pollinator species belonged to the order Hymenoptera (sixteen species), followed by the order Diptera (ten species) followed by the order Lepidoptera (nine species) followed by Hemiptera (three species) and Coleoptera (seven species). Among the families, Apidae was found to be the abundant one comprising of seven species namely, *Apis dorsata, Apis cerana, Apis florea, Apis andreniformis, Amegilla cingulata, Xylocopa latipes, Triepeolus eliseae*. Followed by Lycaenidae comprising four species namely, *Chilades pandava, Nacaduba kurava, Castalius rosimon, Spindasis vulcanus*. The two families shared three species each *i.e*. Formicidae (*Camponotus pennsylvanicus, Solenopsis invicta, Formica fusca*) and Syrphidae (*Episyrphus balteatus, Eristalinus arvorum, Chalcosyrphus femoratus*). Four families shared two species each i.e. Megachilidae (*Coelioxys elongata, Megachile inimica*) Family Pentatomidae (*Euthyrhynchus floridanus, Nezara viridula nymph*), family Chrysomelidae (*Bruchidius villosus, Acanthoscelides obtectus*) and family Meloidae (*Mylabris pustulata, Mylabris phalerata*. Remaining families shared only one species each *i.e*. Halictidae comprising (*Augochlora pura*), Sphecidae (*Chalybion californicum*), Thynnidae (*Myzinum quinquecinctum*) Vespidae (*Rhynchium oculatum*), Asilidae (*Mallophora leschenault*), Muscidae (*Musca domestica*), Rhiniidae (*Stomorhina lunata*), Sarcophagidae *(Sarcophaga bercaea*), Tipulidae (*Limonia phragmitidis*), Tephritidae (*Trupanea crassipes*), Tachinidae (*Carcelia iliaca*), Nymphalidae (*Hypolimnas bolina*), Pieridae (*Eurema hecabe*), Crambidae (*Spoladea recurvalis*), Choreutidae (*Saptha cypridia*), Erebidae (*Amata bicincta*), *Coreidae* (*Gonocerous acuteangulatus*), Scarabidae (*Gametis versicolor*), Phalacridae (*Phalacrus fimetarius*) and *Coccinellidae* (*Menochilus sexmaculata*).

Painkra *et al*. (2015) found 15 species of insect pollinators / visitors on niger flowers namely, *Apis florea* and *Apis dorsata, Danaus chrysippus, Eristalis* sp., *Musca domestica, Nazara virudula, Dysdercus cingulatus, Coccinella septumpunctata, Vespa cincta, Leptocorisa acuta, Amata passelis, Pelopidas mathias, Sarcophaga* sp. and *Chrysomya* sp.



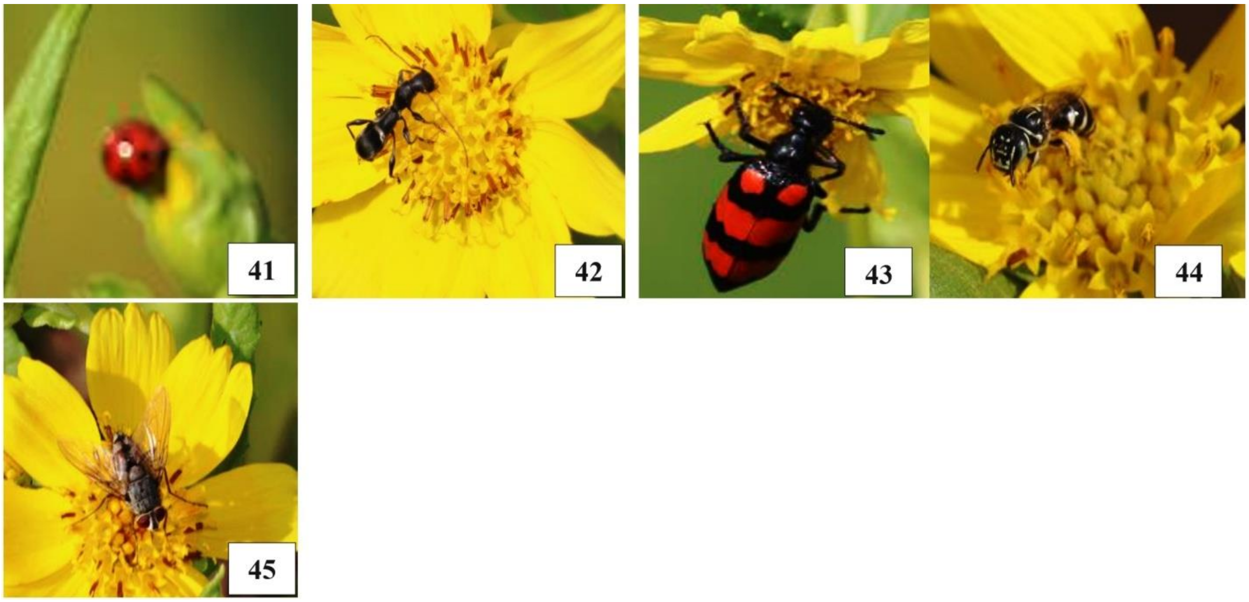
Legend: 1. *Apis dorsata*, **2**. *Apis cerana*, **3**. *Amegilla cingulate*, **4**. *Apis florea*, **5**. *Coelioxys elongate*, **6**. *Apis andreniformis*, **7**. *Episyrphus balteatus*, **8**. *Augochlora pura*, **9**. *Musca domestica*, **10**. *Stomorhina lunata*, **11**. *Chalcosyrphus femoratus*, **12**. *Myzinumquin quecinctum*, **13**. *Chalybion californicum*, **14**. *Xylocopa latipes*, **15**. *Eristalinus arvorum*, **16**. *Camponotus pennsylvanicus*, **17.** *Mallophora leschenault*, **18.** *Sarcophagabercaea*, **19.** *Solenopsis invicta*, **20.** *Limonia phragmitidis*

Plate 1a: Insect pollinators of Black seed /Niger (*Guizotia abyssinica*).



Legend: 21. *Hypolimnas bolina,* **22.** *Chilades pandava,* **23.** *Nacaduba kurava,* **24.** *Castalius rosimon,* **25.** *Eurema hecabe,* **26.** *Spoladea recurvalis,* **27.** *Spindasis vulcanus,* **28.** *Euthyrhynchus floridanus,* **29.** *Bruchidius villosus,* **30.** *Nezara viridula* Nymph, **31.** *Gametis versicolor,* **32.** *Acanthoscelides obtectus,* **33.** *Phalacrus fimetarius,* **34**. *Megachile inimical,* **35.** *Saptha cypridia,* **36.** *Amata bicincta,* **37.** *Mylabris pustulata,* **38.** *Trupanea crassipe,* **39.** *Gonocerous acuteangulatus,* **40.** *Rhynchium oculatum*

Plate 1b: Insect pollinators of Black seed /Niger (*Guizotia abyssinica*).



Legends: 41. *Menochilus sexmaculata*, **42**. *Formica* sps., **43**. *Mylabris phalerata*, **44**. *Triepeolus eliseae*, **45**. *Carcelia iliaca*

Plate 1c: Insect pollinators of Black seed /Niger (*Guizotia abyssinica*).

Thangjam *et al*. (2018) have recorded 19 species of insect pollinators visiting niger were belonged to ten families under five orders *viz*., Hymenoptera, Diptera, Coleoptera, Hemiptera and Lepidoptera. *Andrena* sp., *A. florea, Apis dorsata, A. ceranaindica, Phytomia zonata, Eristalinus taeniops, E. punctulatus, Eristalinus arvorum, Erisyrphus balteatus, Micraspis discolor, Harmonia dimidiata, Popillia* sp., *Aulacophora foveicollis, Dysdercus cingulatus, Tajuria cippus, Chilades* sp., *Pieriscanidia indica, Nyctemera* sp. and *Amata* sp.

The order Hymenoptera made up 78% of the species, followed by the orders Diptera (8%) and Coleoptera (7%), which were then followed by the orders Lepidoptera (5%) and Hemiptera (2%). (Fig. 2) Gebremedhn *et al*. (2014) observed that insect visitors from more than 11 species, representing 4 Orders and 7 Families, on *G. abyssinica* flowers. These observations supported the findings of Gebremedhn *et al*. (2014), who measured the abundance of insect pollinators in Ethiopia and discovered that the Order Hymenoptera had the highest abundance (81.6%), followed by Diptera (12%). Honeybees, in particular, accounted for 79% of all insect visitors. They also discovered that the time of day and flowering period had an impact on the abundance and diversity of insect pollinators. According to Kachhela and Pastagia (2018), *A. dorsata* was the most common flower visitor in Niger (9.33 bees/m^2/5 minute), accounting for 60.74 percent of total flower visitors, followed by *A. florea* (2.30 bees/m^2/5 minute) and *A. cerana* (1.17 bees/m^2/5 minute), accounting for 14.97 and 7.62 percent of total flower visitors, respectively.

Thangjam *et al*. (2018) recorded that *A. cerana* was dominant forager (41.95%) followed by *A. florea* (20.13%) and *A. dorsata* (19.80%).

Table 1: Observations of number and nature of insect visitors on Niger plant.

Sl. No.	Order	Family	Insect Visitors	No. of insect visitor	Nature of insect visitor
1.	Hymenoptera	Apidae	*Apis melifera*	125	PO(P,N)
			Apis dorsat1a	85	PO(P)
			Apis florea	425	PO(N,P)
			Xylocopa latipes	9	PO(P,N)
			Amegilla cingulata	13	PO(N,P)
			Apis andreniformis	82	PO(P)
			Triepelous sps.	11	PO(P,N)
		Megachilidae	*Coelioxys elongata*	03	PO(P,N)
			Megachile inimica	02	PO(P)
		Halictidae	*Augochlora pura*	17	PO(P,N)
		Formicidae	*Camponotus pennsylvanicus*	19	PO(N,EXF N)
			Solenopsis invicta	103	PO(N)
			Formica fusca.	11	PO(P)
		Thynnidae	*Myzinum quinquecinctum*	05	PO(N)
		Vespidae	*Vespa sps*	02	PR
2.	Lepidoptera	Lycanidae	*Nacuduba kuravaNemana*	6	PO(N)
			Castalius rosimon	04	PO(N)
			Chilades pandava	13	PO(N)
			Spindasis vulcanus	07	PO(N)
		Pieridae	*Eurema hecabe*	15	PO(N)
		Nymphalidae	*Hypolimna bolina*	02	PO (N)
		Crambidae	*Spoladea recurvalis*	05	PO(N)
		Choreutidae	*Saptha cypridia*	02	PO(N)
		Erebidae	*Amata bicincta*	02	PO(N)
3.	Diptera	Muscidae	*Musca domestica*	08	PO(N)
		Syrphidae	*Chalcosyrphus femoratus*	02	PO(N)
			Episyrphus balteatus	05	PO(N,P)
			Eristalinus arvorum	11	PO(P,N)
		Sphecidae	*Chalybion californicum*	21	PO(N)
		Tachinidae	*Carcelia sps.*	11	PO(P)
		Sarcophagidae	*Sarcophaga bercaea*	03	PA
		Rhiniidae	*Stomorhina lunata*	06	PO(N,P)
		Limoniidae	*Limonia phragmitidis*	19	PO(N)
		Asilidae	*Mallophora leschenaulti*	02	PR
4.	Hemiptera	Pentatomidae	*Nezara viridula nymph 3rd instar*	05	B
			Euthyrhynchus floridanus	11	PR
		Coreidae	*Gonocerousacuteangulatus*	03	PO(B)
5.	Coleoptera	Scarabidae	*Gametis versicolor*	26	PO(P)
		Chrysomelidae	*Acanthoscelides obtectus*	09	PO(P)
			Bruchidius villosus	31	PO(P)
		Phalacridae	*Phalacrus fimetarius*	08	PO(P)
		Meloidae	*Mylabris pustulata*	05	PO(P)
			Mylabris phalerata	02	PO(P)
		Coccinellidae	*Menochilus sexmaculata*	06	PO(P)

B= Basking, PO(P)=Pollinators who visits flower for Pollen, N= Nectar, EXF N= Extrafoliar Nectar), PA=Parasitoid, PR=Predator

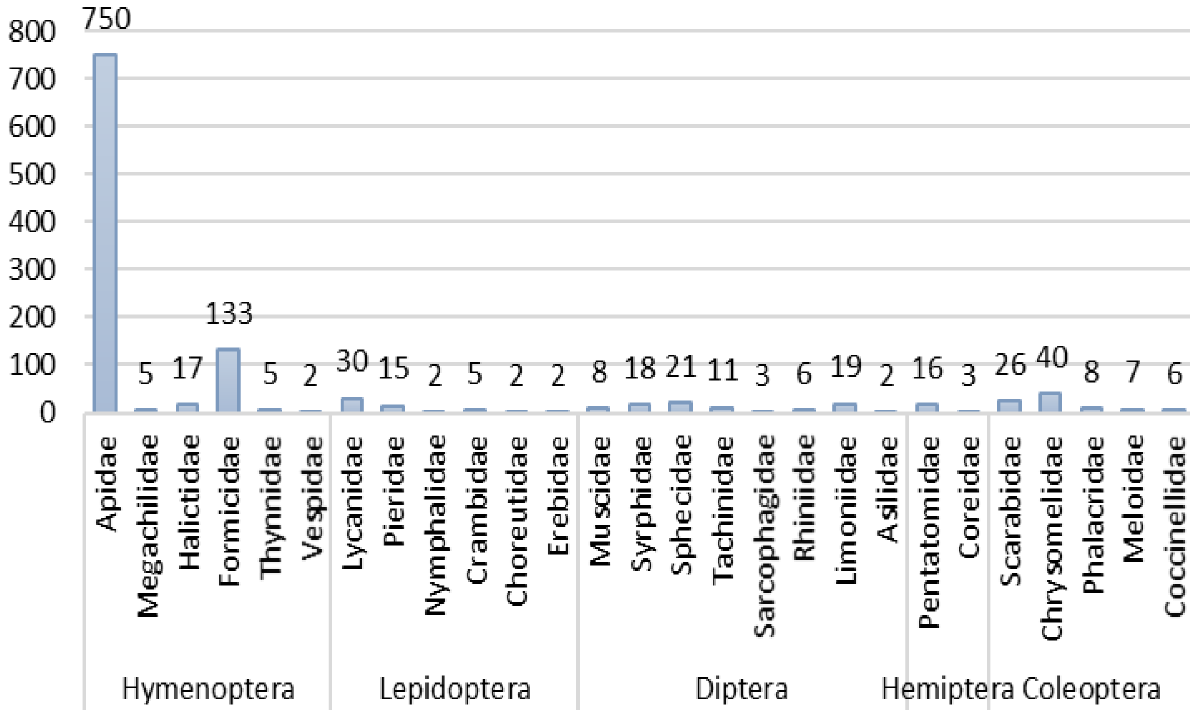


Fig. 1 Order and Family-wise insect visitors on *Guizotia abyssinica*

Tiwari *et al*. (2020) recorded *Apis cerana indica* (19.55), *Apis dorsata* (13.67), *Apis mellifera* (7.24), and *Apis florea* (1.81) as the most abundant pollinators. They also recorded Monarch butterfly – *Danaus chrysippis* (4.22), Rice skipper - *Pelopidas mathias* (4.88), Wasp - *Vespa cincta* (1.95), House fly - *Musca domestica* (3.28), Syrphid fly - *Eristalis* sp. (5.50), Blow fly- *Chrysomya bezziana* (1.82), Red cotton bug- *Dysdercus cingulatus* (1.97), and Tiger moth - *Amata passelis* (2.44) visiting on niger flower throughout the flowering period during rainy season.

The majority of insect visitors were found to visit flowers for pollen and nectar; very few insects visited flowers for warmth. Insects that visited flowers in search of prey included parasitoids and predators. For pollinators to complete their life cycle, food supplies, nesting materials, and nest sites must all be accessible (Potts, 2005; Winfree, 2010). The conservation of natural pollinators and the reproduction of planted species are better served by larger wildflower plantings with more varied flower species mixtures. Pollinator visitation rates fluctuated throughout the season, peaking around the time of flowering, according to Brett and Rufus (2014).

The richness of the associated invertebrate populations and for the birds depends greatly on the development of the ground vegetation within the plantations (Sage *et al.*, 2006; Semere and Slater, 2007; Bellamy *et al.*, 2009; Valentin *et al.*, 2009). Many of these contain pest species that might harm the plantation, including leaf-eating beetles, Coleoptera: Chrysomelidae (Sage, 2008).

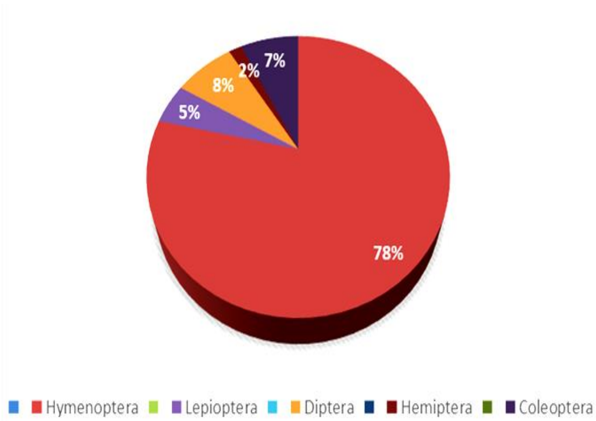


Fig. 2 Order wise Percentage of insect visitors

The availability of highly profitable mass flowering crops, such as oilseed rape, is directly correlated with the density of pollinator densities attracted by mass flowering (Westpal *et al.*, 2003). Insects play a crucial role in many processes, including pollination (Ockinger and Smith, 2007; Ollerton *et al.*, 2011), herbivory and detritivory (Yang and Gratton, 2014), and providing food for higher tropic levels of animals like amphibians, birds, and mammals. Their loss has detrimental effects on ecosystem functioning.

Because of modern agricultural methods, pollinators have insufficient access to the resources they need to survive, including food, nesting habitats, and other physical factors.

4. Conclusion

Numerous creatures contribute to the ecological services that remove our garbage and put food on our tables. It is obvious that the majority of insects are essential for pollinating a variety of crop plants. It is beyond our wildest dreams how important pollinators are to maintaining biodiversity and pollinating an enormous array of flowering plants. Even though these services are essential to maintaining human existence, it can be challenging to give them a proper economic value, which can make conservation efforts more important.

Plate 2 The agricultural fields where study was carried out.

There has to be a greater environmental knowledge today regarding practical habitat management that could contribute to an increase in insect pollinators. In order to preserve pollinators and to fully utilize the potential of crop pollination, exploratory study should also be conducted. Most notably, Niger (*Guizotia abyssinica*) plantations should be expanded since they have significant commercial and ecological importance as oil-producing plants that also play a significant role in maintaining insect biodiversity.

5. Acknowledgement

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6. Conflict of Interest

None declared. The authors affirm no financial or personal relationships that could influence the objectivity or interpretation of the findings.

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Research Article

Correlation and path coefficient analysis among yield and yield attributing traits of rice landraces

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ABSTRACT

Growing under a variety of ecological conditions, rice holds a significant position in Indian agriculture. However, yield mostly depends on net impact of yield component traits. Hence, the present investigation was carried out to study the correlation and path analysis in forty rice (*Oryza sativa* L.) landraces. Trait association of the yield and its attributing traits revealed that significant positive correlation of single plant yield with plant height, days to 50 percent flowering, flag leaf length and hundred seed weight. Thus, selecting for these traits might increase yield. Path coefficient analysis research revealed that the following characters had a favourable direct impact on the single plant yield: kernel breadth, length to breadth ratio, days to fifty percent flowering, number of productive tillers, and hundred seed weight. These characteristics might be used as selection indicators to increase rice yield in future breeding programmes.

KEY WORDS: Rice landraces; Yield; Correlation; Direct and Indirect effects

1. Introduction

Rice (*Oryza sativa* L.) is rich in genetic diversity, with thousands of varieties grown throughout the world. In Asia, rice fulfils 50% of the dietary calorie needs of almost 520 million people (Zhou *et al.*, 2016) and provides, good supply of vitamins and minerals like thiamine, niacin, iron, riboflavin, vitamin D, calcium and fibre (Schenker, 2012). While the demand for rice is continuously increasing with the rapid growth in the human population, the land area available for rice production is decreasing as a result of rapid urbanisation, impeding climate change and changing lifestyles. By 2025, there will be a 758 million tonnes of rice needed to meet the world's consumption, which is currently approximately 600 million tonnes (Nakano *et al.*, 2019). To satisfy future consumer demands, there is an urgent need for new rice cultivars with high yield

Grain yield is a complexly inherited characteristic that is polygenically regulated (Immanuel *et al.*, 2011). It is directly or indirectly impacted by a multitude of component traits and the environment factors. As a result, selection favouring one component may favourably influence linked qualities at the same time. Grain size, grain number and panicle number are reported to play major role and having high correlation with yield

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in rice (Kim *et al*., 2016). Direct selection for yield is not considered effective due to their low heritability. This is mainly due to heterozygosity, environmental influences and G × E (genotype × environment) interactions. To avoid these problems, indirect selection for yield is relatively better by selecting the traits which shows the high positive correlation with high heritability as compared to direct selection.

Correlation studies are useful in identifying the association among grain yield and its component traits (Lakshmi *et al*., 2019), enabling plant breeders to select genotypes possessing desirable traits that are associated to grain yield. Due to various environmental influences on polygenic traits, significant selection based on yield alone may not be effective. In order to improve yield, the selection must be achieved through component traits (Rangare *et al*., 2012; Debsharma *et al*., 2023). With this goal in mind, the current study was carried out to look for any effects on correlations between plant properties with yield potential.

2. Materials and Methods

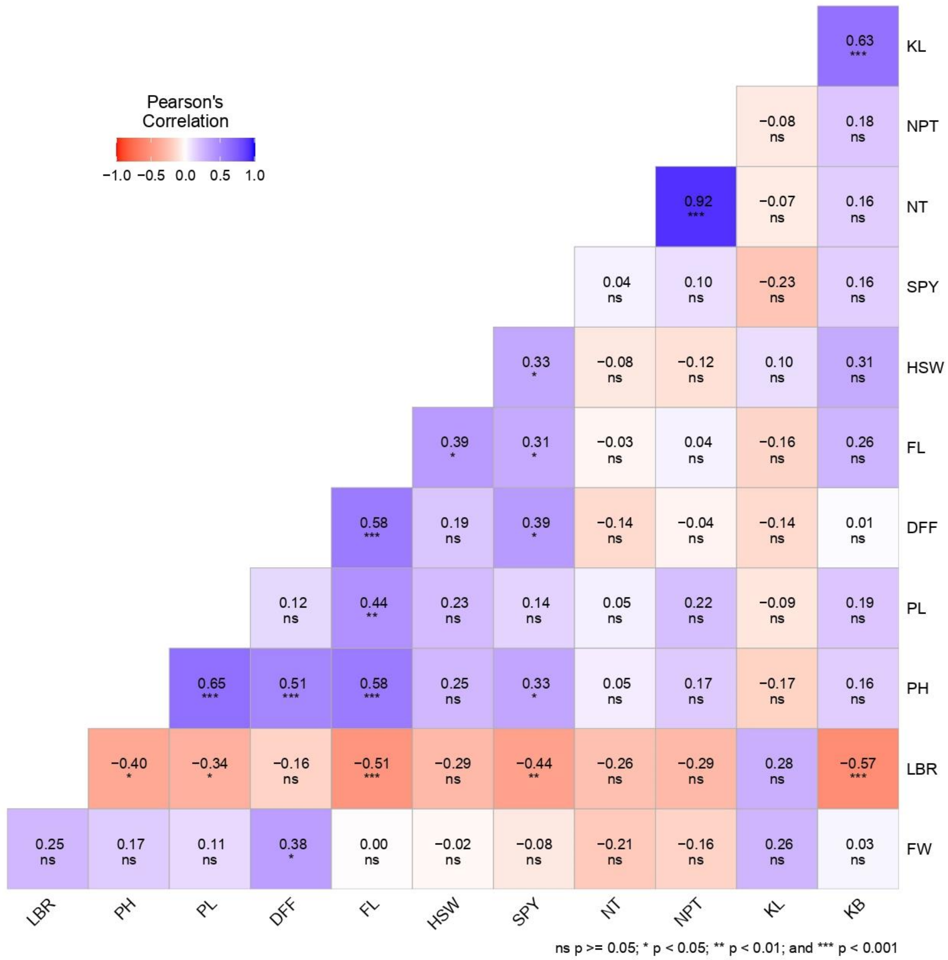
In Rabi, 2022, a set of forty landraces were cultivated at the Tamil Nadu Rice Research Institute (TRRI), Aduthurai, Thanjavur, Tamil Nadu, India. TRRI is located at latitude 10.99ºN and longitude 79.48ºE. Forty landraces were transplanted 21 days after sowing as two seedlings per hill with three replications in randomized block design with spacing of 20 × 20cm. The recommended package of practices for rice was followed for the proper establishment and growth of the crop. Observations were recorded on five random plants in each genotype in each replication for morphological characters *viz*., days to fifty percent flowering, plant height, number of tillers, number of productive tillers, flag leaf length, flag leaf width, panicle length, kernel length, kernel breadth, length to breadth ratio, hundred seed weight and single plant yield.

Analysis of variance for twelve morphological traits were analyzed using R software using "agricolae" package (Mendiburu and Yaseen, 2020). Correlation between the traits were estimated using "corrplot" package (Wei *et al*., 2017). Path analysis were estimated using "Semplot" package (Epskamp *et al*., 2017)

3. Results and Discussion

3.1 Correlation analysis

In the present study, significantly positive association was found between plant height ($r = 0.33$, $p < 0.05$), days to 50 percent flowering ($r = 0.39$, $P < 0.05$), flag leaf length ($r = 0.31$, $p < 0.05$) and hundred seed weight ($r = 0.33$, $p < 0.001$) with single plant yield (Fig. 1). Selection for these traits would be helpful in enhancing grain yield as this indicates a high correlation between these traits with single plant yield (Khan *et al*., 2020; Surjaye *et al*., 2022; Shanmugam *et al*., 2023; Thuy *et al*., 2023). Similar findings were reported by Saha *et al*., 2019; Ata-Ul-Karim *et al*., 2022; Faysal *et al*., 2022). Negative correlation was found between length breadth ratio ($r = -0.44$, $p < 0.01$) with single plant yield. In agreement to this finding, Ata-Ul-Karim *et al*. (2022) reported negative association of LBR with yield per plant. Hence, indirect selection for these traits would improve single plant yield. Since rice consumers have strong preferences for grain types, grain shape is given top consideration in all rice breeding programmes. It is important for breeders to carefully analyse the negative association between traits like grain width, length to breadth ratio and



DFF-Days to 50% flowering; FL-Flag leaf length; FW-Flag leaf width; HSW-Hundred seed weight; KB-Kernel breadth; KL-Kernel length; LBR- Length to breadth ratio; NPT-Number of productive tillers; NT-Number of tillers; PH-Plant height; PL-Panicle length; SPY-Single plant yield

Fig. 1 Correlation analysis of twelve morphological traits.

yield, as goals may limit or compromise efforts to improve yield (Ata-Ul-Karim *et al.*, 2022).

Understanding the interrelationships between yield component traits can help breeders determine the direction and strength of selection pressure to apply to related traits in order to simultaneously improve these traits (Gopikannan and Ganesh, 2013). Days to fifty percent flowering was found to exhibit strong and significant ($0.75 \leq r < 1.00$) correlation with plant height ($r = 0.51$, $p < 0.001$) and flag leaf length ($r = 0.58$, $p < 0.001$) while flag leaf width had a significant positive association ($r = 0.38$, $p < 0.05$). Strong and significant association was noted between number of tillers and number of productive tillers. Panicle length had strong and positive correlation with plant height ($r = 0.65$, p<

0.001) and had highly significant positive correlation with flag leaf length (r = 0.44, p< 0.01) while had negative and significant association with length breadth ratio (r = -0.34, p< 0.05). Flag leaf length detected significant (p < 0.05) and positive association with hundred seed weight (r = 0.33) and plant height (r = 0.58, p< 0.001). Length to breadth had a negative association with kernel breadth (r = -0.57, p< 0.001), flag leaf length (r = -0.51, p< 0.001), panicle length (r = -0.34, p< 0.05) and plant height (r = -0.40, p< 0.05). Strong and significant association was noted between kernel length and kernel breadth (r = 0.63, p< 0.001). Similar findings were reported by Khan *et al*., 2016; Priyanka *et al*., 2018; Prasannakumari *et al*., 2020; Shanmugam *et al*., 2023. It could be concluded from the above discussion that four traits *viz*., panicle length, days to 50 percent flowering, plant height, flag leaf length were directly associated with single plant yield and while length breadth ratio had an indirect association with yield. This implied that traits panicle length, days to 50 percent flowering, plant height, flag leaf length might be important for determining yield. These characteristics all showed a positive, substantial correlation with the single plant yield, suggesting that they are the main factors that contribute to yield and could be relied upon.

3.2 Path analysis

Correlation measures simply the link between two variables while path coefficient analysis, on the other hand, divides the correlations to clearly describe the cause-and-effect relationship and employs additional traits to explain the direct and indirect reasons of association (Wright, 1921). The direct and indirect effects of yield component traits studied as partitioned by path analysis were given in Table 1, Fig. 2.

The study reveals that kernel breadth and length to breadth ratio had high and positive direct effect on single plant yield. Days to fifty percent flowering,

Table 1: Direct and indirect effects of different yield components as partitioned by path analysis

Traits	DFF	PH	NT	NPT	FL	FW	PL	KL	KB	LBR	HSW	SPY
DFF	**0.401**	0.055	0.039	-0.011	-0.139	-0.059	-0.007	0.173	0.012	-0.127	0.052	0.389*
PH	0.206	**0.106**	-0.015	0.047	-0.139	-0.026	-0.038	0.214	0.219	-0.310	0.069	0.334*
NT	-0.056	0.006	**-0.284**	0.254	0.007	0.033	-0.003	0.087	0.217	-0.203	-0.021	0.036
NPT	-0.016	0.018	-0.261	**0.276**	-0.009	0.025	-0.013	0.096	0.246	-0.230	-0.032	0.100
FL	0.233	0.061	0.009	0.011	**-0.240**	0.001	-0.026	0.198	0.358	-0.400	0.109	0.313*
FW	0.152	0.018	0.060	-0.044	0.001	**-0.157**	-0.007	-0.329	0.041	0.196	-0.007	-0.077
PL	0.047	0.069	-0.013	0.060	-0.106	-0.018	**-0.059**	0.111	0.251	-0.267	0.064	0.139
KL	-0.055	-0.018	0.020	-0.021	0.038	-0.041	0.005	**-1.258**	0.847	0.222	0.028	-0.235
KB	0.003	0.017	-0.045	0.050	-0.063	-0.005	-0.011	-0.787	**1.355**	-0.443	0.086	0.157
LBR	-0.065	-0.042	0.074	-0.081	0.123	-0.039	0.020	-0.357	-0.767	**0.782**	-0.082	-0.435**
HSW	0.076	0.027	0.022	-0.032	-0.094	0.004	-0.014	-0.127	0.418	-0.230	**0.277**	0.327*

DFF-Days to 50% flowering; FL-Flag leaf length; FW-Flag leaf width; HSW-Hundred seed weight; KB-Kernel breadth; KL-Kernel length; LBR- Length to breadth ratio; NPT-Number of productive tillers; NT-Number of tillers; PH-Plant height; PL-Panicle length; SPY-Single plant yield

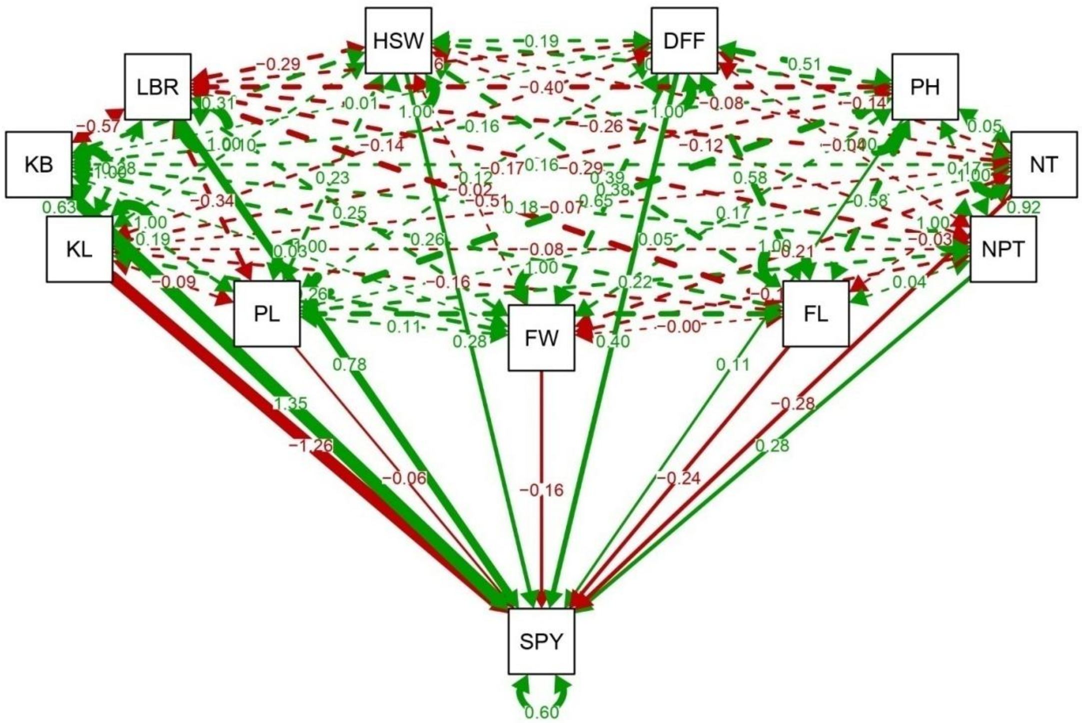


Fig. 2 Phenotypic path diagram

number of productive tillers and hundred seed weight expressed positively moderate direct effects on single plant yield. These results indicates yield improvement is directly associated with these traits. Similar findings were reported by Bhargava *et al*., 2021. The trait kernel length showed negatively high direct effect while flag leaf length and number of tillers had negatively moderate effect on single plant yield.

Kernel length, flag leaf length and hundred seed weight expressed high and positive indirect effects on single plant yield through kernel breadth. Plant height, number of tillers and number of productive tillers and panicle length exhibited moderately positive indirect effects on single plant through kernel breadth and exhibited moderately negative indirect effects through length breadth ratio. The traits *viz*., plant height and flag leaf length showed moderate and positive indirect effects via, days to 50 percent flowering. It could be inferred that kernel breadth, plant height, number of tillers and length to breadth ratio contributed equally through direct and indirect effects for yield improvement. These characteristics are thought to be significant yield factors, and each of them enhances grain yield through the interaction of other yield component traits. Since the studied characteristics contributed more to the variability in single plant yield, the residual effect of the study (0.20) is modest.

Under changing climatic conditions, direct yield enhancement is challenging. It is therefore possible to increase yield by identifying secondary traits that contribute to yield and making selective breeding decisions for those traits. Therefore, selection based on the subsequent secondary traits plant height, number of tillers, days to 50 percent flowering, kernel breadth, panicle length, length to breadth ratio may be utilised as trustworthy criteria for enhancing rice yield.

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5. Conflict of Interest

None declared. The authors affirm no financial or personal relationships that could influence the objectivity or interpretation of the findings.

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Current Innovation in Agriculture Science
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Research Article

Investigating the impact of biofertilizer (*Azotobacter* and Mycorrhiza) on nitrogen losses and yield in Wheat (*Triticum aestivum* L.) fields

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ABSTRACT

Nitrogen stands paramount as a critical nutrient indispensable for wheat growth and productivity. However, nitrogen losses within agroecosystems precipitate diminished soil fertility, decreased crop yield, and environmental perturbations. Ammonia volatilization and denitrification represent primary mechanisms contributing to nitrogen losses in agroecosystems. Consequently, there arises an imperative to explore diverse microbial consortium strategies aimed at enhancing nutrient provision while concurrently mitigating nitrogen losses and aligning with the crop's nutrient demands. In this study, four distinct treatments were implemented: Control (unfertilized, Tc), *Azotobacter* (Ta), Mycorrhiza (Tm), and *Azotobacter* + Mycorrhiza (Tam). Notably, the Tam treatment exhibited the highest levels of available soil nutrients such as phosphorus (P) and potassium (K), alongside fostering robust root proliferation, thereby augmenting soil nutrient uptake. Cumulative ammonia flux emissions ranged from 5.84 to 6.25 kg ha^{-1} in plots receiving microbial consortium treatments, with denitrification losses varying between 3.6 and 3.9 kg ha^{-1}, notably lower compared to Tc-treated plots. Yield assessments revealed a noteworthy yield of 5.34 t ha^{-1} in the Tam-treated plots, surpassing the yield of Tc-treated plots at 2.73 t ha^{-1}. Consequently, the utilization of microbial consortium practices emerges as an efficacious strategy for curbing nitrogen losses while concurrently enhancing crop yield.

KEY WORDS: Ammonium volatization loss; *Azotobacter*; Mycorrhiza; Denitirification loss; Root traits

1. Introduction

Wheat (*Triticum aestivum* L.) stands as the most extensively cultivated crop worldwide, owing to its remarkable adaptability to diverse environmental conditions and its pivotal role as a staple in the human diet. Of the global wheat production, approximately 65% is allocated for human consumption, with the remainder serving as livestock feed and supporting various facets of the food industry. Notably, wheat serves as the primary source of calories for a significant portion of the global population (Braun *et al*., 2019). Despite its paramount importance, the rate of yield gain in wheat cultivation has exhibited a continual decline, with yields stagnating in numerous major wheat-producing regions (Fischer and Edmeades, 2020). Climate change coupled with extensive chemical utilization in soil management exacerbates this trend, introducing variability to

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wheat cultivation conditions and posing challenges to sustainable production intensification. Securing and sustaining future wheat production hinges upon two primary objectives: sustainable intensification of wheat systems to accommodate escalating food demands and comprehending the ramifications of future climate scenarios on wheat yields (Asseng *et al*., 2011). Nitrogen (N) emerges as a vital nutrient elemental to plant growth and development. However, anthropogenic activities have led to a dramatic surge in the volume of reactive nitrogen (Nr) present in both the biosphere and atmosphere, precipitating a cascade of environmental repercussions including eutrophication, acidification, water pollution, and air pollution (Fowler *et al*., 2013). Moreover, N losses via various pathways such as runoff, ammonia volatilization, denitrification, and leaching contribute to groundwater pollution, water eutrophication, greenhouse gas emissions, and ozone (O_3) degradation in the stratosphere (Shi *et al*., 2012). Ammonia volatilization, characterized by the release of nitrogen in the form of free ammonia (NH_3) gas, stands as a significant pathway for nitrogen losses from agroecosystems. Denitrification, prevalent in agricultural systems, facilitates the conversion of nitrate and nitrite into nitric oxide (NO), nitrous oxide (N_2O), and nitrogen gas (N_2), with N_2 being released under anaerobic conditions (Bouwman *et al*., 2013). Terrestrial denitrification processes recycle 30%–60% of Nr back into N_2 (Ciais *et al*., 2013), with rice paddy fields or wetlands being identified as major sites of N loss via denitrification (Wang *et al*., 2017).While nitrogen fertilization stands as a pivotal measure for ensuring high crop yields, its indiscriminate use often leads to problematic agricultural practices. Raiver *et al.* (2017) observed increases in wheat yield, protein content, and nitrogen use efficiency in response to initial nitrogen deficiencies, potentially attributed to altered root growth dynamics during the seedling stage under low-N conditions. Hence, diversifying nitrogen fertilization strategies beyond reliance on inorganic fertilizers becomes imperative to enhance nitrogen use efficiency. Root elongation and seedling growth in wheat are sluggish during early growth stages, resulting in weak nitrogen absorption capacities owing to immature roots, thereby increasing nitrogen retention or losses and subsequently reducing nitrogen use efficiency (Jiang *et al*., 2017). Hu *et al*. (2018) underscored the significance of root morphology and function in nitrogen uptake, with root length, surface area, and branching patterns being primary determinants. Efficient nutrient absorption is facilitated by the upper root system (Tian *et al*., 2018). Despite achieving self-reliance in agricultural production, the extensive use of chemical fertilizers exacts tolls on soil nutrient statuses and biotic communities. In light of the adverse effects associated with chemical fertilizers, the utilization of microbial consortia as alternative nutrient sources emerges as a promising avenue to meet the escalating demands of crop production sustainably while addressing growing concerns regarding environmental pollution. Microbial consortia, comprising living microorganisms applied to soil, plants, or incorporated with seeds prior to sowing, promote plant growth by enhancing nutrient availability (Dakshayini *et al*., 2020).

2. Materials and Methods

The field experiment was conducted at the research farm of the Indian Agricultural Research Institute (IARI), New Delhi, during the winter season of 2018-19. Wheat cultivar "HD 3086" was sown in the third week of November on sandy loam soil with moderate levels of organic carbon (0.45%), nitrogen (347.7 kg/ha), phosphorus (21 kg/ha), and potassium (68.4 kg/ha), accompanied by a soil pH of 8.2 and electrical conductivity of 0.42 dS/m. Meteorological data recorded from the IARI observatory indicated mean minimum and maximum temperatures of 9.18°C and 25.57°C, respectively, throughout the trial period from November to April. The climate of New Delhi is characterized as continental, subtropical, and semi-arid, with an annual average precipitation of 650 mm, primarily attributed to the southwest monsoon.

The experimental design employed a randomized block design with three replications, utilizing wheat variety HD 3086 and four treatments during the winter season: Tc (No Fertilizer), Ta (*Azotobacter*), Tm (Mycorrhiza), and Tam (*Azotobacter* + Mycorrhiza). Biofertilizer treatments comprised Mycorrhiza at a rate of 10 kg/ha and *Azotobacter* seed treatment at 20 kg/ha.

Soil samples were collected from the 0-30 cm soil layer at three distinct locations within each treatment during tillering, flowering, grain-filling, and physiological maturity stages of the crop. Standard procedures were employed to analyze various soil properties, including determination of total nitrogen content using the Kjeldahl method (Kjeldahl, 1883), ammonical nitrogen using continuous flow analyzer, nitrate content using continuous flow analyzer, available soil phosphorus using Olsen's method (Olsen, 1954), available soil potassium using the Ammonium acetate method, and soil organic carbon using the Walkley and Black method (Walkley and Black, 1934). Plant nitrogen content was estimated via the Kjeldahl method. Ammonia volatilization was assessed using the force air graft method, employing closed chambers (20 cm × 20 cm × 50 cm) constructed from 6 mm acrylic sheets placed in the field. Volatilized ammonia gas was collected in 2% boric acid solution containing a mixed indicator (methyl red and bromocresol green), with air inside the chambers collected into boric acid traps using a vacuum pump operating at a flow rate of 3 L/min. Boric acid traps were replaced every 24 hours. Denitrification potential of soils was determined using the acetylene inhibition method. Harvesting occurred in the fourth week of April 2019, with the recording of yield and yield-contributing characteristics such as number of effective tillers, number of grains per spike, grain yield, and straw yield. Statistical analysis was conducted on the collected data, with means compared at a significance level of 5%.

3. Results and Discussion

3.1 Impact of microbial consortium on soil nutrient status

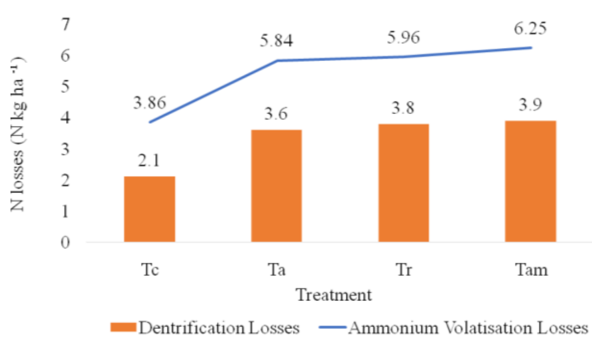
The study revealed significant variations in total nitrogen content among the treatments, ranging from 0.02% to 0.03%. Treatment Tam exhibited the highest total nitrogen content in soil at 0.036%, likely attributable to the application of *Azotobacter* and Mycorrhiza, resulting in a slight enhancement of total nitrogen content (Table 1). These findings align with those of Karad *et al*. (2016), who investigated the effect of biofertilizers on nitrogen dynamics in a groundnut-wheat system. Available phosphorus content ranged significantly from 20.4 kg/ha to 26.98 kg/ha, with the highest content observed in treatment Tam. Similarly, available potassium content ranged from 48.9 kg/ha to 66.56 kg/ha, with treatment Tam recording the highest potassium content in soil. Thus, the results indicate that the application of the mycorrhiza biofertilizer in conjunction with *Azotobacter* enhances soil fertility, thereby contributing to sustainable wheat crop yields. Rama *et al*. (2015) reported similar findings in soil nutrient studies conducted under various microbial treatments in corn (*Zea mays* L.). Notably, regular addition of biofertilizers in combination resulted in a significant improvement in soil organic carbon content, ranging from 0.47% to 0.50%, with treatment Tam exhibiting the highest organic carbon content at 0.50%. This increase in organic carbon content may be attributed to enhanced soil microbial diversity associated with wheat crops. Kaur *et al*. (2018) also observed similar results in their study on soil organic matter dynamics influenced by long-term organic and inorganic fertilizer use in a maize-wheat cropping system.

Table 1. Effect of microbial consortium on soil nutrient status

Treatments	Total Nitrogen (%)	Phosphorus (kg/ha)	Potassium (kg/ha)	Organic Carbon (%)
Tc	0.028	20.42	48.96	0.47
Ta	0.031	24.52	60.48	0.49
Tm	0.032	25.13	61.60	0.49
Tam	0.036	26.98	66.56	0.50
CD	0.02	1.38	4.89	0.02
sEM±	0.008	0.70	1.61	0.008

3.2 Impact of microbial consortium on Nitrogen losses

The investigation revealed significant reductions in ammonium volatilization and denitrification losses in plots receiving *Azotobacter* and Mycorrhiza. The lowest losses were observed in treatment Tc at 3.8 N kg/ha, while treatment Tam recorded the highest loss at 6.2 N kg/ha. Treatments Ta and Tm also exhibited decreased ammonium volatilization losses, with treatment Tam statistically comparable to treatments Tr and Ta. Similarly, for denitrification losses, treatment Tam recorded the highest loss at 3.9 N kg/ha, whereas the lowest losses were observed in treatment Tc at 1.05 N kg/ha. Treatments Ta and Tm demonstrated a two-fold decrease in denitrification losses, with treatments Ta and Tm statistically comparable in denitrification losses (Fig. 1). This reduction in denitrification losses under integrated fertilizer treatment compared to inorganic fertilizer treatment aligns closely with the findings of Aulakh (2020) on the use of biofertilizers for sustainable crop production, improved crop quality, soil health, and reduced environmental pollution.



Note: Ammonium volatisation and Denitrification losses (kg Nha^{-1}). Bar represent denitrification losses whereas line represent ammonia volatisation losses. Tc- (No Fertiliser), Ta-*Azotobacter*, Tm-Mycorrhiza, Tam-*Azotobacter* + Mycorrhiza.

Fig. 1 Effect of microbial consortium on Nitrogen losses

3.3 Impact of microbial consortium on Wheat crop yield attributes

Azotobacter and Mycorrhiza significantly influenced wheat yield attributes, including grain yield, straw yield, test weight, number of grains per ear, and number of spikelets per ear (Table 2). Treatment Tam exhibited the highest grain yield at

4.3 t/ha, significantly higher compared to treatments Tr and Ta, with a percentage increase of 4.7% compared to Tr and Ta. Treatment Tam also demonstrated the highest test weight at 37.9 g, representing a 5.9% increase compared to treatment Tc. Treatment Tam also recorded the highest number of grains per ear and number of spikelets per ear at 40.2 and 13.8, respectively. The harvesting index was highest in treatment Tam at 37.1%, significantly higher compared to treatments Tr and Tc, with treatments Ta and Tr exhibiting statistically comparable values. Significant differences in yield were observed in treatments receiving biofertilizer compared to the unfertilized control, consistent with the findings of Soleimanzadeh and Gooshchi (2018) on the effects of *Azotobacter* and nitrogen chemical fertilizer on wheat yield and yield components. The increase in grain and straw yield may be attributed to the sufficient quantities and balanced fractions of plant nutrients delivered to the crop as needed during the growth period, resulting in enhanced yield attributes. The integrated use of fertilizers with biofertilizers promoted dry matter accumulation, increased the number of effective tillers, grains per spike, and test weight, likely due to stimulated vegetative growth facilitated by the sustained and adequate supply of essential nutrients, as reported by Devi *et al*. (2019) in their study on the effect of microbial consortium on wheat growth and yield.

4. Conclusion

The findings of this study demonstrate that the application of biofertilizers enhances the pool of available nutrients in the soil, thereby increasing their accessibility for plant uptake. Moreover, it improves nitrogen use efficiency, rendering the soil less prone to nitrogen losses, whether through volatilization or denitrification. This was evidenced by the reduction in rates of ammonia volatilization and denitrification losses with the utilization of *Azotobacter* and Mycorrhiza treatments in wheat cultivation. Additionally, the study highlights the positive impact of *Azotobacter* and Mycorrhiza on wheat yield attributes, indicating the agronomic feasibility, economic viability, and environmental sustainability of employing these microbial treatments in crop production systems. By augmenting soil fertility and mitigating nitrogen losses, the integration of both nutrient sources optimizes fertilizer inputs, enhances crop nutrient use efficiency, and ultimately reduces nutrient losses, thereby contributing to sustainable crop production practices.

Table 2: Effect of microbial consortium on yield attributes of wheat crop

Treatment	Wheat Yield (t ha^{-1})	Straw yield (t ha^{-1})	Biological yield (t ha^{-1})	Test weight (g)	No. of grains per ear	No. of spikelet per ear	Harvesting index (%)
Tc	2.7	5.2	8	36.8	38.1	11.9	34
Ta	3.7	6.4	10.1	37.6	39.8	12.6	36.4
Tc	3.4	6.1	9.6	36.9	38.6	12.7	36.1
Tam	4.3	7.2	11.5	37.9	40.2	13.8	37.1
sEM±	0.11	0.19	0.34	0.6	0.7	0.26	0.98
CD	0.35	0.57	1.04	1.85	2.12	0.79	2.98

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Research Article

Unraveling the complexities of casual labour: An analysis of income disparities, market dynamics, and employment challenges in Udaipur, India

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ABSTRACT

The present study analysed the various aspects related to casual labour households, focusing on income levels, market functioning, employment status, wage rates, and constraints faced by households in rural and urban areas of Udaipur district of Rajasthan, India. The study observed that the average income varies between rural and urban areas, with urban households earning more on average due to higher employment opportunities. Skilled labour earns higher wages and works longer hours, often receiving advance payments. Payment methods are predominantly time-based in rural areas and a mix of time-based and piece-rate in urban areas. Discrimination in wages, undercutting of wages, and lack of bargaining power are perceived issues, with many households feeling they have little freedom to reject offered wages. Wage rates vary across quarters and between male and female labourers, with urban areas generally offering higher wages due to more non-agricultural work opportunities. Constraints faced by casual labour households include low wage rates, lack of skills, poor functioning of government employment schemes, seasonal employment nature, prolonged work hours, and drudgery of work. These constraints are more pronounced in rural areas, leading to challenges in securing stable employment and adequate wages.

KEY WORDS: Casual Labour; Rural and Urban divide; Casual Labour Market

1. Introduction

The pattern of economic growth in any country has implications for the structural transformation of its labour force. Globally, majority of the total workforce (3.3 billion) is living in moderate or extremely poor conditions out of which about 61 per cent are employed in the informal sector (International Labour Organization, 2019). In informal/unorganized sector, workers are not able to organize themselves for their common goal due to various constraints like casual nature of employment, illiteracy, ignorance, and scattered and small size of establishments (Government of India, 1969). Casual labour is mainly unskilled or semi-skilled workers whose employment pattern includes a course of short-term jobs. Their investment and consumption pattern cognates into same category as they earn their livelihood by selling their man power and often regenerate it by 'investing' a significant part of their wage-earnings on food items (Mishra and Lyngskor,

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2007). They work either in their own or nearby villages or to nearby cities for employment opportunities.

In India, nearly 95 per cent of the informal/ casual labour does not have any written job contracts, and the chances are also significantly low in getting a daily employment in the market (PLFS, 2018-19). At national level, about 56 per cent and 41 per cent of the below poverty line cards were owned by casual labour working in the agriculture sector and in non-agriculture sector, respectively (National Sample Survey, 68th round, 2011). This distribution shows that casual labour households are living in a low dietary consumption levels in India. Due to the low consumption, casual workers are often poor performers and their efficiency is also very low. The market forces often times impose on them the vicious cycle of inefficiency - low wage rates - low consumption - inefficiency. That's why majority of the casual labour is living in severe food-insecure conditions (Chakravarty and Dand, 2006). The average monthly income of farm households (for which casual labour is the primary occupation) is ₹ 8,931 (NABARD Report, 2016), which is not enough to satisfy their basic livelihood needs and a quality life. This poor sector spends a higher share of their expenditure on food and other essential requirements and oftentimes obtains their food from subsistence production, market, and transfers from government programs or from other relative households (Baiphethi and Jacobs, 2009). This low level of food consumption is a sincere livelihood problem as it increases risk of physical, social and mental issues (Nord, 1999).

In Indian context, urban casual labour is divided mainly into two types: (i) skilled labour or *karigar*, which includes skilled labour in the field of marble or tile fixing, wall plastering, wall preparation, wall painting, etc. and (ii) unskilled workers engaged in activities such as loading and unloading of trucks, preparation of building materials, etc. In case of rural casual labour, skilled labour is related to work such as agriculture, construction and painting of houses and unskilled workers are engaged in agricultural work, MGNREGA (public work programme), and other village work activities. This urban and rural divide of casual labour in emerging countries like India is characterized by comparatively higher income in urban areas on account of higher employment opportunities and wage rates.

Further, despite the fact that scholars have begun to push for the study of casual labour market, there has been little policy implementation at the international level and even less at the local level. Therefore, the current research is an exclusive attempt to study the casual labour cross the rural and urban divide based on primary data concerning various important economic parameters. The implications of the study will be useful in specific programme and policy recommendations that address the insecurity of casual labour and can act as a link to the debate of rural bias particularly among the informal or unorganised sector of casual labour.

2. Materials and Methods

2.1 Data

The study is based on primary data collected at two intervals i.e. August-September, 2019 and January-February, 2020 from the Udaipur district of Rajasthan (largest state of India). Udaipur district was purposively selected as it has the highest number of agricultural labour *i.e.* 3,02,968 (6.13 % of total agricultural labour in Rajasthan) and is fifth largest in terms of total worker population (13,65,783) in the state (Government of India, 2011). The district is a home to various indigenous communities including *Bhil*, *Meena*, *Damor* and *Gharasia* since approximately 50 percent of the tribal population of Rajasthan is located in this district (District Census Handbook, 2011). Poverty and malnutrition are especially prevalent amongst these indigenous communities and their occupation is dominated by agriculture with small landholding, undulated land and irrigated area. These communities have a higher incidence of illiteracy, poverty and malnourishment, and they face geographic and social isolation. The district lacks irrigation facilities, productive land, skill-building and other employment opportunities in the villages. Therefore, a large number of tribal and other rural people migrate to the city to earn their livelihood with makes it suitable to investigate the rural and urban divide in terms of consumption pattern and food security.

2.2 Sampling Design

The casual labour was divided into two categories *viz.,* category I[1] and category II[2] casual labour households. A comprehensive list of all casual labour market points was prepared in the Udaipur city of Rajasthan and four points *i.e.* Hathipole Circle, Pratapnagar Circle, MallaTalai and Reti Stand were selected based on the maximum number of casual labour availability. Thereafter, 40 labourers were selected through a judgmental or purposive sampling method from each point, respectively and were considered under the category I of casual labour. In order to make a valid comparison, an equal number of casual labours in category II based on similar economic conditions were purposively selected from villages or nearby places. Therefore, a total sample size of 320 casual labour households was considered for the present study collected at two intervals from 2019-2020.

2.3 Analytical tools and methods

Data collected were tabulated and analyzed to fulfil the specific objectives of the study. The tools, which were used for the analysis of the data, are presented and discussed below. After the collection of raw data, with the help of the schedules, these were compiled and tabulated as required for analysis.

2.4 Status of the casual labour market

The different features of the labour market such as money wage rate, kind wage, number of days worked, frequency of wage payment, working hours, and employment pattern in Udaipur district of Rajasthan were computed by applying simple statistical tools such as frequency, percentage and sample mean.

2.5 Constraints faced by casual labour

To identify and prioritize the constraints faced by casual labour in the study area, households were asked to rank. These ranks were analyzed through Garrett's ranking technique. Garrett's ranking technique gives the change of orders of constraints into numerical scores. The significant advantage of this technique as compared to simple frequency distribution is that in this technique constraints are

[1]Category I: Urban casual labour households who were engaged in different farm or non-farm activities in Udaipur city for at least 100 days in a year.

[2]Category II: Rural casual labour households who were engaged in different farm or non-farm activities within or nearby the village for at least 100 days in a year.

arranged based on their importance from households.

The Garrett's formula for converting ranks into per cent was given by the following expression:

$$\text{Percent position} = \frac{(R_{ij} - 0.5)}{N_j} \times 100$$

Where,
R_{ij} = rank given for i^{th} constraints by j^{th} individual and
N_j = number of constraints ranked by the j^{th} individual.

The relative position of each rank obtained from the above formula was converted into scores by referring to the critical values given in Table by Garrett and Woodworth (1969) (transmutation of orders of merit into units of amount or scores) for each factor; scores of all individuals were added and then divided by the total number of households for the specific factor (constraint).

3. Results and Discussion

3.1 Income level of casual labour households

The main source of income to the sample casual labour households was the wages of human labour. The income level of casual labour in both categories is depicted in Table 1. The total average income earned by the sample casual labour households was ₹ 10265, which varied from ₹ 8100 in rural areas to ₹ 11931 in urban areas, respectively. Across the class intervals, most of the respondents (35.93%) earned income between ₹ 6001 to 9000 per month followed by between ₹ 3001 to 6000 (22.81%), between ₹ 9001 to 12000 (20.65%), between more than ₹ 15000 (9.06%), between ₹ 12001 to ₹ 5000 (8.43%) and less than ₹ 3000 (3.12%). The number of people earning less than ₹ 3000 per month varied from 5 percent in rural category to 1.25 percent in urban category. In urban category, the majority of casual labour households earned monthly income between ₹ 6001 and ₹ 9000, whereas it was reported to be lower in the case of rural category (₹ 3001 to ₹ 6000). Only 2.50 percent of the sample rural casual labour households were earning a monthly average income of more than ₹ 15000, whereas it was higher in the urban category (15.62%). The urban category was receiving 33.59 percent more than the monthly national average income of ₹ 8,931 (NABARD Report, 2016). Whereas the rural casual labour households were reportedly receiving 9.30 percent lower than the national average. It was because of higher employment opportunities in the urban areas with direct implications on the sample households' income levels and food security.

Table 1: Income-wise distribution of sample casual labour households

(Number)

Sl. No.	Class intervals of monthly income (₹)	Category I (Urban)	Category II (Rural)	Overall
1	Less than 3000	2 (1.25)	8 (5.00)	10 (3.12)
2	3001-6000	15 (9.38)	58 (36.25)	73 (22.81)
3	6001-9000	60 (37.50)	55 (34.37)	115 (35.93)
4	9001-12000	41 (25.63)	25 (15.63)	66 (20.65)
5	12001-15000	17 (10.62)	10 (6.25)	27 (8.43)
6	More than 15000	25 (15.62)	4 (2.50)	29 (9.06)
	Total	160 (100.00)	160 (100.00)	320 (100.00)
	Average Income (₹)	11931	8100	10265

3.2 Status and functioning of casual labour market

The key features of the functioning of the casual labour market in the study area are shown in Table

2. The table showed that the urban casual labour market was divided mainly into two types: I skilled labour or *karigar*, which includes skilled labour in the field of marble or tile fixing, wall plastering, wall preparation, wall painting, etc.; and (ii) unskilled or helpful workers engaged in activities such as loading and unloading of trucks, preparation of building materials, etc. In urban casual labour households, the number of days worked per month for both types of work was found to be 25 days per month. The daily wage rate in the casual labour market in urban casual labour households was found to be ₹ 385 for unskilled labour, and ₹ 690 for skilled casual labour. Reddy and Kumar (2006) also reported higher wage rates for skilled labour on the market. Women in the urban casual labour market are mainly engaged in unskilled types of activities, such as construction and housework. Similar findings were also reported by Dave (2012).

In the urban casual labour market, working hours in the peak season were 9.55 hours per day for unskilled labour, and 10.02 for skilled labour, and in the slack season the corresponding figures were 8 hours per day for unskilled labour, and 8.56 hours per day for skilled labour. Skilled labour normally employed for a longer period, so that their hours of work were found to be greater than unskilled labour in urban casual labour households. Asiwal *et al.* (2018) also reported that in the casual labour market, the average number of hours worked by labour exceeded 8 hours (standard norms).

There was much less provision of advance wages in the urban casual labour market. Employers were found not to be giving advance wages in the case of unskilled labour, because unskilled labour usually works for a very shorter period, and there was also a chance or fear of them not coming to work after receiving advance wages. The skilled urban casual labour households mainly work for a longer period in the construction sector or any other activity, so sometimes employers or contractors find that they provide advance wages for casual labour, which is only 7 percent. Two factors could be attributed to the advance payment of wages for skilled casual labour. The first factor was that both employers and workers belonged to the same villages and did not suffer from moral hazards. Second, the payment of a certain amount of wages in advance ensures that the employer secures the supply of labour at the required time and saves the cost of the transaction.

The basis for payment in the urban casual market is divided into time and piece rate. Payment of wages for casual labour households based on a piece rate, mainly or for works such as unloading of trucks, was also made for construction work based on a time rate. It was found that 80.05 percent of the payment of wages made in terms of time and 19.95 percent of the payment of wages was made based on the rate of pay for unskilled casual labour. The corresponding figures were 91.67 percent and 8.33 percent for skilled urban casual labour. The wages for urban casual labour were revised back in the study area two years ago.

The table also showed that, in the rural casual labour market, labour was also divided into two types, i.e. skilled labour, which includes skilled labour related to agricultural work, construction and painting of houses, etc., and unskilled workers engaged in agricultural work, MNREGA, and other village work activities. Most of the labourers have worked in their own villages as well as in nearby villages for most of the time. The average number of casual working days per month, ranging from 17 days in the case of skilled casual labour, to 15 days in the case of unskilled casual labour in rural casual labour market. The daily wage rate in rural casual labour was found to be equal to ₹ 275 for unskilled labour, and ₹ 575 for skilled casual labour. In some cases, it has also been observed that there is a small amount of money borrowed by casual labourers who have repaid their wages by deduction. Women in the rural casual labour market are mainly engaged in agriculture, construction, and household-related work. A majority of casual labour has been found to work in public works in rural areas.

Average working hours for rural casual labour, during peak season, exceed 8 hours of standard labour, which were 9.75 hours per day for unskilled labour, 9.89 hours for skilled labour, and in the slack season, approximately 7 hours per day for unskilled labour, and 7.45 hours per day for skilled labour. Employers or contractors found that they were providing advance wages to casual labour, which was 5 percent in the case of skilled rural casual labour, and 3 percent in the case of unskilled rural casual labour. The majority of casual labours did not receive any snacks/meals from the employer, yet some of them received special snacks/meals in the case of agricultural labour.

On average, 92.75 percent of unskilled rural casual labour received wages on a time-based basis and 8.25 percent on a piece-based basis, compared with 87.65 percent and 12.35 percent for skilled rural casual labour, respectively, on the rural market. The wages of rural casual labour households, such as urban casual labour households, have not been revised in the study area for the last two years.

The results of the study clearly showed that there were more opportunities for urban casual labour, compared to rural casual labour. Wage rates were also found to be higher for both skilled and unskilled casual labour in the urban labour market. Casual work hours were also higher in urban casual labour in both peak and slack seasons. In the urban casual labour market, the piece rate was found to be higher compared to rural casual

Table 2: Functioning of casual labour market

Sl. No.	Particulars	Unit	Category I (Urban)		Category II (Rural)	
			Unskilled	Skilled	Unskilled	Skilled
1	Days worked	Days in a month	25	25	15	17
2	Average wage Rate	₹/day	385	690	275	575
3	Working hours					
(i)	Peak season	Hours/day	9.55	10.02	9.75	9.89
(ii)	Slack season	Hours/day	8.00	8.56	7.00	7.45
4	Advance wage					
(i)	Yes	Percent	0.00	7.00	3.00	5.00
(ii)	No	Percent	100.00	93.00	97.00	95.00
5	Basis of payment					
(i)	Time rate	Percent	80.05	91.67	92.75	87.65
(ii)	Piece rate	Percent	19.95	8.33	8.25	12.35
6	Wage revision	Years	2	2	2	2

labour, and the rate of time was found to be more prominent in rural casual labour, compared to urban casual labour. The last wage revision in both categories of casual labour was carried out in the study area two years ago. It can also be concluded that prevailing wage rate in the study area was found to be higher than the minimum wage fixed by the government of Rajasthan in both skilled and unskilled casual labours. Employment opportunities in urban areas were found to be far better than rural areas and that the wage rate was also higher, leading to a higher level of income for casual work in urban casual labour.

3.3 Salient features of the functioning of casual labour market

The opinion of the sample of casual labour households on some other important aspects of the functioning of the labour market is summarized in Table 3. In the case of casual labour in the study area first, 74.06 percent of labour households believed in loyalty to their employers. Second, 76.62 percent of labour households reported no conflict with their employers, and 24.38 percent reported that they had a conflict mainly over non-payment of wages on time. Third, the sample of labour households reported that they covered 21 km. Fourth, they were free to work elsewhere, and fifth, there was no labour union in labour households. Sixth, 80.74 percent of casual labour households believed that there was discrimination in wages in other places. Seventh, 56.92 percent of the sample of casual labour households faced undercutting wages and the remaining 43.08 percent did not face cuts in wages. Eighth, 53.12 percent of casual labour reported having the freedom to reject the wage offered, and 46.88 had no freedom to reject the wage offered. Ninth, 77.28 percent reported that there was no collusion between employers, and the remaining 22.72 percent reported that there was collusion between employers. Tenth, 72.78 percent of urban casual labour objected to the same wage paid to all, and the remaining 27.22 percent did not have any objection about the same wage paid to all.

First, 61.87 percent of urban casual labour households believed in loyalty to their employers. Second, 69.38 percent of labour households did not report any conflict with their employers; there were some cases of conflict mainly over non-payment of wages on time. Third, all the sample labour reported that they had travelled 37 km to work. Fourth, they were free to work elsewhere, and fifth, there was no labour union in casual labour. Sixthly, 70.23 percent of urban casual labour households believed that there was discrimination in wages in other places. Seventh, 51.49 percent of the sample urban casual labour households did not face undercutting wages and the remaining 48.51 percent faced lower wages. Eighth, 62.12 percent of urban casual labour households reported that they had the freedom to reject the wage offered and 37.88 percent were not free to reject the wage offered. Ninth, 80.44 percent reported that there was no collusion between employers, and the remaining 19.66 percent reported that there was collusion between employers. Tenth, 67.22 percent of urban casual labour households objected to the same wage paid to all, and the remaining 32.88 percent had no objection to the same wage paid to all.

First, in rural casual labour households, 86.25 percent of workers believed in loyalty to their employers. Second, 81.87 percent of labour households reported no conflict with their employers, and 18.13 percent reported some cases of conflict with their employers. Third, all the

Table 3: Salient features of the functioning of casual labour market

Sl. No.	Particulars	Unit	Category I (Urban)	Category II (Rural)	Overall
1	Loyalty towards employees				
(i)	Yes	Percent	61.87	86.25	74.06
(ii)	No	Percent	38.13	13.75	25.94
2	Conflicts between employer/contractor and labour				
(i)	Yes	Percent	30.62	18.13	24.38
(ii)	No	Percent	69.38	81.87	76.62
3	Distance have to covered for work	Km	37	4	21
4	Freedom to seek work in other places				
(i)	Yes	Percent	100.00	100.00	100.00
(ii)	No	Percent	0.00	0.00	0.00
5	Existing of labour union				
(i)	Yes	Percent	0.00	0.00	0.00
(ii)	No	Percent	100.00	100.00	100.00
6	Discrimination in wages in other places				
(i)	Yes	Percent	70.23	91.24	80.74
(ii)	No	Percent	29.77	8.76	19.26
7	Undercutting of wages				
(i)	Yes	Percent	48.51	65.33	56.92
(ii)	No	Percent	51.49	34.67	43.08
8	Freedom to reject wage offered				
(i)	Yes	Percent	62.12	44.11	53.12
(ii)	No	Percent	37.88	55.89	46.88
9	Collusion among employers				
(i)	Yes	Percent	19.56	25.88	22.72
(ii)	No	Percent	80.44	74.12	77.28
10	Objection about same wage paid to all				
(i)	Yes	Percent	67.22	78.34	72.78
(ii)	No	Percent	32.88	21.66	27.22

rural casual labour households reported going 4 km to work every day. Fourth, one-hundred percent of rural casual labour households were free to work elsewhere, and fifth, there was no labour union. Sixthly, 91.24 percent of rural labour households believed that there was discrimination in wages elsewhere. Seventh, 65.33 percent of the sample of rural labour households faced undercutting wages and the remaining 34.67 percent did not face cuts in wages. Eighth, 55.89 percent of urban casual labour households reported that they had no freedom to reject the wage offered, and 44.11 were free to reject the wage offered because of the less availability of work in rural areas. Ninth, 74.12 percent reported that there was no collusion between employers, and the remaining 25.88 percent reported that there was collusion between employers. Tenth, 78.34 percent of rural labour households objected to the same wage paid to all, and the remaining 21.66 percent had no objection to the same wage paid to all.

It can be summed up that the majority of casual labour households, who believed in loyalty to their employers, did not report any conflict with their employers, covered 21 km, were free to work elsewhere, did not have a labour union, believed in discrimination in wages elsewhere, faced undercutting wages, reported freedom to reject the wage offered, reported no collusion and had objection to the same wage paid to all.

3.4 Employment status of casual labour

The employment and unemployment status of casual labour is shown in Table 4, which shows that, in the urban category of casual labour households, average working days were 25 days per month with 4 days of unemployment and urban casual labour was not available on one day in a month due to various issues such as family or individual health, social reasons, etc. The probability of employment was 0.86 in the case of urban casual labour. This indicates that the chances of working for rural casual labour were 86 times and that they could not get work 14 times.

Average working days for rural casual labour households were found to be 16.00 days per month with 12.50 days of unemployment and 1.50 days of non-working days per month due to various issues such as family or individual health, social reasons, etc. The probability of employment for rural casual labour was 0.55. This indicates that the chances of working for rural casual labour households were 55 times higher.

Table 4: Employment status

(Days per labour)

Sl. No.	Particulars	Category I (Urban)	Category II (Rural)
1	Average working days	25.00	16.00
2	Unemployed days	4.0	12.50
3	Not available for work	1.0	1.50
4	Probability of Employment (E_P)	0.86	0.55
5	Probability of unemployment (UE_P)	0.14	0.45

Note: (i) E_P may be defined as the number of days for which person is employed, (ii) $UE_P = 1 - E_P$

Average working days in urban casual labour households were 25 days higher than rural casual labour households (16 days). The probability of employment was also found to be higher in the case of urban casual labour households (0.86) compared to rural casual labour households (0.55).

It can be summed up that urban casual labour households had better job opportunities than rural casual labour households, that's why a lot of casual labour travelled daily from their villages to Udaipur to earn wages.

3.5 Average daily wages of casual labour

Wages are, by and large, the only source of income for casual labour in the study area. As such, the only determinant of casual labour earnings is the rate of pay and the extent to which work is available in a month or year (Chitodkar, 1992). Table 5 and Table 6 show the average daily wage earnings of all the casual labour sampled during the study period. It is important to note that the average urban casual labour wage was found to be the highest in the quarter I (April-June), which was ₹ 323 for male labour and ₹ 291 for female labour with an average wage of ₹ 304 due to the summer season, and mainly construction activities were carried out at the prime level during that period. The lowest average wage earnings (₹ 287) were recorded during quarter II (July-Sep) consisting of a maximum of ₹ 296 per day for male labour and a maximum of ₹ 278 per day for female labour due to less availability of work during the rainy season. The average wage earnings for quarter III (Oct-Dec) and quarter IV (Jan-March) were ₹ 317, and ₹315 for male and ₹ 280, and ₹ 282 for female labour with an average wage of ₹ 298 and ₹ 299, respectively. Asthana and Medrano (2001) also reported the difference in wages for male and female labour.

Table 6 shows that, in rural casual labour, average daily wage earnings in non-public works ranged between ₹ 252 (quarter I) and ₹ 297 (quarter IV) for males, and similarly, ranged from ₹ 192 (quarter I) to ₹ 203 (quarter IV) for females during different periods. The highest wage earnings were recorded in quarter IV (approximately ₹ 250) and quarter III (approximately ₹ 243) due to higher employment in agricultural activities such as harvesting in winter and rainy season. Swamikannan and Jeyalakshmi (2015) also reported maximum employment in agriculture during the rainy and winter seasons. The MGNREGA Act enhances the security of households' livelihoods in rural areas of the country and provides for at least 100 days of employment in every household in the financial year for which adult members volunteer to do unskilled manual work. Table 6 shows average wage earnings in public works ranging from ₹ 134 (quarter II) to ₹ 165 (quarter IV) for males and ₹ 100 (quarter II) to ₹ 130 (quarter IV) for females. In public works such as MGNREGA, the lowest wage earnings (₹ 117) were recorded during the quarter II period due to the rainy season in the study area.

Table 5: Average daily wage earnings of urban casual labour

Sl. No.	Quarters	Gender wise wage (₹)		Quarterly Average wage (₹)
		Male	Female	
1	Quarter I (April-June)	323	291	304
2	Quarter II (July- Sep)	296	278	287
3	Quarter III (Oct- Dec)	317	280	298
4	Quarter IV(Jan March)	315	282	299
5	Annual average wage	313	283	297

The results suggest that these casual labour working partly in agriculture and in partly in non-agricultural activities in rural areas and casual labour working only in non-agricultural activities in urban areas. The average wage rate for urban casual labour, therefore, was higher than that for rural casual labour. All those labours who go for work outside the village are earning a much higher daily wage than those who worked in the village. It can be concluded that wage earnings ranged from one quarter to the next quarter, as well as between male and female labour. The differential wage rate is one of the important factors for labour migration in the study area.

Table 6: Average daily wage earnings of rural casual labour

Sl. No.	Particulars	Wages in public work (₹)		Quarterly Average wage (₹)	Wages in other than public work (₹)		Quarterly Average wage (₹)
		Male	Female		Male	Female	
1	Quarter I (April-June)	145	105	125	252	192	222
2	Quarter II (July- Sep)	134	100	117	256	193	224
3	Quarter III (Oct- Dec)	145	121	133	286	199	243
4	Quarter IV (Jan- March)	165	130	148	297	203	250
5	Annual average wage	147	114	131	273	197	235

3.6 Constraints faced by casual labour

In order to analyze the constraints faced by casual labour households, major problems identified during the visits were presented to the sample of casual labour households and asked to rank them according to the severity of the constraints and analyzed using the Garrett ranking technique. All the constraints faced by casual labour households discussed in the following sub-headings:

3.7 Constraints related to employment opportunities

Table 7 depicted the constraints related to employment opportunities and perceived that low wage rates and unavailability of work as a major problem with a Garrett score of 78.27, lack of skills (63.78), poor functioning of MGNREGA (63.47), seasonal nature of employment (51.33), prolonged work hours (48.57), drudgery of work (44.55), low bargaining power (39.04), no labour union (32.48) and misbehave with labour by employment provider (27.08) were the major constraints related to employment opportunities securing second, third, fourth fifth, sixth, seventh, eighth and ninth rank, respectively. There are a very large number of casual workers working in urban and rural areas, so there were not enough job opportunities for all casual workers, which also reduced their bargaining power. Dave (2012) also reported constraints such as lack of skills, long working hours, poor working conditions, occupational hazards, low wages for labour.

Urban casual labour perceived that low wage rates and unavailability of work ranked first with a Garrett score of 77.32, lack of skills (69.21), poor functioning of MGNREGA (56.21), seasonal nature of employment (53.20), prolonged work hours (50), drudgery of work (45.40), low bargaining power (39.32), no labour union (32.30) and misbehave with labour by employment provider (27.04) were the major constraints related to employment opportunities securing second, third, fourth fifth, sixth, seventh, eighth and ninth rank, respectively.

Rural casual labour households perceived that low wage rates and unavailability of work ranked first with a Garrett score of 79.22, poor functioning of

MGNREGA (70.74), lack of skills (58.34), seasonal nature of employment (52.32), prolonged work hours (47.14), the drudgery of work (43.70), low bargaining power (38.76), no labour union (32.66) and misbehave with labour by employment provider (27.12) were the major constraints related to employment opportunities securing second, third, fourth fifth, sixth, seventh, eighth and ninth rank, respectively.

It can be concluded that the main constraint faced by households was low wage rates and unavailability of work and, at the very least, misbehaviour of labour by the employment provider.

It can be summed up that casual labour households in the study area believed that large family size, lack of capital, lack of availability of hospitals/health centres and health services, lack of availability of schools/Anganwadi centres/colleges, lack of public contact of labour with information sources, lack of pucca houses, low wage rates and unavailability of work were the major constraints faced by the casual labour households.

4. Suggestions and Recommendations

The study noted that there is no single platform for casual labour, where they can unite to discuss various issues related to them. There is often a conflict between casual labour and employers on a variety of issues, such as undercutting wages, working hours, and many other disputes. The Government should, therefore, set up a common platform to address various issues related to casual labour. The study shows that a low wage rate; a lower number of working days and low family earnings prevail in rural areas. Existing rural activities must be renovated in light of the minimum needs of casual labour. Rural development opportunities need to be created through the establishment of agro-based industries, particularly in rural areas, so that rural labour can work at a decent wage rate for a reasonable number of days.

Table 7: Constraints related to employment of casual labour

Sl. No.	Particulars	Category I (Urban)		Category II (Rural)		Overall	
		Garrett Score	Rank	Garrett Score	Rank	Garrett Score	Rank
1	Low wage rates and unavailability of work	77.32	I	79.22	I	78.27	I
2	Lack of skills	69.21	II	58.34	III	63.78	II
3	Poor functioning of MGNREGA	56.21	III	70.74	II	63.47	III
4	Seasonal nature of employment	53.20	IV	52.32	IV	51.33	IV
5	Prolonged work hours	50.00	V	47.14	V	48.57	V
6	Drudgery of work	45.40	VI	43.70	VI	44.55	VI
7	Low bargaining power	39.32	VII	38.76	VII	39.04	VII
8	No labour union	32.30	VIII	32.66	VIII	32.48	VIII
9	Misbehave with labour by employment provider	27.04	IX	27.12	IX	27.08	IX

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6. Conflict of interest

The authors declared that they have no conflict of interest in this study.

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Research Article

Exploring the impact of biofertilizers on Tomato crop growth and yield: A comprehensive research study

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ABSTRACT

Experiment was conducted to investigate the effect of different combination of biofertilizers and inorganic chemical fertilizers on the growth and yield parameters of tomato crop. The experiment was designed using a Completely Randomized Design with three replicates and seven treatments, including the sole application of individual strains and their combinations with 75% of recommended dose of fertilizers. The investigation was prompted by the combined effects of inorganic fertilizers and the potential benefits of microbial inoculants. The results revealed that the 100% RDF (T_2) and combined inoculation of 75% RDF with *Azotobacter* + PSB + KSB (T_7) recorded significant increase in the growth and yield parameters of tomato over control (T_1). Further, treatment (T_2) and (T_7) found on par to each other. Tomato plants treated with T_2 exhibited significantly greater plant height (126.8 cm at harvest stage), Number of branches per plant (14.66), Number of leaves per plant (113.33), fresh weight (412.66 g plant^{-1}), and dry weight (133 g plant^{-1}). The study highlights the potential of combined inoculation of inorganic fertilizers and beneficial microbial inoculants promote the growth and yield parameters of tomato plants, this results also suggest that the 75% RDF in combination with Nitrogen fixing, phosphorous and potash solubilising bacteria may helps in reducing the 25% of inorganic chemical fertilizer application and which intern leads to reduce in input cost to the farmers.

KEY WORDS: Tomato; Biofertilizers; *Azotobacter*; Phosphorus solubilisers; Potassium solubilisers

1. Introduction

Tomato (*Solanum lycopersicum*) is a widely cultivated and economically important vegetable crop worldwide (Sadashiva *et al*., 2013; Venema *et al*., 2005). Its nutritional value, versatility in culinary applications, and growing demand make it a significant component of the global agricultural industry. In recent years, there has been a growing emphasis on sustainable agricultural practices that minimize the use of synthetic inputs (Pretty, 2008) while ensuring optimal crop productivity.

The tomato crop in India has not been able to reach its full potential in terms of yield. This can be attributed to various factors, one of which is the inadequate use of fertilizers. In particular, hybrid varieties of tomatoes require higher nutrient levels for optimal growth (Kamal *et al*., 2018). However, the low utilization of fertilizers and the imbalanced application ratio of nitrogen, phosphorus, and potassium (NPK) contribute to the subpar yield. Relying solely on chemical fertilizers is insufficient to sustain higher yields, and the high cost of inorganic fertilizers often leads to poor profit margins (Ritu and Dash, 2022). To address these challenges, the integration of biofertilizers alongside inorganic fertilizers emerges as a promising solution. Biofertilizers offer several advantages, including their eco-

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friendliness and economic viability, which can help reduce the dependence on chemical fertilizers. These biofertilizers have the ability to fix significant amounts of atmospheric nitrogen in the soil, enhance plant growth through the production of organic acids and growth hormones, and improve nutrient availability for the plants. By incorporating biofertilizers into tomato cultivation practices, farmers can optimize nutrient uptake, stimulate plant growth, and enhance overall crop productivity. In this context, the utilization of biofertilizers has gained attention as a potential alternative to conventional fertilizers for enhancing tomato crop growth, yield, and overall sustainability.

Furthermore, the use of biofertilizers has demonstrated positive effects on the soil ecosystem. The introduction of beneficial microorganisms can help improve soil structure, enhance nutrient cycling, and suppress soil-borne pathogens, thereby reducing the reliance on chemical inputs and promoting sustainable soil health management. However, despite the growing interest in biofertilizers for tomato cultivation, there is still a need for comprehensive research that investigates their efficacy under different environmental conditions, examines their long-term effects on crop growth and yield, and elucidates the underlying mechanisms involved. This research article aims to fill this gap by presenting a comprehensive study that evaluates the effects of various biofertilizers on tomato crop growth and yield performance. Through a combination of field experiments, laboratory analyses, and statistical modeling, this study seeks to provide valuable insights into the potential benefits and practical implications of integrating biofertilizers into tomato production systems.

2. Materials and Methods

The field experiment was conducted to evaluate the impact of different treatments on tomato crop growth and yield. The experiment consisted of seven treatments, which included 75 % and 100 % of the recommended dose of NPK. These treatments were combined with the application of N-fixing bacteria (*Azotobacter*), phosphorus-solubilizing bacteria (PSB) and Potassium solubilizing bacteria (KSB), in addition to a control that does not received any chemical and bio fertilizers.

The experimental design followed a randomized block design, with three replications to ensure statistical validity. Seedlings, aged thirty-eight days, were transplanted into the experimental plots and applications of biofertilizers were imposed as per the respective treatment combinations. All necessary cultural operations were performed in accordance with the specific requirements of the tomato crop across all experimental plots. Plant height measurements were taken at various intervals: 45, 90 days after transplantation (DAT) and at the final harvest.

At the time of the final harvest, the plants were uprooted and their fresh weight was measured. These plants were then subjected to sun drying for a period of 3-4 days, followed by drying in an oven at a temperature of 55 °C for 6-8 days. The dried plants were weighed to obtain the plant dry weight. Additionally, observations were made for the number of primary branches and the number of fruits per plant at the final harvest stage. The total fruit yield (measured in quintals per hectare) was computed based on the yield obtained from each individual plot.

By conducting these measurements and observations, valuable data was collected to assess various parameters of tomato plant growth and yield. This comprehensive analysis allowed for a detailed evaluation of the effects of the experimental treatments on the different plant characteristics, providing insights into the performance and productivity of the tomato crops.

3. Results and Discussion

3.1 Plant height (cm)

In this study, plant height was monitored at various stages of growth, and the effects of microbial inoculants and inorganic chemical fertilizer treatment combinations were recorded. The results showed a continuous increase in plant height from 45 days after transplanting (DAT) until the harvesting stage in all the treatment combination, regardless of the presence of microbial inoculants. However, treatments with 100% RDF (126.8 cm) and 75% RDF with *Azotobacter*, PSB and KSB (125.3 cm) showed significantly greater plant height compared to all other treatments at all stages of plant growth (45, 90 and at harvest stage) (Table 1, Fig. 1). Conversely, the control treatment (T_1) that did not received any chemical fertilizers and microbial inoculants exhibited the lowest plant height (37.5 cm).

These findings highlight the positive impact of specific microbial inoculants combinations in promoting plant height, providing valuable insights for optimizing plant growth in agricultural practices. Similar results were obtained by (Argaw, 2012) who revealed that the parameters like the height of the plant were enhanced by the co-inoculation of PSB and *Azotobacter* than single inoculation.

3.2 Number of branches/plant

The experiment examined the impact of different treatments on the number of branches per plant. The results revealed that Treatment T_2, which involved applying the full recommended dose of fertilizer (100 % RDF), led to the highest average of 14.66 branches per plant. This indicates that providing plants with the complete fertilizer dosage significantly promoted branch development (Table 2, Fig. 2).

Furthermore, Treatment T_7, which combined 75% RDF with *Azotobacter*, PSB, and KSB, demonstrated the second-highest average of 13.33 branches per plant. This suggests that the synergistic effects of microbial inoculants, along with a reduced fertilizer dose, positively influenced branch growth, similar findings were also recorded by (Gajbhiye *et al.*, 2003). In contrast, treatments involving specific microbial inoculants (T_3, T_4, and T_5) resulted in moderate to slightly lower numbers of branches per plant, ranging from 10.33 to 8.00. Finally, Treatment T_1 which is not treated with any biofertilizer and inorganic chemical fertilizer exhibited the lowest average of 4.00 branches per plant.

Table 1: Effect of biofertilizers on plant height (cm) at various stages of crop growth

Treatment	Plant Height (cm)		
	45 DAT	90 DAT	At final harvest
T_1 – Control	18.2	32.4	37.5
T_2 – 100% RDF	76.1	117.3	126.8
T_3 – 75% RDF + *Azotobacter*	70.4	112.2	121.7
T_4 – 75% RDF + PSB	69.5	109.5	120.4
T_5 – 75% RDF + KSB	69.0	108.1	119.6
T_6 – *Azotobacter* + PSB + KSB	55.9	91.7	116.2
T_7 – 75% RDF + *Azotobacter* + PSB + KSB	74.3	116.2	125.3
S.Em ±	0.82	1.21	1.12
CD at 5%	2.41	3.60	3.31

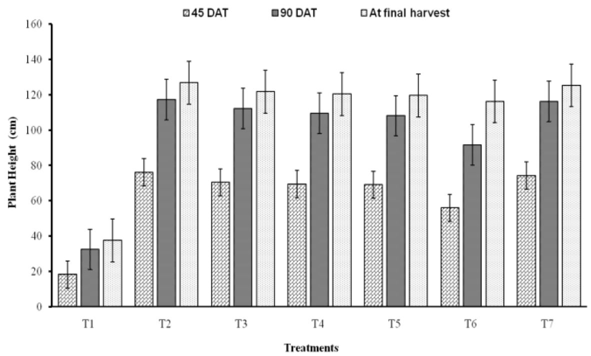

Fig. 1 Effect of biofertilizers on plant height (cm) at various stages of crop growth

3.3 No. of leaves/plant

The results of the experiment suggest significant differences in the number of leaves per plant among certain treatments. Treatment T_2, applying the full recommended dose of fertilizer (100% RDF), demonstrated a significantly higher leaf count with an average of 113.33 compared to the control group (Treatment T_1) with 51.00 leaves per plant. Similarly, Treatment T_7, combining 75% RDF with *Azotobacter*, PSB, and KSB, exhibited a significantly higher leaf count with an average of 109.66. However, treatments T_3, T_4, T_5, and T_6, which involved combinations of microbial inoculants and reduced RDF, showed non-significant differences in leaf count compared to T_2 and T_7. Nonetheless, these treatments still displayed improved leaf development compared to the control (Table 2, Fig. 2). Overall, the results indicate that the full RDF application and the combined use of microbial inoculants with a reduced RDF had significant positive effects on leaf growth, while the other treatments showed non-significant differences but still contributed to enhanced leaf development, in an another research conducted by (Mamatha and Bagyaraj, 2003) demonstrated the similar results.

3.4 Fresh weight/plant (g)

The results showed that, fresh weight per plant across different treatments, revealing both significant and non-significant differences. Treatment T_2, involving the application of the full recommended dose of fertilizer (100% RDF), displayed a significantly higher fresh weight per plant with an average of 412.66 g, compared to the control (T_1) with 195.33 g. Similarly, Treatment T_7, combining 75% RDF with *Azotobacter*, PSB, and KSB, exhibited a significantly higher fresh weight per plant with an average of 410.33 g, results obtained were similar to the research done by (Mahdi *et al.*, 2011) who investigated the effect of *Azotobacter chroococum*, was more than *Pseudomonas putida* and the combined inoculation produced the higher results than the control or sole application of either inoculants.. The findings demonstrate that the full RDF application and the combined use of microbial inoculants with a reduced RDF had significant positive effects on plant weight (Table 2, Fig. 2).

3.5 Dry weight/plant (g)

The analysis of dry weight per plant among the different treatments revealed that, Treatment T_2 which received the full recommended dose of fertilizer (100% RDF), exhibited a significantly higher dry weight per plant with an average of 133 g compared to the control group (T_1) with 42.66 g. Similarly, Treatment T_7, combining 75% RDF with *Azotobacter*, PSB, and KSB, displayed a significantly higher dry weight per plant with an average of 129.66 g, our results are in accordance with (Singh *et al.*, 2004). In contrast, treatments T_3, T_4, T_5, and T_6, which involved combinations of microbial inoculants with a reduced RDF, showed non-significant differences in dry weight

Table 2: Effect of biofertilizers on growth and yield parameters of tomato crop

Treatment	No. of branches/Plant	No. of leaves/Plant	Fresh weight/Plant (g)	Dry weight/Plant (g)	Number of fruits/Plant	Yield (q/ha)
T_1 – Control	4.00	51.00	195.33	42.66	14.66	238.71
T_2 – 100% RDF	14.66	113.33	412.66	133.00	53.00	712.46
T_3 – 75% RDF + *Azotobacter*	10.33	102.00	386.00	103.33	41.33	624.35
T_4 – 75% RDF + PSB	9.66	98.66	383.66	101.00	38.00	617.97
T_5 – 75% RDF + KSB	8.00	96.00	379.33	94.66	35.66	613.52
T_6 – *Azotobacter* + PSB + KSB	7.66	92.33	322.00	63.00	26.33	413.53
T_7 – 75% RDF + *Azotobacter* + PSB + KSB	13.33	109.66	410.33	129.66	51.66	697.34
S.Em ±	0.76	2.37	6.23	8.16	2.82	13.28
CD at 5%	2.25	7.08	18.64	24.45	8.41	39.81

compared to T_2 and T_7 (Table 2, Fig. 2). However, these treatments still contributed to increased dry weight relative to the control.

3.6 Number of fruits/plant

The analysis of the number of fruits per plant revealed significant differences among the treatments. Treatment T_2, which applied the full recommended dose of fertilizer (100% RDF), exhibited a significantly higher fruit count per plant compared to the control (T_1). Similarly, Treatment T_7, combining 75% RDF with *Azotobacter*, PSB, and KSB, demonstrated a significant increase in fruit production. In contrast, when compared to T_2 and T_7, treatments T_3, T_4, T_5, and T_6, which employed combinations of microbial inoculants with a reduced RDF, did not exhibit statistically significant disparities in fruit count. However, these treatments still contributed to an enhanced number of fruits per plant (Table 2, Fig. 2).

3.7 Fruit yield/plant

The analysis of the fruit yield per plant (q/ha) across various treatments showed that the Treatment T_2, which involved the application of the full recommended dose of fertilizer (100% RDF), displayed a significantly higher fruit yield per plant compared to the control (T_1) and which was followed by the treatment T_7 which received combined application 75% RDF with Azotobacter, PSB, and KSB. These results are in agreement with the findings of (Dhanasekaran and Bhuvaneswari, 2005). Notably, Treatment T_1 which does not received inorganic fertilizer and biofertilizers demonstrated the lowest fruit yield among the treatments (Table 2, Fig. 2).

From this study, it was concluded that the significant variations in various plant growth and yield parameters. Treatments involving the full recommended dose of fertilizer (100% RDF) consistently demonstrated superior performance across multiple metrics. Specifically, T_2 displayed increased plant height, a higher number of leaves, branches and fruits per plant, as well as greater fresh and dry weights, similar results were also obtained by (Sengupta *et al.,* 2002). Additionally, Treatment T_7, combining 75% RDF with microbial inoculants, showcased notable improvements in plant height, number of branches, fresh weight, and fruit yield (Rama and Naik, 2017).

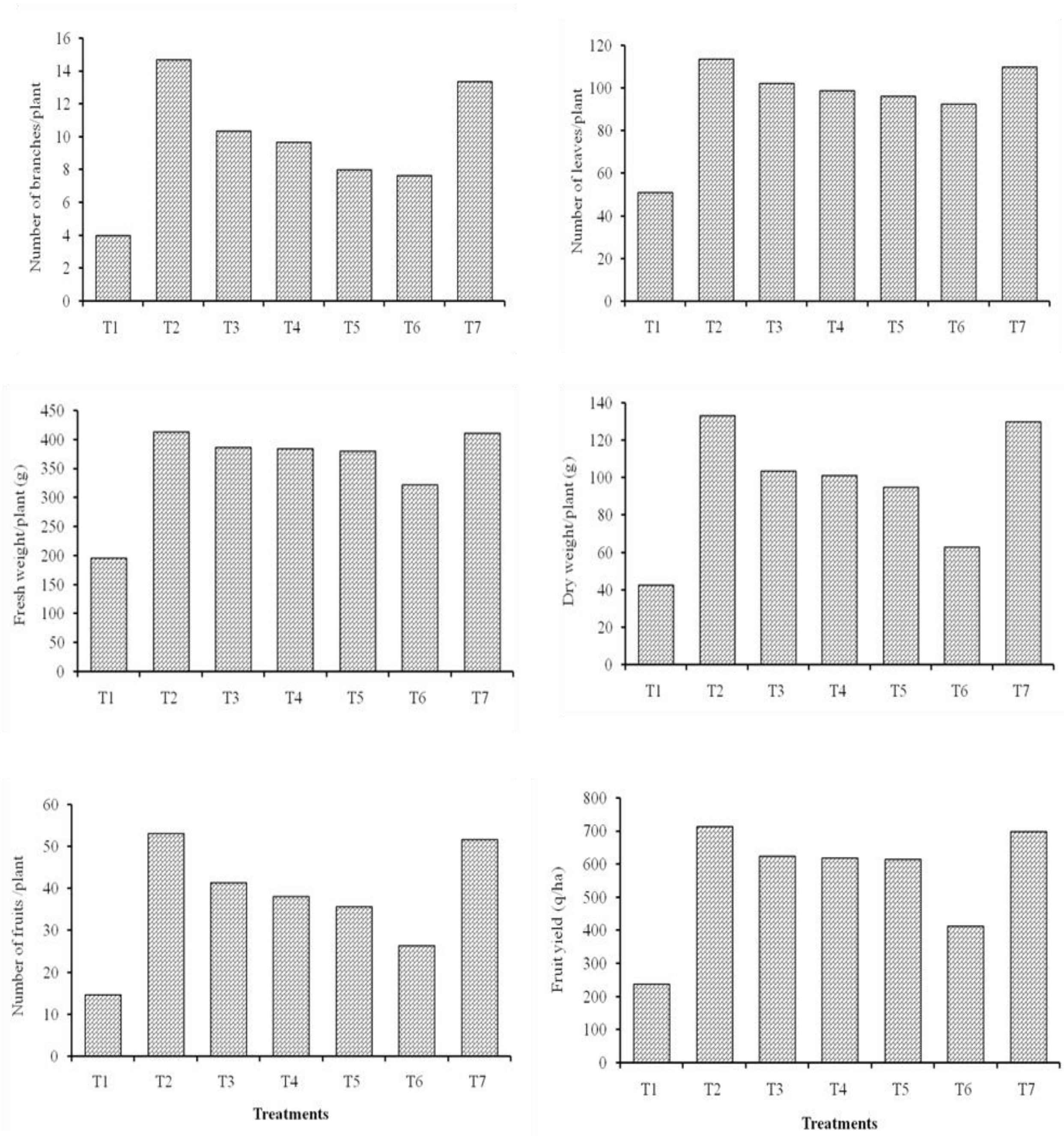

Fig.2 Effect of biofertilizers on growth and yield parameters of tomato crop

While treatments with reduced RDF and microbial inoculants (T_3, T_4 and T_5) did not yield statistically significant differences in various growth and yield parameters of tomato, they still exhibited positive effects compared to the control (T_1).

The observed enhancements in all the plant growth and yield parameters may be attributed to various factors. One potential contributor is the secretion of ammonia into the rhizosphere, which can have a positive impact on plant growth (Harikrishna *et al.*, 2002). Another factor that could play a role is the accelerated movement of photosynthates facilitated by root exudates.

The presence of beneficial microorganisms in the soil might also contribute to the observed improvements. These microorganisms may engage in various biological activities that enhance the soil's condition, transforming it into a fertile zone capable of readily supplying essential nutrients to the plant's root system. Similar positive outcomes were documented in studies conducted on coriander (Subramanian and Vijayakumar, 2001) and maize (Rama *et al.*, 2015). Collectively, these findings offer valuable insights for improving growth and yield parameters of tomato by adapting different treatment combinations to reduced the use of inorganic chemical fertilizers.

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5. Conflict of Interest

None declared. The authors affirm no financial or personal relationships that could influence the objectivity or interpretation of the findings.

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Research Article

Genetic analysis of Maize crop resistance to Polysora Rust: A comprehensive investigation

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ABSTRACT

This study investigates the inheritance pattern of *Puccinia polysora* rust resistance in maize, focusing on a segregating F2 population derived from a cross between the susceptible inbred line CM 202 and the resistant inbred line MAI 105. Disease severity was assessed using the Percent Leaf Area Covered (PLAC) method, revealing a spectrum of resistance within the population. Genetic analysis confirmed a single dominant gene controlling rust resistance, aligning with previous research indicating monogenic inheritance in maize. The identification of this dominant gene offers significant implications for breeding programs, enabling the development of rust-resistant varieties through marker-assisted selection and gene pyramiding. Additionally, the presence of moderate resistance and susceptibility suggests the involvement of modifiers or quantitative trait loci. Understanding these mechanisms is crucial for comprehensive rust resistance elucidation. Overall, this study provides valuable insights into the genetic basis of polysora rust resistance, informing breeding strategies to enhance maize productivity and resilience against rust diseases.

KEY WORDS: Maize; *Puccinia polysora*; Disease resistance; Inheritance pattern; Genetic analysis

1. Introduction

Maize (*Zea mays* L.) is a fundamental crop globally, serving as a staple food for millions of people and playing a crucial role in various industries. Its versatility extends beyond dietary consumption, encompassing applications in livestock feed, biofuels, and industrial production. This cereal crop's adaptability to diverse climates and soil conditions underscores its significance in ensuring food security and economic stability worldwide. However, maize production faces formidable challenges, with diseases posing a significant threat to yields and food security. Among these pathogens, Polysora rust, caused by the fungus *Puccinia polysora*, emerges as a prominent adversary. This biotrophic fungus primarily targets maize leaves, manifesting as rust-like lesions that compromise photosynthetic efficiency and weaken the plant's overall vigor, ultimately leading to yield losses.

Understanding the genetic basis of resistance to Polysora rust is paramount for developing effective management strategies to mitigate its impact. Previous research endeavors have made notable progress in unraveling the genetic mechanisms underlying resistance, shedding light on the intricate interplay between host genetics and pathogen virulence. Studies have highlighted the existence of genetic diversity among maize

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genotypes concerning Polysora rust resistance. Chungu *et al*. (2007) conducted research on tropical maize inbred lines, revealing substantial variation in resistance levels. This diversity provides a valuable resource for breeding programs aimed at enhancing resistance in maize cultivars, thereby bolstering resilience against Polysora rust.

Furthermore, investigations into the resistance of maize hybrids and inbred lines have yielded insights into the genetic architecture of resistance traits. Munkvold and Mengistu (1998) evaluated various maize genotypes for their resistance to northern leaf blight and Polysora rust, identifying promising candidates with durable resistance. These findings underscore the importance of genotype-specific responses to pathogen challenges and emphasize the need for tailored breeding strategies to enhance resistance in maize cultivars. Characterization studies have contributed to elucidating the mechanisms underlying resistance to Polysora rust in maize. Takan *et al.* (1998) conducted comprehensive analyses to delineate the genetic basis of resistance traits, identifying key genomic regions associated with resistance. By dissecting the molecular pathways involved in host-pathogen interactions, such studies provide a foundation for targeted breeding efforts aimed at developing maize cultivars with enhanced resistance to Polysora rust.

Moreover, advances in molecular genetics and genomic technologies have facilitated the identification of candidate genes and markers linked to Polysora rust resistance in maize. These molecular tools offer valuable resources for marker-assisted selection (MAS) programs, enabling breeders to expedite the development of rust-resistant maize cultivars with improved agronomic traits.

In summary, maize stands as a cornerstone crop in global agriculture, but its productivity is threatened by diseases such as Polysora rust. Understanding the genetic basis of resistance to this pathogen is crucial for developing sustainable management strategies and ensuring food security. Through genetic diversity studies, characterization of resistance mechanisms, and leveraging molecular tools, researchers are making significant strides towards enhancing maize resilience to Polysora rust. Continued interdisciplinary efforts will be essential for harnessing the full potential of genetic resistance and safeguarding maize production against emerging challenges in the agricultural landscape.

2. Materials and Methods

The materials utilized, experimental procedures employed, and data analysis methods are comprehensively detailed below.

2.1 Selection of plant material

The base material for experimentation comprised one polysora rust-resistant inbred, MAI 105, and one susceptible inbred, CM 202. These lines were maintained through successive generations by selfing.

2.2 Crossing programme

The two selected inbred lines were planted during the late rainy season. The crossing procedure involved uniform silk cutting the previous evening, followed by crossing and pollination with pollen the following day between 8-10 AM. Subsequently, F1 progeny were harvested. The F1 generation was grown during the early rainy season and self-pollinated within the same season, with subsequent harvest. In late rainy season, the F2 population (consisting of 150 plants) was cultivated in three plots, each measuring 5 meters in length and arranged in four rows with a spacing of 70cm × 20cm. Polysora rust-susceptible genotype 219 J was planted as a border row, and artificial inoculation was conducted as previously described.

2.3 Assessment of Disease Severity

Disease severity/incidence data were recorded on individual plants at two intervals: first at anthesis and then at the dough stage, using a standard scale ranging from 1 to 5 (Cramer, 1967). Subsequently, these scores were converted to percentages of disease severity (0-100%) following the method outlined by James (1971).

2.4 Statistical Analysis

The segregation pattern in the cross was statistically analyzed using the Chi-square test to assess the inheritance pattern of the resistance trait.

By adhering to standardized protocols for plant material selection, crossing procedures, disease assessment, and statistical analysis, this study aimed to elucidate the inheritance pattern of polysora rust resistance in maize and contribute to the genetic improvement of maize cultivars for enhanced disease resistance.

3. Results and Discussion

The F2 population, comprising 150 plants resulting from the cross between the susceptible inbred line CM 202 and the resistant inbred line MAI 105, was meticulously cultivated and assessed for polysora rust disease severity on an individual plant basis. Disease severity was quantified using the PLAC (Percent Leaf Area Covered) method, which assigns a severity score ranging from 0 to 100% to each plant based on the proportion of leaf area affected by the disease.

To elucidate the inheritance pattern of polysora rust resistance, a genetic ratio analysis was conducted. The observed segregation ratio of 3 (resistant):1 (susceptible) was evaluated using the Chi-square test. The calculated Chi-square value was found to be lower than the tabulated Chi-square value, leading to the acceptance of the null hypothesis (Table 1). This statistical analysis supports the conclusion that the trait of polysora rust resistance in this population is governed by the control of a single dominant gene.

Table 1: Analysis of Polysora rust severity on F2 population

Description	No. of Plants in F2 Population		Total
	Resistant	Susceptible	
Observed frequency (O)	118	32	150
Expected frequency (E)	113	37	150
$\chi^2 = \sum(O-E)2/E$	0.22	0.68	0.90 (NS)

NS – Non Significant

Table 2a and 2b presents the comprehensive data on disease severity within the F2 population. Among the 150 plants assessed, 53 demonstrated resistance to polysora rust, while 66 exhibited moderate resistance. Additionally, 27 plants displayed moderate susceptibility to the disease, while the remaining 5 plants were categorized as susceptible. This result underscores the genetic basis of polysora rust resistance in maize, providing valuable insights into the inheritance pattern of this trait.

Table 2a: Reaction of F2 population of maize line (CM-202×MAI-105) to Polysora rust

Sl. No.	Disease severity (%)	Disease reaction	Sl. No.	Disease severity (%)	Disease reaction	Sl. No.	Disease severity (%)	Disease reaction	Sl. No.	Disease severity (%)	Disease reaction
1	25	MR	26	45	MS	51	1	R	76	25	MR
2	1	R	27	45	MS	52	5	R	77	45	MS
3	5	R	28	25	MR	53	25	MR	78	5	R
4	1	R	29	15	MR	54	25	MR	79	45	MS
5	15	MR	30	15	MR	55	15	MR	80	5	R
6	5	R	31	15	MR	56	15	MR	81	1	R
7	15	MR	32	5	R	57	25	MR	82	5	R
8	25	MR	33	1	R	58	15	MR	83	15	MR
9	45	MS	34	1	R	59	1	R	84	15	MR
10	25	MR	35	1	R	60	45	MS	85	15	MR
11	5	R	36	1	R	61	1	R	86	45	MS
12	1	R	37	1	R	62	5	R	87	45	MS
13	1	R	38	15	MR	63	25	MR	88	25	MR
14	5	R	39	45	MS	64	15	MR	89	45	MS
15	25	MR	40	5	R	65	5	R	90	45	MS
16	25	MR	41	1	R	66	1	R	91	1	R
17	25	MR	42	1	R	67	1	R	92	5	R
18	15	MR	43	25	MR	68	45	MS	93	15	MR
19	5	R	44	5	R	69	15	MR	94	5	R
20	15	MR	45	15	MR	70	45	MS	95	25	MR
21	45	MS	46	5	R	71	45	MS	96	15	MR
22	5	R	47	25	MR	72	45	MS	97	25	MR
23	5	R	48	15	MR	73	45	MS	98	5	R
24	1	R	49	25	MR	74	25	MR	99	15	MR
25	1	R	50	1	R	75	45	MS	100	1	R

Note: R=Resistant MR=Moderately Resistant S=Susceptible HS=Highly Susceptible

Table 2b: Reaction of F2 population of maize line (CM-202×MAI-105) to Polysora rust

(Contd..)

Sl. No.	Disease severity(%)	Disease reaction	Sl. No.	Disease severity(%)	Disease reaction	Sl. No.	Disease severity (%)	Disease reaction
101	15	MR	121	15.00	MR	141	5	R
102	15	MR	122	5	R	142	15	MR
103	5	R	123	5	R	143	15	MR
104	15	MR	124	1	R	144	45	MS
105	25	MR	125	25	MR	145	65	S
106	25	MR	126	45	MS	146	65	S
107	15	MR	127	45	MS	147	25	MR
108	25	MR	128	25	MR	148	15	MR
109	15	MR	129	45	MS	149	25	MR
110	5	R	130	65	S	150	65	S
111	15	MR	131	45	MS		**CM202**	**MAI105**
112	1	R	132	45	MS		65	1
113	1	R	133	15	MR		45	1
114	25	MR	134	45	MS		65	1
115	5	R	135	25	MR		85	5
116	5	R	136	45	MS		4	10
117	25	MR	137	25	MR		45	1
118	15	MR	138	65	MS		45	1
119	25	MR	139	45	MR		45	5
120	15	MR	140	25	MR		25	1

Note: R = Resistant MR = Moderately Resistant S = Susceptible HS = Highly Susceptible

Such findings hold significant implications for maize breeding programs aimed at developing cultivars with enhanced resistance to polysora rust, thereby contributing to sustainable maize production and food security.

4. Conclusion

The findings of this study shed light on the inheritance pattern of polysora rust resistance in maize, providing valuable insights into the genetic mechanisms underlying this important trait. The observed segregation ratio of 3 (resistant):1 (susceptible) in the F2 population suggests that the resistance to polysora rust is governed by the control of a single dominant gene. This finding aligns with previous research demonstrating the monogenic inheritance of rust resistance in maize (Takan *et al*., 1998; Chungu *et al*., 2007). Furthermore, understanding the genetic basis of rust resistance enables breeders to employ advanced breeding strategies such as gene pyramiding, which involves stacking multiple resistance genes to enhance durability and broaden the spectrum of resistance (Singh *et al*., 2017).

The moderate resistance observed in a substantial proportion of the F2 population suggests the presence of modifiers or quantitative trait loci (QTLs) influencing the expression of rust resistance. These modifiers may interact with the major resistance gene, influencing the degree of resistance conferred by the gene and contributing to the observed phenotypic variation (Kuchel *et al*., 2007). Further research is warranted to elucidate the genetic basis of moderate resistance and identify potential QTLs associated with this trait.

It is noteworthy that a small percentage of plants in the F2 population exhibited susceptibility to polysora rust despite originating from a resistant × susceptible cross. This phenomenon could be attributed to factors such as incomplete penetrance, environmental influences, or genetic background effects (Michelmore *et al*., 1991). Understanding the mechanisms underlying susceptibility in these individuals is essential for comprehensive elucidation of rust resistance in maize and may uncover novel genetic factors contributing to disease susceptibility.

The utilization of the PLAC method for disease severity assessment enabled precise quantification of rust symptoms, providing valuable data for genetic analysis. However, it is essential to acknowledge potential limitations associated with this method, such as subjectivity in symptom evaluation and variability in disease expression under different environmental conditions (Jeger *et al*., 2018). Integrating additional phenotypic and molecular approaches, such as histological analysis and gene expression profiling, could provide deeper insights into the mechanisms of rust resistance and complement the findings of this study.

In conclusion, the identification of a single dominant gene controlling polysora rust resistance in maize represents a significant advancement in our understanding of rust resistance genetics. This knowledge lays the foundation for targeted breeding efforts aimed at developing rust-resistant maize varieties with enhanced productivity and resilience. Continued research into the genetic basis of rust resistance, coupled with advancements in breeding technologies, holds promise for addressing the challenge of rust disease in maize and ensuring global food security.

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CRISPR-Cas revolution in Agriculture: From precision genome editing to sustainable crop improvement

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ABSTRACT

The CRISPR-Cas system has emerged as a transformative tool in agricultural biotechnology, revolutionizing the landscape of crop improvement. This review paper explores the multifaceted applications of CRISPR technology in agriculture, from its fundamental principles to its practical implementations for precision genome editing. We delve into the various strategies employed to enhance crop growth and yield traits, including disease resistance, abiotic stress tolerance, and nutritional content, thereby contributing to the development of sustainable agriculture practices. Furthermore, we discuss the regulatory frameworks and ethical considerations surrounding the deployment of CRISPR-edited crops, highlighting the challenges and opportunities for its widespread adoption. Through a comprehensive analysis of recent advancements and future prospects, this review aims to provide insights into the role of CRISPR-Cas in shaping the future of agriculture and global food security.

KEY WORDS: CRISPR; Cas9; Agriculture; Gene editing

1. Introduction

The application of CRISPR-Cas9 technology in agriculture has catalyzed a paradigm shift in crop improvement strategies, offering unparalleled precision and efficiency in genome editing. CRISPR (Clustered Regularly Interspaced Short Palindromic Repeats) and its associated protein Cas9 constitute a groundbreaking molecular toolkit that enables targeted modifications of DNA sequences within plant genomes (Jinek *et al.*, 2012). This revolutionary genome editing platform has not only expedited the pace of genetic manipulation in crops but also unlocked novel avenues for sustainable agriculture and global food security (Hsu *et al.*, 2014).

The versatility and simplicity of the CRISPR-Cas9 system have propelled research efforts worldwide, driving significant advancements in crop biotechnology. By precisely targeting genes linked to agriculturally significant traits, such as yield, stress tolerance, and nutritional composition, researchers have achieved remarkable success in developing crops with enhanced productivity and resilience (Puchta and Fauser, 2014). For instance, CRISPR-mediated editing has been instrumental in conferring resistance to devastating pathogens like powdery mildew in wheat (Wang *et al.*, 2014), bacterial blight in rice (Li *et al.*, 2012), and citrus canker in citrus plants (Jia and Wang, 2014).

Moreover, CRISPR technology holds promise for

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promoting sustainable agricultural practices by mitigating the environmental impacts associated with conventional farming methods. Through targeted genome editing, researchers can engineer crops that require fewer chemical inputs, such as pesticides and fertilizers, thereby reducing environmental pollution and preserving soil health (Schindele *et al*., 2018). Additionally, CRISPR-edited crops with improved nutritional profiles offer a viable solution to malnutrition and food insecurity, particularly in regions where staple crops lack essential vitamins and minerals (Wurtzel *et al*., 2019).

Despite the transformative potential of CRISPR-Cas9 in agriculture, several challenges persist, ranging from regulatory complexities to ethical considerations and public acceptance (Waltz, 2016). The regulatory landscape governing the cultivation and commercialization of genetically modified organisms (GMOs) varies across jurisdictions, presenting obstacles to the global adoption of CRISPR-edited crops (Chawla *et al*., 2017). Moreover, concerns regarding unintended off-target effects and the potential for gene flow to wild relatives underscore the need for rigorous risk assessment and environmental monitoring (Bortesi and Fischer, 2015).

In this comprehensive review, we aim to provide an extensive exploration of the CRISPR-Cas revolution in agriculture, spanning its diverse applications, underlying mechanisms, regulatory frameworks, and ethical implications. By synthesizing recent research findings and emerging trends, we seek to elucidate the transformative potential of CRISPR technology in driving sustainable crop improvement and addressing the multifaceted challenges confronting modern agriculture.

2. CRISPR-Cas: components and mechanism

2.1 Components of CRISPR-Cas system

At its core, the CRISPR-Cas system consists of two primary components: the Cas protein and the guide RNA (gRNA). The Cas protein, typically Cas9, serves as the molecular scissors responsible for cleaving DNA at specific target sequences. The gRNA, composed of a CRISPR RNA (crRNA) and a trans-activating CRISPR RNA (tracrRNA) fused together, guides the Cas protein to the target DNA sequence through complementary base pairing (Jinek *et al*., 2012).

2.2 Mechanism of CRISPR-Cas action

The mechanism of CRISPR-Cas-mediated genome editing involves several key steps. Initially, the gRNA forms a complex with the Cas protein, leading to the formation of the Cas-gRNA ribonucleoprotein (RNP) complex. This complex scans the genomic DNA for sequences complementary to the gRNA, facilitating target recognition (Doudna and Charpentier, 2014).

Upon binding to the target DNA sequence, the Cas protein undergoes a conformational change, resulting in the activation of its endonuclease activity. The endonuclease domains of the Cas protein then catalyze the cleavage of the DNA, generating double-strand breaks (DSBs) at the target site (Gasiunas *et al*., 2012).

Following DNA cleavage, the cell's DNA repair machinery comes into play to resolve the DSBs. Two primary pathways involved in DNA repair are non-homologous end joining (NHEJ) and homology-directed repair (HDR). NHEJ often leads to small insertions or deletions (indels) at the site of the DSB, resulting in gene knockout or disruption. In contrast, HDR utilizes a template DNA molecule to precisely repair the DSB, enabling gene editing and insertion of desired sequences (Doudna and Charpentier, 2014).

3. CRISPR-Cas9: evolution to precision genome editing

The journey of the CRISPR-Cas system began with the elucidation of clustered regularly interspaced short palindromic repeats (CRISPR) in the genomes of bacteria and archaea. Initial studies in the late 1980s and 1990s identified these repetitive DNA sequences, which sparked curiosity about their function. However, it was not until the early 2000s that researchers began to unravel the significance of CRISPR in bacterial immunity against viral infections.

The versatility of CRISPR-Cas9 lies in its ability to target virtually any genomic locus by simply modifying the sequence of the gRNA. This programmable nature, coupled with its high efficiency and specificity, has revolutionized genome editing and facilitated the study of gene function and regulation in various organisms.

4. Importance of CRISPR-Cas in Agriculture

The escalating impacts of climate change have intensified the specter of food scarcity, necessitating innovative approaches to bolster agricultural productivity. Climatic shifts, characterized by erratic weather patterns, extreme temperatures, and unpredictable precipitation, have destabilized traditional farming practices, leading to reduced crop yields and compromised food security. In response to these challenges,

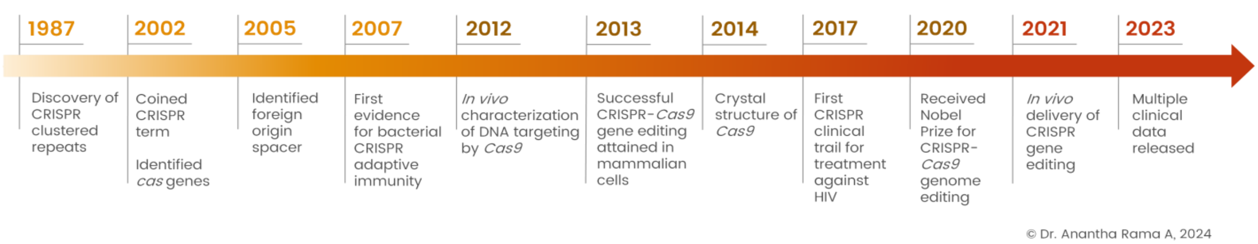

Fig. 1: Evolution of the CRISPR-Cas System: Key Milestones

The breakthrough came in 2012 when Doudna and Charpentier demonstrated the programmable nature of CRISPR-Cas9 for targeted genome editing in bacteria. Their landmark paper published in Science described how the Cas9 protein, guided by a short RNA molecule, could precisely cleave specific DNA sequences. This pivotal discovery laid the foundation for a myriad of applications in genetic engineering, ranging from gene knockout and knock-in to gene regulation and functional genomics.

targeted genome editing, facilitated by advanced molecular tools such as CRISPR-Cas systems, has emerged as a promising approach to boost agricultural productivity. In rice and wheat, targeted genome editing has been instrumental in increasing grain size, weight, and number, as well as enhancing protein content, tiller spread, and tiller number. These improvements have been reported in various studies (Wang *et al*., 2020; Oliva *et al*., 2019; Zhang *et al*., 2019).

Moreover, targeted genome editing has led to significant enhancements in the quality of crops such as rice and corn. Modified crops utilizing the CRISPR–Cas system have been tailored to reduce the levels of toxic steroidal glycoalkaloids, thereby enhancing the color and extending the shelf-life of fruits and vegetables, rendering them more commercially appealing. Additionally, these modifications have resulted in an increase in desirable traits such as amylose and starch content, as well as good fats like oleic acid levels. Furthermore, improvements in fragrance, gluten protein reduction, and decreased unsaturated fatty acids content have been achieved (Li *et al*., 2012; Cermak *et al*., 2015; Clasen *et al*., 2016; Jia *et al*., 2017). These advancements highlight the transformative potential of CRISPR-based targeted genome editing in agriculture, offering precise and tailored solutions to address the complex challenges posed by climate change and food insecurity.

4.1 Enhancing crop yield and quality

CRISPR-Cas9 genome editing, targeting the OsNAS2 promoter, specifically deleting the cis-regulatory element ARR1AT at position -933, significantly increased Zn concentration per plant in rice and also led to an augmented spikelet number per main panicle, resulting in increased grain yield per plant (Ludwig *et al.*, 2024). In an another study conducted by Usman *et al*. (2020) reported that precise editing of the OsPYL9 gene by RNA-guided Cas9 nuclease increased the grain yield in rice by regulating circadian rhythm. CRISPR/Cas9-mediated multiplex genome editing targeted three key genes - GW2, GW5, and TGW6, known as negative regulators of grain weight and the outcomes demonstrated a notable increase in grain size and thousand grain weight (Xu *et al*., 2016).

Bioactive compounds, characterized as additional nutritional constituents found in small quantities in foods, often contribute to the prevention of cardiovascular disease and cancer. Anthocyanin, malate, γ-aminobutyric acid (GABA), and lycopene are among these bioactive compounds. Utilizing CRISPR-Cas9 technology, researchers have enhanced the levels of anthocyanin, GABA, and lycopene in tomato fruits by modulating the expression of key genes in their metabolic pathways (Cermak *et al*., 2015; Nonaka *et al.*, 2017).

The function of TM6 in strawberry was elucidated using the CRISPR-Cas9 system applied to an octoploid species. Phenotypic analysis of tm6 mutants unveiled pronounced defects in anthers, underscoring TM6's crucial role in flower development (Martín-Pizarro *et al*., 2019). Furthermore, CRISPR-Cas9 was employed to explore the biological role of YUCCA 10 (YUC10) in auxin synthesis during strawberry fruit development. Knocking out YUC10 resulted in a significant reduction in free auxin in yuc10 mutants (Feng *et al*., 2019).

4.2 Disease resistance

CRISPR–Cas13a presents an efficient tool for targeting RNA viruses, predominantly plant viruses. Aman *et al*. 2018 utilized LshCas13a to target Turnip mosaic virus (TuMV) which cause Turnip mosaic disease in *Nicotiana benthamiana*, achieving significant reductions in viral gene expression. The predominant approach for pathogen control via the CRISPR/Cas9 system involves disrupting the host's susceptibility gene (S gene), thus impeding plant-pathogen interactions and preventing pathogen establishment (Zaidi *et al*., 2018). This disruption can be achieved by targeting either the promoter sequence of the S gene or interrupting the effector-binding site. Ali and his coworkers effectively demonstrated virus targeting by inducing indels in the genome of tomato yellow leaf curl virus, thus imparting viral resistance. This resistance was achieved through CRISPR/Cas9 binding to the viral genome, subsequently obstructing the viral genome's access to replication units, or by generating blunt-end cuts or indel mutations on the viral genome. Thomazella *et al*. (2016) utilized the CRISPR-Cas9 system to deactivate the DMR6 ortholog in tomatoes. The resulting dmr6 mutants exhibited disease resistance against a range of pathogens, such as *Pseudomonas syringae*, *Phytophthora capsica*, and *Xanthomonas* spp., with minimal adverse effects. *Pseudomonas syringae* induces bacterial speck disease in tomato plants, which significantly impacts their productivity and market value. Given the role of Jasmonatezim domain protein 2 (JAZ2) in defense against *P. syringae* in *A. thaliana*, scientists employed CRISPR-Cas9 to produce dominant JAZ2 repressors in tomatoes with the C-terminal jasmonate associated (Jas) domain removed (JAZ2Δjas). These JAZ2Δjas repressors confer resistance to *P. syringae*. Nekrasov *et al*. (2017) employed CRISPR-Cas9 technology to create a tomato loss-of-function mlo1 mutant. This mutant exhibited complete resistance to the powdery mildew fungus *Oidium neolycopersici*.

4.3 Herbicide resistance

The application of the CRISPR-based gene editing technique has led to the successful development of crop varieties resistant to herbicides that target the ALS enzyme. This technique has been implemented across various crops, such as rice (Zhang *et al*., 2021), maize (Li *et al*., 2020), wheat (Zhang *et al*., 2019), watermelon (Tian *et al*., 2018), oilseed rape (Wu *et al*., 2020), tobacco (Kang *et al*., 2019), tomato and potato (Veillet *et al*., 2019). Additionally, wheat has shown tolerance to herbicides inhibiting ACCase through cytidine-deaminase-mediated base editor (CBE). To enhance the efficiency of CRISPR/Cas technology, the target-activation induced cytidine deaminase (Target-AID) system has been introduced, facilitating the simultaneous improvement of multiple traits in crops.

In the development of herbicide-resistant crop varieties, only resistance to ALS-inhibiting herbicides, ACCase-inhibiting herbicides, and glyphosate has seen significant success. However, research on the widespread adoption and effective management of weeds with herbicides that target 4-hydroxyphenyl pyruvate dioxygenase and protoporhyrinogen oxidase is lacking.

4.4 Plant stress resistance

Stress poses a formidable challenge to agricultural productivity, with abiotic and biotic stressors exerting detrimental effects on crop yield. Abiotic stressors, encompassing factors such as drought, floods, temperature extremes, salinity, heavy metals, and radiation, disrupt plant growth and development. Conversely, biotic stress arises from attacks by various pathogens including viruses, bacteria, fungi, and herbivores, further compromising crop health and productivity. To mitigate these challenges, crops such as rice, tomato, cucumber, and grapefruits have been genetically modified through induced mutations to enhance resistance to both abiotic (Klap *et al*.,

2017) and biotic stresses (Lu *et al*., 2018). While earlier attempts at site-specific genomic mutation relied on DNA-binding endonucleases such as zinc finger nucleases (ZFN) and transcription activator-like effector nucleases (TALEN), these approaches have inherent limitations (Christian *et al*., 2010). The advent of the CRISPR–Cas system marked a significant breakthrough, enabling precise genome editing in a wide range of crops including rice, wheat, *Nicotiana benthamiana*, and *Arabidopsis* (Chen *et al*., 2019). In their study, Li *et al*. (2018) discovered that C-repeat binding factor 1 (CBF1) plays a crucial role in safeguarding plants against cold injury. The cbf1 mutant, created using CRISPR-Cas9, displayed exacerbated chilling-injury symptoms with

Table 1: CRISPR-Based crop improvement studies in important agriculture and horticulture crops.

Crops	Targeted gene	Result	References
Agricultural crops			
Rice	*OsSEC3A, OsSWEET13, OsERF922*	Resistant to blast and bacterial blight	Ma *et al*., 2018
Rice	*ALS*	Herbicide resistance	Chen *et al*., 2019
Rice	*UVb1-1*	Resistant to false smut	Mishra *et al*., 2018
Rice	*OsGS3*	Increase in grain size	Miao *et al*., 2013
Wheat	*EDR1*	Resistant to powdery mildew	Zhang *et al*., 2017
Barley	*ENGase, HvPM19, BolC.GA4.a*	Increase in number of grains	Kapusi *et al*., 2017
Maize	*ARGOS8*	Drought resistance	Svitashev *et al*., 2016
Horticultural crops			
Tomato	*SlMLO1*	Resistant to powdery mildew	Nekrasov *et al*., 2017
Potato	*ALS*	Herbicide resistance	Choudhury *et al*., 2016
Cucumber	*eIF4E*	Broad virus resistant	Sauer *et al*., 2016
Apple	*DIPM1, DIPM2, DIPM4*	Resistant to fire blight disease	Malnoy *et al*., 2016
Kiwifruit	*CEN4, CEN*	Rapid flower and fruit development	Varkonyi *et al*., 2018
Grape	*VvMLO3*	Resistant to powdery mildew	Wan *et al*., 2020
Citrus	*CsLOB1*	Resistant to citrus canker	Jia *et al*., 2017
Cocoa	*TcNPR3*	Resistant to *Phytophthora tropicalis*	Fister *et al*., 2018
Watermelon	*ALS*	Herbicide resistance	Tian *et al*., 2018
Papaya	*alEPIC8*	Resistance to *Phytophthora palmivora*	Gumtow *et al*., 2018
Cassava	*EPSPS*	Herbicide resistance	Hummel *et al*., 2018
Soybean	*GmSPL9a, b, c*	Increase in yield	Bao *et al*., 2019
Mushroom	*PPO*	Browning resistant	Waltz, 2016

increased electrolyte leakage compared to wild-type (WT) plants. Additionally, MAPK3, known for its involvement in resisting gray mold disease (Zhang *et al*., 2018), also contributes to tomato drought response by shielding cell membranes from oxidative damage.

Customized sgRNA-Cas9 systems have emerged as a widely employed tool for genome modification in crops like rice and wheat, showcasing the ease and efficiency of genome editing (Shan *et al*., 2013). Notably, Cas12a, formerly known as Cpf1, presents advantages over Cas9 in plant genome editing due to its requirement of shorter guiding nucleotides, ability to create larger deletions at target sites, and facilitation of NHEJ-mediated donor DNA insertion (Kim *et al*., 2017).

In *Arabidopsis*, Feng and coworkers successfully demonstrated the mutation and heritability of five endogenous target genes – *brassinosteroid insensitive 1 (bri1), jasmonate-zim-domain protein 1 (jaz1), gibberellic acid insensitive (gai), magnesium chelatase subunit i (chli),* and *transparent testa 4 (tt4)*, in addition to the *apetala1* (*ap* 1) gene, using CRISPR–Cas tools (Feng *et al*., 2014). Furthermore, CRISPR–Cas technology can be harnessed for the regulation of genes responsible for epigenetic modification, methylation, and/or demethylation, enabling simultaneous induction and repression of gene expression (Puchta, 2016).

Hybrid breeding, alongside precision plant breeding facilitated by CRISPR–Cas, holds promise for increasing crop productivity (Chen *et al*., 2019). CRISPR–Cas has been instrumental in producing thermosensitive male-sterile lines in rice (Zhou *et al*., 2016) and maize (Svitashev *et al*., 2016), facilitating the production of high-quality hybrid varieties. Additionally, knockout mediated by CRISPR–Cas has enabled the development of herbicide-resistant crops in rice (Shimatani *et al*., 2017), *Arabidopsis* (Chen *et al*., 2017), and watermelon (Tian *et al*., 2018), further expanding the scope of genome editing applications in plants.

5. CRISPR crop regulations and ethics

The regulatory framework surrounding CRISPR-modified crops varies significantly among countries and regions. In some jurisdictions, CRISPR-edited crops that do not involve the insertion of foreign DNA are subject to less stringent regulations compared to traditional genetically modified organisms (GMOs). For example, the European Union (EU) has classified some CRISPR-edited crops as non-GMOs, thereby exempting them from rigorous regulatory requirements (Eckerstorfer *et al*., 2019).

In contrast, other countries, such as the United States, have adopted a case-by-case approach to regulate CRISPR-modified crops, evaluating them based on their characteristics and potential risks to human health and the environment. The U.S. Department of Agriculture (USDA), the Environmental Protection Agency (EPA), and the Food and Drug Administration (FDA) play key roles in assessing the safety and environmental impact of CRISPR-modified crops (Waltz, 2018).

5.1 Ethical Implications

CRISPR-mediated genome editing in agriculture raises various ethical considerations that must be carefully addressed. One of the primary concerns revolves around unintended consequences and potential ecological impacts of genetically modified crops. Altering genes in crops could inadvertently affect ecosystems, biodiversity, and non-target organisms, leading to unforeseen environmental consequences (Lassoued *et al.*, 2019).

Additionally, ethical considerations extend to issues of social justice and equity in access to CRISPR technology and its benefits. There is a risk that CRISPR-based agricultural innovations could exacerbate existing inequalities, favoring large agro-industrial companies and marginalizing small-scale farmers and resource-constrained regions. Ensuring equitable access to CRISPR technology and its benefits is essential for promoting social justice and addressing global food security challenges (Levidow and Carr, 2020).

Furthermore, questions surrounding informed consent, transparency, and public engagement in decision-making processes related to CRISPR-modified crops are paramount. Stakeholder involvement, including farmers, consumers, policymakers, and civil society organizations, is crucial for fostering transparency, accountability, and democratic governance in agricultural biotechnology.

6. Conclusion

The advent of CRISPR-Cas technology heralds a new era in agriculture, offering unparalleled precision, efficiency, and adaptability in crop enhancement. With its transformative potential, CRISPR-Cas has emerged as a powerful tool for addressing the multifaceted challenges confronting contemporary agriculture. By leveraging the capabilities of CRISPR-Cas, researchers are poised to revolutionize crop breeding practices, enabling the development of resilient, high-yielding, and nutritionally enriched cultivars.

CRISPR-Cas-mediated genome editing facilitates the targeted modification of specific genes, thereby accelerating the breeding process and circumventing the limitations of traditional breeding methods (Zhang *et al*., 2019). This precision breeding approach holds tremendous promise for enhancing crop traits such as disease resistance, abiotic stress tolerance, and nutritional quality, thereby bolstering agricultural productivity and resilience in the face of climate change and environmental pressures (Wang *et al*., 2020). Furthermore, the versatility of CRISPR-Cas extends beyond genetic modification to encompass epigenome editing and gene regulation, offering novel avenues for crop improvement (Shimatani *et al*., 2017). By precisely modulating gene expression patterns and regulatory networks, CRISPR-Cas9 enables fine-tuning of agronomically important traits, such as flowering time, yield components, and nutrient utilization efficiency.

Moreover, CRISPR-Cas technology holds immense potential for promoting sustainable agriculture and addressing global food security challenges (Nalley *et al*., 2019). By enhancing crop productivity, reducing input requirements, and minimizing environmental impacts, CRISPR-edited crops offer a pathway towards achieving food security goals while mitigating the ecological footprint of agricultural production systems.

In conclusion, CRISPR-Cas technology represents a paradigm shift in agriculture, offering unprecedented opportunities for crop improvement and sustainable development. By harnessing the power of CRISPR-Cas, researchers can accelerate the pace of genetic improvement in crops, cultivate resilience to environmental stresses, and contribute to the realization of a food-secure future for generations to come.

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