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Unraveling nutrient stress in aonla (*Emblica officinalis*) : comprehensive insights into research and utilization- a review

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Abstract

Aonla (*Emblica officinalis* Gaertn), also known as Indian gooseberry, is a time-honored fruit tree species belonging to the Euphorbiaceae family. Widely distributed across diverse ecological regions, aonla demonstrates remarkable adaptability to varying edapho climatic conditions. The crop is highly valued for its rich nutritional value, therapeutic properties, and significant post-harvest and industrial applications. Historically, aonla's pharmaceutical benefits have been documented in ancient litrature, both as a standalone remedy and in combination with other ingredients. To enhance the utilization and improvement of this invaluable genetic resource, it is crucial to delve into studies on its origins, diversity, and geographical distribution. To achieve a deeper insight into its taxonomy, systematics, and biological traits, including phenology, floral biology, and pollination mechanisms, detailed investigation is essential. In addition to fundamental biological aspects, agro-technologies, propagation methods, canopy architecture, and strategies for managing physiological disorders, pests, and diseases play a critical role in conserving aonla germplasm and boosting production. This review aims to comprehensively address these areas, offering insights into the genetic, ecological, and agronomic aspects of aonla cultivation, with a focus on its conservation and enhanced production techniques.

Key words: Indian gooseberry, *Emblica officinalis*, Crop improvement, Agronomy, Genetic resource, Biotechnology, High-density planting

Aonla (*Emblica officinalis* Gaertn), commonly known as Indian gooseberry, is a significant fruit crop native to India, cultivated across the country in a wide range of agro-climatic conditions (Tiwari *et al.*, 2007). Revered as a sacred tree, it has been referred to as 'Amritphal' in ancient texts. Beyond India, Aonla is found growing in natural forests across various regions, including Cuba, the USA, Pakistan, Sri Lanka, Malaysia, China, Java, and the West Indies. The fruit plays a pivotal role in Ayurvedic medicine, particularly in the preparation of well-known formulations such as Triphala and Chyavanprash (Tiwari *et al.*, 2008). Owing to its robustness, adaptability to diverse wasteland conditions, high productivity, and remarkable nutritional and therapeutic attributes, aonla has emerged as one of the most promising fruits of the 21st century. Aonla is known for its medicinal benefits, including its effectiveness in treating conditions like dysentery, diarrhea, jaundice, anemia, bronchitis, and cough. The fruit is commonly processed into various value-added products such as murabba, candy, pickles, and jellies. Additionally, aonla powder, known for its high vitamin C content, is considered superior to synthetic alternatives in combating deficiencies.

It is a highly adaptable plant, thriving in diverse agro-climatic conditions. Although classified as a subtropical fruit, it successfully grows in tropical, arid, and rainfed semi-arid regions. In India, it is cultivated from the coastal areas of South India to the foothills of North India, and natural plants can even be found up to 1,800 m above sea-level (Pathak, 2003; Malik *et al.*, 2010). Mature aonla trees are resilient to both freezing temperatures and heat up to 48°C, although they can be susceptible to frost during winter, particularly in the hot, arid ecosystems of Rajasthan (Pathak *et al.*, 1993.

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Genetic resources and varietal wealth

Aonla, a hardy and versatile fruit, has exhibited remarkable genetic variability across India, particularly in regions like Uttar Pradesh, Uttarakhand, Bihar, Gujarat, Madhya Pradesh, and Rajasthan (Bala *et al.*, 2009; Pathak, 2003). This genetic richness primarily stems from seedling propagation and wild relatives found in diverse agro-climatic zones, including the North-Eastern states of India (Pathak, 2003). However, the risk of genetic erosion is high due to deforestation, natural calamities, and the growing dominance of a few popular cultivars in select regions.

Although genetic erosion threatens aonla diversity, significant untapped variability persists among its wild and cultivated forms. Strategic exploration and conservation of these resources are crucial for improving resilience and productivity. Related *Emblica* species, including *E. fischeri* and *E. myrobalan*, may further enrich breeding programs through their stress-tolerant traits (Shukla *et al.,* 2005; Rai *et al.*, 1993; Chandra *et al.,* 1998). Over the years, numerous cultivars have been developed through systematic selection and evaluation of seedlings, particularly in regions like Uttar Pradesh. For instance, the Banarasi genotype, originally selected from wild trees in the Vindhyan hills, laid the foundation for commercial aonla cultivation. Similarly, other cultivars such as *Chakaiya* and *Francis* have been cultivated, although they come with specific limitations like small fruit size or susceptibility to disease (Pathak, 2003; Pandey *et al.*, 2013).

Breeding and cultivar development

Breeding efforts have focused on selecting superior seedlings and cultivars with desirable traits like large fruit size, disease resistance, and high yield. Notable cultivars like NA-4, NA-5, NA-6, and NA-7 have been developed through seedling selection and are now commercially cultivated in different parts of India (Pathak, 2003). *NA-7,* in particular, is the leading cultivar, occupying the maximum cultivation area in the country.

Clonal selection from elite trees has also led to the development of high-yielding varieties like BSR-1 (43.05 t/ha), Goma Aishwarya, and Anand series (Pathak, 2003). However, challenges remain in ensuring stable yields and quality across diverse agro-climatic conditions. The introduction of biotechnological tools offers promising solutions to accelerate the development of disease-resistant and high-yielding cultivars.

Biotechnology and improvement

Conventional breeding methods have struggled to address certain challenges in aonla cultivation, such as susceptibility to diseases like leaf rust, wilt, and anthracnose. The plant's small flower size, varied chromosome numbers, and complex taxonomy have further complicated breeding efforts (Pathak, 2003). However, genetic engineering presents an exciting avenue for overcoming these obstacles.

Research has shown potential in developing transgenic aonla through somatic embryogenesis. A protocol for high-frequency somatic embryogenesis from juvenile leaf tissues has been successfully developed by Thilaga *et al.*, (2013), providing a robust foundation for genetic transformation in aonla. In addition, protocols for in-vitro regeneration from various explants, including endosperm, epicotyls, and hypocotyls, have been reported, albeit with some variability in reproducibility.

Seed propagation and rootstock management

Seedlings raised from seeds are not true-to-type plants because they have prolonged juvenility and wide variability. Multiplication through seed is mostly done for raising rootstock. For these purpose seeds from fully mature fruits of *desi* trees should be taken. During March /April before rain, pretreated seeds are sown into the field or perforated polythene bag (40 cm × 15cm) under irrigated hot arid region. Seeds germinate within two weeks. Six months to one year old seedling is used as rootstock, and scion of desired varieties are patch budded on to them during the period from May to September for optimum success. Generally, the seed sown in first week of March in perforated polythene bag size (40 × 15cm) becomes ready for budding in July, which saves about six months, and also helps in effective nursery management than conventional methods (Saroj *et al.*, 2001). For better germination and healthy seedling, seeds should be soaked in 500 ppm GA_3 over night. Effect of age and position of scion shoot on mineral composition in aonla has been reported by Awasthi *et al.*, (1993). Murali, (1997) studied the pattern of seed size, germination and seed viability of tropical trees species in southern India.

Vegetative propagation

Among the various vegetative propagation methods for aonla, budding has proven to be the most practical and effective. Patch budding has consistently shown the highest success rates, followed by modified ring budding. For optimal success, budding operations should be carried out between mid-May and mid-August, though success remains significant if performed up to September (Pathak *et al.*, 2006). For a more traditional method, softwood grafting on rootstocks raised in-situ for one year or more achieved a 70% success rate. *In-situ* patch budding is considered the most effective propagation method under dryland conditions. Singh and Singh (2014) reported that aonla seedlings become graftable when their stems reach about 3.0 cm in diameter at 5–6 months of age. The minimum size for rootstocks should be 0.5 cm in diameter at the time of budding or grafting. Moreover, cuttings treated with IBA (15000 ppm) and given bottom heat (33°C) showed 87.30% rooting success.

In terms of improving the chemical composition and growth of aonla seedlings, studies by Singh and Singh, (2002) explored the effects of gypsum and distillery effluent on shoot growth under north Indian conditions. They found that shoot regeneration was most successful when both BA (0.2 mg/l) and IBA (0.1 mg/l) were included in the media. In South India, where the climate allows extended propagation periods, aonla can be propagated almost 8-10 months a year, thanks to the use of greenhouses and net houses (Pathak, 2003). These controlled environments significantly extend the propagation window, leading to more efficient multiplication and stronger plant development. Overall, a combination of traditional and innovative propagation methods, along with optimal environmental conditions, has led to more efficient cultivation and improved aonla plant production across diverse agroclimatic regions.

Micropropagation

The micropropagating aonla comes with its own set of challenges, particularly in-vitro oxidative browning and contamination during shoot bud culture, which has been a persistent issue. To address the oxidative browning problem, various strategies have been developed, including explant waxing, which has shown about 80% success in initiating cultures, although it does delay bud induction and subsequent proliferation (Mishra *et al.*, 1998). The best time for initiating cultures under North Indian conditions has been found to be August to November, with the highest bud induction rate (76.4%) occurring during this period, followed by April to June. Shoots taken from the 10–15th nodes of slightly green and moderately hard stems yielded the best results, with indeterminate shoots being essential for further propagation (Mishra *et al.*, 2005). For shoot proliferation, Mishra *et al.*, (2006) found that a combination of 4.33 µM GA3, 13.9 µM Kinetin, and 342.11 µM Glutamine promoted the highest shoot proliferation rate—13.33 shoots per culture. A major challenge in aonla micropropagation, however, has been *in-vitro* leaf fall. This issue has been mitigated by the addition of Glutamine (342.11 µM) to the media, which has been found to reduce leaf drop, a common problem in tissue culture (Mishra *et al.*, 2006; Mishra and Pathak, 2001).

For *in-vitro* rooting, ½ MS medium supplemented with 49.20 µM IBA and 10.74 µM NAA has been found to work best for rooting aonla plantlets. This medium results in minimal callus formation at the base of the rooted plants, ensuring strong root development without unnecessary tissue growth. Acclimatization is a vital step in micropropagation, as the plantlets are prone to wilting and environmental stress during this period. The use of Paclobutrazol, a growth retardant, has shown promise in improving acclimatization by enhancing cuticular wax deposition, stomatal closure, and root thickening, thereby reducing wilting in plantlets. When grown in a substrate mixture of soil, sand, and compost, plantlets thrive better than in coconut husk substrates, which were prone to in-vitro leaf fall (Patidar *et al.*, 2010).

Additionally, successful regeneration of plantlets from mature endosperm explants has been reported on MS medium supplemented with auxins (2, 4-D or IAA) and cytokinins (Kinetin or BAP). Cultures grown with both BAP (0.2 mg/l) and IAA (0.1 mg/l) exhibited the highest shoot regeneration rates, resulting in the formation of plantlets and even embryo-like structures.

Other successful strategies have included the use of BMS medium supplemented with various growth hormones such as 2, 4-D and Kinetin to form callus and induce shoot formation. Moreover, secondary embryos derived from the culture also produced multiple shoots, which could be rooted using NAA or IBA to generate healthy, acclimatized plants..In conclusion, micropropagation of aonla holds great potential for the rapid and efficient mass production of planting material. While challenges like oxidative browning, leaf fall, and acclimatization need to be carefully managed, ongoing research and refinements in tissue culture protocols continue to enhance the success of this technique. By overcoming these obstacles, we can ensure a steady and reliable supply of high-quality aonla plants for commercial cultivation.

Planting and orchard establishment

For better orchard establishment, pits of 1 m^3 (1 m x 1 m x 1 m) are dug during May at a spacing of 8–10 m and exposed to the hot sun for about one month. Each pit is then refilled with the top one-third layer of surface soil thoroughly mixed with 20–25 kg of farmyard manure (FYM) and drenched with chlorpyriphos (Singh *et al.,* 2014f). Healthy budded plants are planted during rainy season, preferably soon after occurrence of first rain. Rootstock can also be raised *in-situ* at appropriate distance (8m x 8m or 10 x 10m) for budding with superior clone at pencil thickness stage to get maximum success particularly in semi-arid regions. For *in-situ* budding, rootstock should be planted in the already prepared pit in June and after 30-45 days budding may be carried out on them in the field itself.

High-density planting

A field trial was conducted to determine the effects of various planting systems and plant densities on vegetative growth pattern and their influence on yield and quality attributes of NA-7 aonla. The average plant height was recorded highest in double hedgerow system (8.87m) and the lowest in square system (7.95m), whereas the rootstock (60.52cm), scion girth (58.53cm) and plant spread (7.20m) was measured the maximum in square system of planting. However, these parameters were measured the lowest in double hedgerow followed by hedgerow and cluster planting systems. Results of the study revealed that the mean yield/plant (121.20 kg) was recorded the highest in square, but the yield/ha were recorded the maximum in double hedgerow (284.02 q) and it was the lowest in square system (121.20 q). An increase in yield/ha in double hedgerow system over square, paired, cluster, hedgerow system ranged between 25.62-134.34 per cent under semi-arid conditions, whereas an increase in yield in double hedgerow, hedgerow, cluster and paired over square systems was recorded 134.34, 121.10, 97.51 and 53.77 per cent purely under rainfed conditions (Singh *et al.,* 2014h). Among the different planting systems, the square system exhibited better values for physical qualities, whereas chemical attributes like TSS, total sugar, vitamin C and total phenols were observed the highest in double hedgerow followed by hedgerow planting system. (Singh *et al.,* 2024b; Singh *et al.,* 2007g; Singh *et al.,* 2005b; Singh *et al.,* 2024a; Singh *et al.,* 2011a; Singh *et al.,* 2014g; Singh *et al.*, 2010c; Singh *et al.*, 2014f). There were significant differences amongst different planting system with regards to fruit physico-chemical attributes. The net economic return was computed the highest in double hedgerow (₹ 2,34,020.00) and it was the minimum in square (₹ 1,01,200.00) system of planting under rainfed hot semi-arid conditions (Singh *et al.*, 2018a;Singh *et al.*, 2004h; Singh *et al.,* 2007a; Singh *et al.,* 2014b; Singh *et al.,* 2014a).

Irrigation management

Mature aonla orchards need no irrigation under normal rainfall. Avoid watering during flowering (February–March), but irrigate after applying manures and fertilizers if soil moisture is low. In areas facing water scarcity, the drip irrigation system has shown promising results and is particularly effective in rainfed conditions. Studies have demonstrated that plants irrigated through drip systems, with water applied every alternate day and 60% wetted area, experienced improved growth, yield, and fruit quality (Singh *et al.*, 2008c; Singh *et al.*, 2016c).

Additionally, construction of low-cost earthen dams and a series of check dams have had a significant positive impact on soil moisture conservation, thereby enhancing the productivity of fruit crops under the semi-arid conditions of Gujarat (Meshram *et al.*, 2008; Meshram *et al.*, 2009).

Training, pruning and canopy management

Aonla trees generally do not require regular pruning; however, during the early years, proper training and shaping are essential to develop a strong and well-balanced framework. To achieve this, the tree should be trained with a single stem up to a height of approximately 0.70 meters, and primary branches should be spaced evenly around the trunk. The recommended training method is the modified centre leader system, where the framework is developed by encouraging the growth of four to six well-spaced branches at a wide angle.

The main branches should emerge around 0.70 meters above the ground. Unwanted branches should be pinched off during March-April to maintain a balanced structure. In the following years, 4 - 6 branches should be allowed to grow. Subsequent pruning involves removing dead, infested, broken, weak, or overlapping branches after harvest. It is important to note that limb breakage has been observed in the aonla cultivar NA-7, particularly due to overbearing and narrow angles between branches and the main stem. Therefore, maintaining a balanced canopy is essential for this variety to ensure its structural integrity (Pathak *et al.*, 2006; Singh *et al.*, 2014f).

Integrated nutrient management

The dosage of manures and fertilizers for aonla cultivation depends on factors such as soil fertility, the age of the plant, and the desired production levels. For one-year-old aonla plants, a recommended dose of 15 kg of FYM (Farm Yard Manure), 100 g of nitrogen (N), 50 g of phosphorus (P), and 100 g of potassium (K) should be applied. This dose should gradually increase each year until the plant reaches ten years of age, after which a constant dose should be maintained (Singh, 1998). The growth, yield, and quality of aonla are significantly influenced by various nutrient sources. Additionally, the physical, biological, and chemical properties of the soil are also positively affected by organic nutrient sources.

Several nutrient sources, including biofertilizers like Azotobacter, Azospirillum, PSB (Phosphate Solubilizing Bacteria), and VAM (Vesicular Arbuscular Mycorrhiza), as well as organic matter such as FYM and cakes (neem, mahua, castor, and groundnut), have been proven beneficial for the sustainable production of high-quality aonla fruits (Korwar *et al.*, 2006; Yadav *et al.*, 2007;Singh *et al.*, 2008a;Singh *et al.*, 2012b;Singh *et al.*, 2007b;Singh, 2007;Singh *et al.*, 2014f). Among various organic nutrient sources, combinations such as FYM + neem Cake + CPP (Crop-Promoting Proteins) and FYM + Azotobacter + VAM have been particularly effective in improving soil fertility, enhancing microbial activity, increasing earthworm populations, and boosting both the yield and quality of aonla fruits under semi-arid conditions of western India (Pathak *et al.*, 2006;Singh *et al.*, 2007a;Singh *et al.*, 2008a;Singh *et al.*, 2012a;Singh *et al.*, 2012b;Singh *et al.*, 2014a;Singh *et al.*, 2014c).

Soil moisture conservation

In rainfed semi-arid regions, mulching with materials like paddy straw, maize straw, rice husk, grasses, or subabul loppings was practiced by applying about 20 kg per tree over a 4 m^2 basin area in September, with the mulch maintained for a prolonged duration. Organic materials like paddy straw, maize straw, grasses, and rice husk, which are locally available and easily decomposable, were applied in 20 cm thick layers after the rainy season. These mulching materials have been found to significantly improve soil quality, plant growth, and the yield of NA-7 aonla (Singh *et al.*, 2006; Singh *et al.*, 2014g; Singh *et al.*, 2007e and Singh *et al.*, 2008d). Each year, these materials were thoroughly incorporated into the basin soil at the end of the monsoon season.

Under rainfed conditions, organic mulching has proven to be very beneficial for the successful cultivation of aonla (Singh and Singh, 2004a;Singh *et al.*, 2015a;Singh *et al.*, 2014c;Singh *et al.*, 2010b). Mulches not only conserve soil moisture but also contribute multiple benefits to soil fertility, plant growth, and yield (Rao and Pathak, 1996a and 1998b). Even the leaf litter under the aonla tree canopy serves as an effective mulch, retaining moisture during summer months and improving soil properties.

Among mulching materials, highest average yield per plant (87.48 kg) was recorded with paddy straw mulch, followed by maize straw mulch (84.00 kg), grasses (80.37 kg), and rice husk (79.30 kg). The lowest yield was observed in the control group (76.29 kg) under rainfed conditions in the hot semi-arid ecosystem during the 10th year of orchard life (Singh *et al.*, 2008d). The yield in paddy straw mulch was 14.66% higher than in the control (Singh *et al.*, 2010). In terms of quality, the highest levels of total soluble solids (TSS) (9.50° Brix), total sugar (6.00%), and vitamin C (510 mg/g) were observed in the paddy straw mulch, while the lowest values were recorded in the control (Singh *et al.*, 2010b; Singh *et al.*, 2008a).This demonstrates that mulching, especially with organic materials like paddy straw, plays a key role in enhancing the yield and quality of aonla under rainfed semi-arid conditions.

Crop diversification

Aonla, being a deep-rooted deciduous tree with sparse foliage, is ideally suited for intercropping. The large space between rows and the 2-7 years required for the trees to reach economic production make aonla orchards perfect for cultivating intercrops in the interim. This not only provides growers with an additional source of income in the early years but also helps in maintaining maximum soil cover.

Under the rainfed conditions of Gujarat, cucurbitaceous vegetables such as bottle gourd, sponge gourd, bitter gourd, cucumber, and pumpkin can be successfully grown under aonla based cropping system. Economic analysis of aonla-based cropping systems

Fig.1: Intercropping in aonla orchard with A: bottle gourd and, B : cucumber. (source: Singh *et al.*, 2014f.)

Table 1. Net income, cost benefit ratio under different intercropping systems in aonla under rainfed hot semi-arid conditions

Inter crops	Details of cost and benefits from intercrops					Details of cost benefits from sole crop aonla							
	Produce of intercrop (q/ha)	Rate (₹/kg)	Cost of cultivation /ha (₹)	Gross income (₹)	Net income (₹)	Produce of main crop (q/ ha)	Rate (Rs/ kg)	Cost of cultivation (₹)	Gross income (₹)	Net income (₹)	Input costs (₹)	Total net Return (₹)	B:C ratio
Bottle gourd	79.95	15.00	18142.20	119925.00	101782.80	60.53	10	15000	60530.05	45530.50	33142.20	147312.80	4.44
Pumpkin	74.40	10.00	17225.00	074400.00	057175.00	58.35	10	15000	58350.03	43350.00	32225.00	100525.00	3.11
Bitter gourd	49.14	15.00	18035.50	073521.00	055485.50	56.03	10	15000	56030.50	41030.40	33035.50	96515.00	2.92
Cucumber	68.61	10.00	16054.83	068610.15	052555.17	52.85	10	15000	52850.15	37850.00	31054.83	90405.17	2.91
Sponge gourd	43.73	10.00	15147.57	043730.20	028582.43	59.50	10	15000	59500.00	44500.07	30147.57	73082.43	2.42
Control	--------	------	---------	----------	--------	64.65	10	15000	64650.30	49650.52	01500.00	49650.00	------

has shown that the highest net returns of Rs. 147,312.80 per hectare and Rs. 1,000,525.00 per hectare can be obtained from aonla + bottle gourd and aonla + pumpkin intercropping, respectively, in the semi-arid ecosystem (Singh *et al.*, 2013b;Singh *et al.*, 2008b;Singh *et al.*, 2007d). Detailed information on net income and cost-benefit ratios can be found in Table 1.

Hiwale, (2014) reported that aonla + okra intercropping yielded a net return of Rs. 72,505 per hectare without negatively affecting the aonla crop. Among other suitable intercropping options, guava (Sardar) and karonda can also serve as effective filler crops in aonla plantations (Singh, 1998). A variety of other intercrops like okra, bottle gourd, cauliflower, coriander, matricaria, gladiolus, and marigold have been successfully grown alongside aonla plantations) Krishna *et al.*, 2013; Krishna *et al.,* 2018).This intercropping strategy not only enhances farm profitability but also improves the sustainability of aonla orchards, making it a win-win for growers in semi-arid regions.

Fruit setting, fruit growth and development

In North India, aonla trees begin shedding their determinate shoots starting from February, leaving the indeterminate shoots devoid of foliage by mid-March. New determinate shoots emerge at the nodes from the scars left by the abscission of the previous season's determinate shoots, beginning in late-February and continuing through the first week of April. Blossom buds appear on the newly developed determinate shoots, but shoots that emerge after mid-April do not bear flowers (Pathak *et al.*, 2006).

The zygote remains dormant for 120-130 days, and the endosperm nucleus persists for 70-80 days after fertilization (Bajpai, 1968;Ram, 1971). After fruit set, the embryo stays dormant, and the ovary shows no noticeable external growth until mid-August. The fruit, however, grows rapidly during the rainy season from the second fortnight of August to the last week of September, completing around 70% of its growth during this period. Growth slows between the second week of October and the first week of November, then picks up slightly thereafter. The variability was observed among different germplasm of Aonla fruit such as small, medium and large. Growth ceases after November. Thus, the fruit growth of aonla follows a double sigmoid pattern.

Auxin levels rise in the fruit as dormancy sets in during April, gradually peaking by the end of May, then decrease in June, reaching their lowest levels in July before dormancy breaks. These elevated auxin levels halt fruit growth, a phenomenon confirmed by the application of IAA, which translocates from the shoot tip to the fruit. It was also found that external auxin applications do not break dormancy and shoot tips were found to contain similar auxin levels as dormant fruits. Additionally, restricting shoot growth and inducing flowering through defoliation from July to October prevents fruit dormancy. The levels of cytokinin and gibberellins were also found to be inadequate in dormant fruits.

Since aonla is self-incompatible, planting about 10% compatible varieties as pollinizers is recommended to ensure adequate fruiting. The physiology and vitamin content in aonla have been studied by Amal and Raghwan, (1957). Significant differences in the physical and chemical composition of aonla cultivars during their growth and development have been documented by Ojha and Pathak, in North India, and by Singh *et al.*, (2014b) under semi-arid conditions of western India.

Maturity, harvesting, yield and quality attributes

Aonla fruits should be harvested once they reach full maturity to ensure optimal quality. The maturity of aonla fruits can be assessed visually by the change in color from greenish to yellowish-green or light-green to pinkish tinge, along with a shift in seed color from creamy white to brown fig. In addition to these visual cues, physiological maturity can also be determined based on factors such as specific gravity (ranging from 1.07 to 1.24), fiber content, and TSS/acid ratio (which typically falls between 5 and 6).

Varieties like NA-7, Banarasi, and Agra Bold generally mature by the last week of October, while Kanchan and Chakaiya are ready for harvest by the last week of November. Varieties like Anand-1and Anand-2 mature by the last week of November under the semi-arid conditions of Gujarat (Singh *et al.*, 2008a; Singh *et al.*, 2006b).

For proper harvesting, aonla fruits should be individually picked and placed carefully in lined baskets to avoid bruising and spoilage. Figure 9 depicts the variability in fruit surface among different aonla germplasm, showing both shiny and dull types.Aonla trees begin fruiting from the third year after budding, with commercial yields typically starting from the fifth year. Under rainfed conditions in Gujarat, when planted at a spacing of 10m × 10m, 10-year-old trees of varieties like *Chakaiya, NA-7, Francis, Kanchan,* and *Krishna* yield 14-15, 15-18, 13-15, 8-9, and 7-8 tons per hectare, respectively (Hiwale, 2015; Singh,2007). The yield and quality attributes of various aonla cultivars have been thoroughly studied. Furthermore, the physico-chemical characteristics of aonla cultivars have been examined under the rainfed semi-arid conditions of Gujarat.

Post-harvest handling, storage and value-addition

Aonla fruits can be stored on the tree itself for up to 30 days post-maturity without a significant loss in quality

(Singh *et al.,* 2007c). Varieties such as Chakaiya and Anand-2 have a shelf life of 7 days, while Francis, NA-7, and Banarasi*can* last up to 5 days at ambient temperature after harvesting. However, when stored in brine solution, the shelf life can be extended up to 75 days. The fruits of NA-7, Banarasi, and Agra Bold mature by the last week of October, while Kanchan and Chakaiyaare ready for harvest by the last week of November. Additionally, Anand-1 and Anand-2 mature by the last week of November under the semi-arid conditions of Gujarat.

Treating aonla fruits with chemicals and storing them in a zero-energy cool chamber significantly enhance their shelf life (Singh *et al.*, 2010). Effects of post-harvest treatments such as calcium nitrate (1%), GA3 (50 ppm), and Borax (4%) on the shelf life of aonla fruits. They found that physiological weight loss and pathological damage increased with longer storage periods. Calcium nitrate (1%) minimized weight loss, while Borax prevented pathological loss for up to 9 days of storage. Among aonla varieties, NA-10 and Krishnashowed a better shelf life of 10 days, retaining high vitamin C content and a glossy green appearance. In contrast, *NA-7* fruits could be stored for 6-8 days (Scartezzini *et al.*, 2006) under North Indian conditions.

Five aonla varieties (Banarasi, Chakaiya, Francis, Kanchan, and Krishna) were assessed for their yield performance, physico-chemical properties, and sensory quality of processed products. The study revealed that Kanchan and Krishnaare suitable for making candy and jam, while *Banarasi* is best for drying (Pragati *et al.*, 2003). The Chakaiyavariety, known for its desirable attributes, received the highest scores for products such as pickles, chutneys and syrups.

The shelf-life of aonla fruits can be further enhanced by grading, packaging, and using pre- and post-harvest chemical treatments, as well as plant hormone sprays (Singh and Singh, 2006; Kumar *et al.,* 2013). After harvesting, fruits should be graded based on their size and shape. Grade A fruits (with a diameter and length greater than 4.00 cm) are ideal for making *murabba* and candy, Grade B fruits (smaller size) are used for *chavanprash* and *trifla,* while Grade C fruits, which are blemished, are suitable for powder and shampoo production. In the hot semi-arid ecosystem of Gujarat, fruits treated with calcium nitrate (1.5%) or GA3 (50 ppm) and stored in perforated polythene bags showed the least physiological weight loss (2.12-16.00%) and spoilage (2.40-15.60%) and exhibited a shelf life of 11 days (Singh *et al.*, 2007b).

For packaging, wooden crates with a 20-25 kg capacity lined with polyethylene bags and CFB boxes (10 kg capacity) with newspaper liners are most effective for long-distance transportation, while for local markets, fruits are collected and transported in plastic crates lined with newspaper.

Studies on changes in ascorbic acid and total phenols during the processing of aonla products have been conducted by Agarwal and Chopra, (2004). They found that decay loss was lowest (26.56%) under modified storage conditions by the 24th day, while the highest decay loss (48.70%) occurred in the zero-energy chamber. Aonla fruits can also be stored in cold storage for 7-8 days at 0-2°C and 85-90% relative humidity.

Although, aonla is not typically consumed as a table fruit, it has significant commercial value in processing industries. The fruits are used to produce a variety of products such as *murabba,* candy, chutney, toffee, shreds, sauce, pulp, powder, juice, laddu, supari, liqueur, sharbat, and Ayurvedic medicines like *chawanprash, trifla,* syrup, diabetic powder, and aonla powder. Additionally, it is used in the cosmetic industry for products like shampoo, hair oil, and dyes (Goyal *et al.*, 2008).

Pests management

Aonla is generally resistant to serious diseases; however, insect pests can cause significant damage, particularly under favorable environmental conditions. The key pests affecting aonla and their recommended control measures are outlined here. To enhance the production potential of aonla, effective and economical Integrated Pest Management (IPM) strategies have been developed, taking into account the seasonality and peak occurrence periods of these pests.

Notable pests of aonla include aphids (*Cerciaphis emblica*), mealy bugs (*Nipaecoccus vastator*), leaf twisters (*Caloptilia acidula*), hairy caterpillars (*Euproctis flava*), shoot gall makers (*Betousa stylophora*), fruit borers (*Virochola isocrates, Meridarchis scyrodes*), and bark-eating caterpillars (*Inderbela terraonis* Moore) (Pathak *et al.*, 2006;Singh *et al.*, 2014f). Orchard sanitation plays a vital role in managing bark-eating caterpillars and borers. Injecting kerosene or petrol, applying Diclorvos, and sealing the holes with mud are effective methods for controlling bark-eating caterpillars.

For general pest management, a tri-weekly application of Dimethoate (0.05%) in the evening is recommended, as it effectively controls a wide range of pests. Based on pest seasonality, a schedule alternating Dimethoate (0.05%) with 5% NSKE (Neem Seed Kernel Extract) every 10 days, from fruit set to fruit development, has proven highly effective in reducing borers in aonla (Pathak *et al.*, 2006; Bajpai,1957; Singh *et al.*, 2010c). For bark-eating caterpillars, spraying Dimethoate at 1.5 ml/l in the evening, every 15 days in January, has also shown significant control (Singh *et al.*, 2007d).

Diseases management

Rust (*Ravenaliia emblicae*), anthracnose (*Colletotrichum* state of *Glomerella cingulata*), and Penicillium fruit rot (*Penicillium indicum*, *P. oxalicum*, *Aspergillus niger*) are significant diseases that cause considerable losses to aonla growers (Singh *et al*., 2010c). To effectively manage rust, a spray of 2% wettable sulphur or 0.2% Mancozeb 75 WP should be applied at 10-12 day intervals starting in August. For anthracnose control, sprays of 0.2% Mancozeb 75 WP, 0.3% Cuman-L, 0.2% Captaf, 0.2% Chlorothalonil, or 0.3% Copper oxychloride are recommended, along with practices like deep ploughing and healthy cultivation. Penicillium fruit rot, primarily a post-harvest issue, can be controlled by sorting and destroying infected fruits, and avoiding injuries during harvesting, handling, transportation, or storage. Pre-harvest sprays (one week before harvesting) with Blitox, Bavistin, or KH_2PO_4, and post-harvest treatments with 1.5% $CaCl_2$ and 200 ppm GA_3, have also proven effective in managing the disease (Singh *et al*., 2014f;Singh *et al*., 2010c).

Physiological disorder

Fruit necrosis, a physiological disorder in aonla, has been associated with boron deficiency. The disorder initially manifests as browning of the mesocarp, which gradually extends to the epicarp, resulting in a brownish-black discoloration of the fruit flesh. This issue can be managed by spraying a 0.6% borax solution three times at 15-day intervals, starting from early September (Pathak, 2003; Pathak *et al*., 2006).

CONCLUSION AND FUTURE THRUST

Aonla holds immense potential to contribute substantially to India's economy, underscoring the need for its wider popularization. This can be achieved through the development of new self-fruitful cultivars possessing desirable horticultural traits and by expanding its cultivation in arid and semi-arid regions. To enhance the crop's performance, emphasis must be placed on harnessing available genetic resources through extensive exploration and germplasm collection. In addition to selection, further research on the inheritance patterns of specific traits is essential. Advanced breeding techniques, including hybridization, mutation breeding, and molecular approaches, should be adopted to expedite the development of aonla cultivars suited to specific industrial and regional requirements. Furthermore, intensified research on high-density planting systems, value addition, and efficient marketing strategies will be instrumental in boosting the production and productivity of aonla.

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From soil to shelf: challenges and opportunities in medicinal plant research

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Abstract

Medicinal plants playing a vital role in traditional healthcare, have emerged as a cornerstone in the development of modern pharmaceuticals. Increasing global demand for natural therapies, along with advances in science and technology, is transforming the medicinal plant sector. This review highlights emerging trends like DNA barcoding for identifying and conserving medicinal plants, as well as the use of genomics, nanotechnology, and artificial intelligence in medicinal plants. Despite this progress, the sector faces challenges, including biodiversity depletion, adulteration, lack of standardization, weak regulatory frameworks, biopiracy, and climate-related threats. Addressing these issues needs global collaboration to ensure sustainable cultivation, ethical resource use, and effective quality control. The integration of cutting-edge tools with traditional knowledge offers a promising path toward developing safe, effective, and eco-friendly plant-based healthcare solutions.

Key words: Biodiversity conservation, Medicinal plants, Pharmaceuticals, Sustainable cultivation, Traditional knowledge, Quality control

With growing global interest in natural therapies and sustainable living, medicinal plant industry is undergoing significant changes. Medicinal plants have been used by humans for thousands of years, forming the foundation of traditional healing systems across cultures (Das, 2014). Ancient systems like the Charaka Samhita, Unani medicine, and Patanjali's practices highlight the importance of medicinal plants in managing various health conditions. However, this surge in demand is accompanied by complex challenges ranging from biodiversity loss and quality control issues to regulatory and ethical concerns, limit its full potential (Raju and Das, 2024). At the same time, emerging technologies such as genomics, nanotechnology, artificial intelligence, and DNA barcoding are opening new opportunities for innovation, quality enhancement, and sustainable use in medicinal plant conservation (Singh *et al*., 2025; Nazar *et al*., 2025). Although limited literature is available, most studies cover only a few aspects of medicinal plant research. This review highlights current trends, challenges, and future directions, emphasizing the need for an integrated approach. It addresses key issues across the entire value chain, from soil to shelf, and promotes the use of both traditional knowledge and modern science to create safe, effective, and eco-friendly plant-based products.

Current trends in medicinal plant research

Medicinal plant research is undergoing rapid growth driven by technological advancements, evolving consumer preferences, and greater integration with modern healthcare systems. There is a rising global demand for herbal medicines, nutraceuticals, and cosmeceuticals as people seek safer, natural alternatives to synthetic drugs (Sarkar *et al*., 2024). This trend is encouraging industrial investment and policy support for the development of plant-based products. At the same time, traditional knowledge is being scientifically validated, leading to its incorporation into mainstream medicine and expanding therapeutic options. Advanced tools such as genomics, metabolomics, and artificial intelligence are also being increasingly used for plant identification, quality assurance, and drug discovery, making research more precise and efficient (Singh *et al*., 2025). These developments collectively signal a strong shift toward innovation, sustainability, and holistic healthcare in medicinal plant sector.

DNA barcoding inauthentication and conservation

The DNA barcoding is a powerful molecular tool used to verify the botanical identity of medicinal plants and prevent adulteration. By analysing short, standardized genetic sequences, it ensures the authenticity, safety, and quality of herbal products (Nazar *et al*., 2025). Since its introduction, technique has evolved to include advanced methods like high-resolution melting (HRM), next-generation sequencing (NGS), and metabarcoding, enabling the identification of multiple species within complex samples (Mahima *et al*., 2022). Technologies such as DNA microarrays further enhance rapid screening capabilities. As the herbal medicine market expands,

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DNA barcoding supported by comprehensive databases of authentic and adulterant species plays a crucial role in quality control, biodiversity conservation, and future drug discovery.

Use of nanotechnology

Nanotechnology is revolutionizing medicinal plant research by enhancing the delivery, stability, and effectiveness of plant-derived compounds. Many phytochemicals have poor solubility, low bioavailability, and rapid degradation, limiting their therapeutic potential. Nano-formulations such as nanoparticles and nanocapsules overcome these challenges by improving absorption, targeting specific organs, and reducing toxicity (Singh *et al*., 2025). This enables lower dosages with enhanced efficacy. Additionally, plant extracts are increasingly used in the green synthesis of nanoparticles, offering sustainable and eco-friendly applications such as herbal herbicides and antimicrobial agents. The integration of nanotechnology with herbal medicine opens new avenues for developing safer, more efficient plant-based therapies.

AI, Machine Learning and GIS

Artificial Intelligence (AI), Machine Learning (ML), and Geographic Information Systems (GIS) are revolutionizing medicinal plant research and conservation. AI and ML tools, such as Convolutional Neural Networks (CNNs) and pre-trained models like VGG16 and VGG19, are being used to predict bioactivity, identify plant species, optimize extraction methods, and discover synergistic phytochemical interactions (Dey *et al*., 2024). India is the first country to use AI to digitize traditional medical knowledge through the Traditional Knowledge Digital Library (TKDL), covering Ayurveda, Siddha, Unani, and Sowa-Rigpa to preserve ancient practices and support research. These technologies also support remote identification of rare or inaccessible plant species through drone imagery and automated analysis. Simultaneously, remote sensing and GIS technologies help map the spatial distribution of medicinal plants, identify potential cultivation zones, and monitor endangered species (Hirapara *et al*., 2024). By integrating environmental variables, GIS enables predictive modeling and precision farming, aiding sustainable cultivation and resource management (Singh *et al*., 2025). Together, these advanced tools enhance research, conservation, and commercialization of medicinal plant resources.

Application of genetics and biotechnology

Genetics and biotechnology play a crucial role in improving the quality, yield, and sustainability of medicinal plants. Tools like genome sequencing, molecular markers, and CRISPR-Cas9 enable precise gene editing to enhance traits such as metabolite production, disease resistance, and adaptability (Singh *et al*., 2025; Raju *et al*., 2025). Techniques like RNA-seq and RAD-seq help identify genes linked to biosynthesis and support conservation and breeding efforts. Tissue culture methods, including callus and cell suspension cultures, allow controlled, large-scale production of bioactive compounds. Cryopreservation of synthetic seeds ensures long-term conservation and sustainable propagation of elite medicinal plant germplasm. Omics-based tools namely genomics, transcriptomic, metabolomics, and proteomics along with bioinformatics platforms, are essential for understanding biosynthetic pathways, and selecting elite genotypes, ensuring a consistent and high-quality raw material supply for pharmaceutical industries (Yang *et al*., 2023).

Challenges

Despite significant advancements, the medicinal plant sector faces several critical challenges that limit its sustainable growth and potential. Unsustainable harvesting and overexploitation of wild plant populations have led to the decline of many valuable species, pushing some toward extinction (Nayak *et al*., 2025). Habitat destruction due to urbanization, agriculture, and deforestation further threatens plant biodiversity. Additionally, the illegal trade of herbal materials and the lack of standardized regulations contribute to quality issues, adulteration, and loss of consumer trust. Inadequate enforcement of conservation laws and insufficient cultivation efforts exacerbate these problems. Addressing these challenges requires coordinated action in conservation, cultivation, regulation, and awareness to ensure the long-term viability and integrity of medicinal plant resources. The major challenges hindering the growth of medicinal plant sector are discussed (Fig. 1).

Biodiversity loss and overexploitation

Unregulated wild harvesting and habitat destruction are critically threatening medicinal plant biodiversity. Many valuable species like *Nardostachys jatamansi* (Jatamansi), *Picrorhiza kurroa* (Kutaki), *Commiphora wightii* (Guggul), and *Rauvolfia serpentina* (Sarpagandha) are becoming endangered due to overexploitation and slow natural regeneration. This not only disrupts ecological balance but also jeopardizes the sustainability of traditional and modern healthcare systems. Urgent steps like sustainable harvesting, habitat conservation, cultivation, strict bans on wild collection, and seed banking are vital to protect medicinal plants for future generations (Das, 2021).

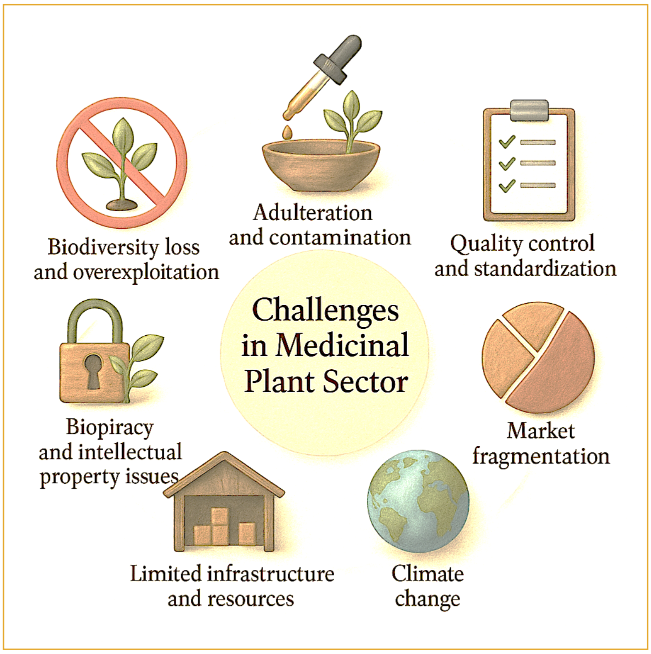

Fig. 1. Overview of major challenges in medicinal plant sector

Adulteration and contamination

Adulteration and contamination of herbal formulations pose significant threats to public health and the integrity of traditional medicine systems. Common issues include the substitution of genuine herbs with inferior or unrelated species, presence of different part of same plant. *Saraca asoca* (Ashoka tree) is often adulterated with the bark of *Polyalthia longifolia*, an ornamental tree with no equivalent medicinal value (Devan and Warrier, 2021). Additionally, contamination from heavy metals, pesticides, and microbial toxins due to poor handling and storage further compromises safety. These challenges highlight the urgent need for strict quality control measures, including DNA barcoding, chemical profiling, and adherence to Good Agricultural and Collection Practices (GACP) to ensure the authenticity, purity, and efficacy of herbal products (Sarkar *et al*., 2024). Common adulterants in medicinal plants and their detection methods are outlined (Table 1).

Quality control and standardization

Inconsistencies in plant identity, cultivation conditions, and processing methods often lead to variability in the quality and efficacy of herbal medicines. The concentration of active compounds in most medicinal plants can vary significantly due to differences in soil conditions, climate, and post-harvest handling practices. Such variability poses challenges in ensuring safety, therapeutic consistency, and global acceptance. Standardization through DNA barcoding, HPTLC/HPLC profiling, and adherence to Good Agricultural and Collection Practices (GACP) and pharmacopoeial guidelines is essential to maintain quality, build consumer trust, and meet international regulatory standards (Das, 2023).

Regulatory and trade barriers

The absence of harmonized global regulations creates major challenges for the development and international trade of medicinal plant products. Due to their multi-component nature, herbal formulations often do not fit into existing pharmaceutical or dietary supplement categories, leading to regulatory ambiguity. EU laws require extensive safety data for herbal products,

Table 1: Common adulteration in medicinal plants and their detection methods

Medicinal plant	Part used	Common adulterant	Method of detection	References
Rauvolfia serpentine (Sarpagandha)	Root	*Rauvolfia tetraphylla* (Devil-pepper)	HPTLC (high-performance thin-layer chromatography) and LC/MS analysis Microscopy, Alkaloid profiling, DNA Barcoding	Sulaiman *et al.* (2020)
Asparagus racemosus (Shatavari)	Root tubers	*Hemidesmus indicus* (Anantamul)	HPTLC, Microscopy	Rai *et al.* (2012)
Centella asiatica (Mandukparni)	Whole plant	*Hydrocotyle sibthorpioides* (Water Pennywort)	DNA barcoding, HPTLC	Maulidiani *et al.* (2012)
Eclipta alba (Bhringraj)	Whole plant	*Wedelia calendulacea* (Water Zinnia)	HPTLC, DNA barcoding, FT-IR, HPTCL	Vadivel *et al.* (2017).
Glycyrrhiza glabra (Licorice)	Root	*Abrus precatorius* (Rosary Pea)	Chemical profiling (TLC, HPLC), Microscopy	Kumar *et al.* (2022)
Terminalia arjuna (Arjuna)	Bark	*T. bellirica* (Baheda)	HPTLC	Tulsi and Vidhu (2022)
Mallotus philippensis (Kamala)	Glandular hair powder from fruit	*Bixa Orellana* (Annatto)	HPTLC, UV-Vis Spectroscopy, Microscopy	Gangwar *et al.* (2014)
Commiphora wightii (Guggul)	Resin	*Boswellia serrata* (Salai guggul)	HPTLC, DNA barcoding, HPLC, GC-MS	Bhardwaj and Alia (2019)

while the U.S. classifies them as dietary supplements with limited health claims (Silpi, 2025). Ayurvedic products like Triphala and ashwagandha face export hurdles due to differing standards across countries. This lack of regulatory alignment delays approvals, restricts market access, and discourages global investment in the herbal sector.

Biopiracy and intellectual property issues

Biopiracy the commercial exploitation of traditional knowledge without proper consent or benefit-sharing poses serious ethical and legal concerns in the medicinal plant sector. Cases like patenting of neem and turmeric by foreign entities, despite their long-standing traditional use in India, highlight the issue. Similarly, *Hoodia gordonii* (Kalahari cactus) traditionally used by the San people to curb hunger and thirst, was commercialized without initially acknowledging their knowledge (Wynberg and Chennells, 2009). Although frameworks like the Nagoya Protocol aim to address such issues, inconsistent implementation especially in developing countries continues to challenge fair access and benefit-sharing.

Limited infrastructure and resources

The development of medicinal plant sector is significantly constrained by inadequate infrastructure, limited funding, and a lack of skilled personnel, especially in developing countries. Despite rich biodiversity, many regions lack advanced laboratories, modern equipment, and institutional support necessary for high-quality research and commercialization, which in turn hampers standardization, value addition, and access to global markets. Strengthening infrastructure, investing in capacity-building, and fostering public–private partnerships are essential to fully realize the potential of medicinal plants (Raju and Das, 2024).

Climate change impacts

Climate change significantly threatens the growth, distribution, yield, and phytochemical integrity of medicinal and aromatic plants (MAPs). Altered temperature regimes, irregular rainfall patterns, and frequent extreme weather events such as droughts, storms, and floods disrupt cultivation cycles and reduce the availability and quality of plant-derived bioactive components (Das *et al.*, 1999). Medicinal plants often reduced yields and altered phytochemical profiles under stress, compromising therapeutic efficacy. Shifts in phenology and geographic distribution of wild medicinal species further highlight their vulnerability. These changes challenge sustainable harvesting, cultivation, and conservation, necessitating urgent development of climate-resilient practices, germplasm preservation, and adaptive management strategies to safeguard the future of medicinal plant resources.

Market fragmentation

The global medicinal plant market is highly fragmented due to the vast diversity of herbal products, traditional healing systems, and region-specific practices. Different countries follow their own systems such as Ayurveda in India, Traditional Chinese Medicine (TCM) in China, Unani in the Middle East, and Kampo in Japan each with unique formulations, plant sources, and therapeutic philosophies (Rohan *et al.*, 2024; Ansari, 2021). *Glycyrrhiza glabra* (Mulethi) is used differently in Ayurveda and TCM, resulting in variations in processing, dosage, and intended use. Such cultural diversity makes it difficult to establish common standards for quality, safety, and effectiveness. This fragmentation makes global marketing difficult as regulations, labeling rules, and consumer expectations vary widely across countries. As a result, manufacturers often face difficulties in aligning their products with international standards, limiting scalability and cross-border trade. Greater collaboration among countries, development of global herbal monographs, and harmonized regulatory frameworks are essential to overcome these barriers and strengthen the global medicinal plant market.

Future directions for research and development

Future research in medicinal plants must adopt strategic, interdisciplinary approaches to overcome current limitations and unlock new opportunities. Key areas include genetic improvement through biotechnology, precision farming for optimized cultivation, and advanced extraction methods like supercritical fluid and nano-based techniques for enhanced efficacy. Technologies such as DNA barcoding and blockchain are strengthening quality control and traceability, while tools like AR (Augmented Reality), VR (Virtual Reality) are revolutionizing education and research. By integrating traditional knowledge with modern science and fostering collaboration among researchers, farmers, and policymakers, these innovations can ensure sustainable cultivation, improved product quality, and global competitiveness of herbal medicines.

Sustainable cultivation and biodiversity conservation

Sustainable cultivation of medicinal plants is essential amid habitat loss, climate change, and shrinking farmland. Eco-friendly practices like organic farming, *in vitro* propagation, and tissue culture help conserve resources and protect ecosystems, ensuring long-term availability of plant material (Nayak *et al.*, 2025). Innovative methods such as hydroponics, vertical gardening, and orchard integration provide space-efficient and climate-resilient cultivation options. Advanced systems like aeroponics and controlled-environment agriculture offer precise growth control, consistent bioactive compound production, and reduced ecological impact. Climate-smart agriculture and robust *in situ* and *ex situ* conservation are vital for protecting genetic diversity and strengthening supply chains (Das and Sharma, 2014). Scientific institutions and industries are also promoting sustainable sourcing through eco-friendly harvesting and biotechnological approaches.

Advanced drug discovery and phytopharmaceutical development

The integration of cutting-edge technologies such as artificial intelligence (AI), network pharmacology, and genetic engineering is revolutionizing drug discovery from medicinal plants. AI-driven platforms can rapidly screen vast phytochemical libraries to predict bioactivity, toxicity, and potential therapeutic targets. Machine learning models have been used to identify novel anti-cancer compounds from *Taxus brevifolia* and anti-inflammatory agents from *Curcuma longa* (Chunarkar-Patil *et al.*, 2024). Network pharmacology allows researchers to understand the multi-target, multi-pathway mechanisms of complex herbal formulations like Triphala or Ashwagandha, providing scientific validation for traditional uses. Genetic engineering and synthetic biology techniques are now being employed to enhance metabolite production, as seen in engineered microbes producing artemisinin (originally from *Artemisia annua*) on a commercial scale (Pulice *et al.*, 2016). These interdisciplinary tools not only accelerate the development of safer and more effective phytopharmaceuticals but also bridge traditional knowledge with modern drug innovation, paving the way for next-generation herbal therapeutics.

Expansion of personalized and integrative medicine

The rise of personalized and integrative medicine is reshaping the future of healthcare by combining plant-based therapies with conventional treatments tailored to an individual's genetic makeup, lifestyle, and health condition. Advances in pharmacogenomics allow for the customization of herbal interventions, enhancing efficacy and minimizing adverse effects (Shaman, 2024) . Integrative approaches are also gaining ground in managing chronic diseases (Kalariya *et al.*, 2023; Bora *et al.*, 2019). Personalized Ayurveda, which aligns constitution types (Prakriti) with genomic data, is another emerging trend. This fusion of traditional wisdom with modern precision medicine improves treatment outcomes, reduces side effects, and enhances overall patient satisfaction.

Enhanced quality control and global standardization

Ensuring the quality, safety, and efficacy of medicinal plant products requires robust quality control measures and globally harmonized standards. Techniques such as HPTLC and HPLC-based metabolite fingerprinting, along with DNA barcoding, are increasingly used to authenticate plant species and detect adulterants. Regulatory frameworks like the Central Drugs Standard Control Organisation (CDSCO), Ayurvedic Pharmacopoeia of India (API), and adherence to Good Manufacturing Practices (GMP) set clear benchmarks for herbal medicine quality. Institutions implementing Good Agricultural and Collection Practices (GACP) further ensure consistency and sustainability (Raju and Das, 2024). Resources like the Traditional Knowledge Digital Library (TKDL) safeguard against biopiracy, while the Herbal Drugs Consultative Committee (HDCC) contributes to regulatory guidance. International cooperation with organizations such as WHO and the U.S. Pharmacopeia supports the global standardization of herbal products, enhancing safety and export potential (Rohan *et al.*, 2024). Integrating modern innovations like green extraction methods, nano-formulations, and high-throughput omics platforms alongside machine learning for precision breeding will accelerate the development of safe, effective, and standardized herbal medicines worldwide.

Ethical bioprospecting and benefit-sharing

Ethical bioprospecting ensures that the exploration and commercialization of medicinal plant resources respect indigenous knowledge and comply with biodiversity regulations. It involves obtaining prior informed consent (PIC), ensuring equitable benefit-sharing, and acknowledging the contributions of local communities (Wynberg and Chennells, 2009). India's Traditional Knowledge Digital Library (TKDL) was developed to prevent biopiracy by documenting indigenous knowledge and making it accessible to patent examiners. Future research must align with frameworks like the Nagoya Protocol, ensuring that local custodians of biodiversity are recognized and rewarded fairly. This approach not only protects community rights but also promotes sustainable and inclusive innovation in the medicinal plant sector.

Climate resilience research

Climate resilience research is vital for sustaining medicinal plant cultivation amid changing environmental conditions. Climate change, including rising temperatures and disrupted seasonal patterns, adversely affects medicinal plant growth, yield, and phytochemical composition particularly secondary metabolites (Das *et al*., 2016). To meet global demand and enhance climate resilience, region-specific cultivation of selected elite varieties under appropriate agro-climatic conditions is being promoted (Saran *et al*., 2025). In parallel, DMAPR has effectively carried out the screening of germplasm suitable for cultivation under low-water conditions in various medicinal plants (Das, 2010). Developing climate-resilient cultivars and adapting cropping systems like intercropping and using drought-tolerant species can help maintain productivity and quality. Integrating predictive climate models and precision farming tools will further support adaptive strategies, ensuring the long-term viability of medicinal plant resources.

CONCLUSION

From soil to shelf, the journey of medicinal plants sector is filled with both challenges and opportunities. While issues like habitat loss, poor quality control, and regulatory gaps pose serious concerns, there is great promise in sustainable cultivation, scientific innovation, and community involvement. Strengthening research, conserving biodiversity, protecting traditional knowledge, and ensuring fair benefit-sharing are key to unlocking the full potential of this sector. Support from institutions like NMPB and DMAPR, along with coordinated efforts across all stakeholders, can lead to a future where medicinal plants play a vital role in health care, rural development, and environmental sustainability. Consumer awareness, improved education about medicinal plants, and the integration of traditional wisdom into modern practices will further enhance the credibility, acceptance, and responsible use of herbal products.

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Retrofitting microbes for inorganic nutrients in citrus nursery: new perspectives

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Abstract

Nutritional health of citrus nursery is the foundation of quality production of mature citrus trees coupled with extended productive life. Retrofitting microbes for nutrient requirement is one of the novel approaches of not only ensuring good health of citrus nursery but cutting down the intensity of mortality during planting into new citrus field. We attempted to tailor the nutrient requirement of citrus nursery through the microbial consortium (*Aspergillus flavus,* MF113270; *Bacillus pseudomycoides,* MF113272; *Acinetobacter radioresistens,* MF113273; *Micrococcus yunnanensis,* MF113274; and *Paenibacillus alvei,* MF113275) developed through extensive isolation, characterization and value addition of different microbial inoculants. The progressive microbial response studies showed that the magnitude of response with microbial consortium outweighed the response of individual microbes with regard to large number parameters, comprising germination percentage, vigour index soil microbial population, changes in available pool of nutrients and leaf nutrient composition, without any additional supplement of inorganic fertilizers. Introducing further the mycorrhizal inoculants, biochars, microbes from rhizospheres of other fruit crops would facilitate towards much better rhizosphere resilience to be fitted in substrate for pre-evaluation of citrus, that could be easily extended to even grown-up orchards as well. With these efforts, we succeeded in retrofitting microbes in place of nutrients to be added from outside sources, synonymous to organic citrus nursery.

Key words: Biochars, Citrus, Plant growth, Leaf nutrients, Microbes, Microbial consortium, Nutrients, Rhizosphere manipulation, Seed vigour, Shelf-life, Soil fertility,

Recognition of the importance of soil microorganisms has led to an increased and thoroughly renewed interest in measuring the quantum of nutrients held in their biomass (Joseph *et al.,* 2015). An increase in the microbial biomass often goes along with increased nutrient immobilization. Plant growth promoting microorganisms play an important role exerting various mechanisms such as biological nitrogen fixation, growth hormone production, phosphate solubilisation siderophore production, hydrolytic enzymes production, antagonistic activity, individually or collectively leading to improved nutrient use efficiency (Srivastava *et al.,* 2015; Srivastava and Bora, 2023). These metabolites can be either overproduced or combined with appropriate biocontrol strains to obtain new formulations for their more effective applications. Studies have demonstrated that *Azotobacter* inoculation alone can substitute up to 50% nitrogen requirement of banana and 25% phosphorus requirement of papaya (Keditsu and Srivastava, 2014).

Microbes have also been reported to substantially improve nutrient acquisition capacity of host plant, and fruit yield in addition to enriching the rhizosphere biologically in a much activated form (Srivastava *et al.,* 2022). Mineral fertilizers on the other hand have limited direct effects, but their application can enhance soil biological activity via increases in system productivity, crop residue return, and soil organic matter (Kohli *et al.,* 1998; Malhotra and Srivastava, 2015). Another important indirect effect especially of nitrogen fertilization is the soil acidification, with considerable negative effects on soil organisms (Ngullie *et al.,* 2015 ; Srivastava *et al.,* 2008).

There are ample evidences accrued through worldwide research that nutrient-microbe synergy is the launching pad for any fruit crop to mobilize and accumulate the required nutrients as per the metabolic nutrient demand, a pre-requisite to improved nutrient-use-efficiency (Srivastava *et al.,* 2015) . While reviewing molecular responses of pants to nutrient stresses, many genes play a central role in the acquisition and distribution of nutrients, including many protein-coding genes as well as microRNAs (miR395, miR398, miR397, and miR408) reported that higher tolerance to nutrient deficiency could be explained by better activation of their antioxidant system (Islam *et al.,* 2022: Srivastava *et al.,* 2022). However, for the other genotypes, tetraploidization did not induce greater tolerance to nutrient deficiency. Rengel *et al.* (1996) observed that the total number of bacterial colony-forming units increased in the rhizosphere of Zn-efficient genotypes of wheat under Zn-deficiency and in Mn-

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efficient genotypes under conditions of Mn-deficiency. In contrast, a Zn-deficiency treatment acted synergistically with the number of fluorescent *Pseudomonas* in the rhizospheres. Fruit crops have displayed an excellent synergy with a variety of microbes, which could play an important role in improving the use efficiency of applied nutrients ((Srivastava, 2013a ; 2013b ;Joseph *et al.*, 2015).

A still bigger question emerges, whether rhizosphere competent microbes could collectively contribute toward improved resilience of plant's rhizosphere against potential nutrient mining. And if those microbes are so successful in promoting growth response, addition of starter nutrients in such combination may further magnify the magnitude of response called nutrient-microbe synergy (Srivastava and Ngullie, 2009 ; Srivastava *et al.*, 2015) . Our earlier studies have shown that rhizosphere effective microbes have the tendency to play multiple roles to overcome various biotic and abiotic stresses while interacting with an environment (Dzuvichu *et al.*, 2023) . Rhizosphere modification through roots by soil microorganisms exudation is an important attribute that regulates not only the availability of nutrients in the soil but also their acquisition by plants (Hota *et al.*, 2020). A number of studies have suggested that whole range of microorganisms have helped to alleviate different abiotic stresses in citrus crop and aid in improving the use efficiency of applied nutrients. Ironically, no citrus crop systematic efforts in the past have been made to tailor the nutrient requirement of nursery plants through microbial interventions (Srivastava and Sharma, 2025). Since healthy nursery plants are pre-requisite to robust citrus industry of future , such issues are mandatory to be addressed.

Microbial Consortium, a Novel Concept

The most common objective of developing microbial consortium is to capitalize on both the capabilities of individual microbes and their interactions to create useful systems in tune with enhanced productivity, and soil health improvements through efficient metabolic functionality (Srivastava and Wu , 2012). Two major underlying principles are applied in the whole process of development of microbial consortium. The first one is resource ratio theory which uses both qualitatively and quantitatively to assess the outcomes between component microorganisms competing for shared limiting resources. This permits coexistence of multiple microbes or the competitive exclusion of all but a single microbe. And the second principle theory relevant to microbial consortium is maximum power principle initially proposed and later modified at various levels, is value for analyzing consortial interactions.

It also dictates that biological systems that maximize fitness by maximizing power, is analogous to metabolic rate or the capacity to capture and utilize energy (Kohi et al., 1998; Srivastava *et al.*, 2002). The microbial consortium is classified as artificial (carrying two or more wild type microbes whose interactions do not typically occur naturally), synthetic (carrying microbes which are modified through manipulations of genetic content), and natural (carrying microbes having much wider applications like bioremediation, wastewater treatment, biogas synthesis etc.).

In the past, a number of studies have suggested the co-inoculation of different microbes as microbial consortium (referred as group of diverse microorganisms having ability to act together in a community and capitalize both the capabilities of individual microbes and their interactions with enhanced response on productivity and soil health improvements through efficient metabolic functionality) is more effective than single inoculation summarized as: *A. brasilense – P. striata/B. polymyxa, A. lipoferem – Agrobacterium radiobacter/A. lipoferem-Arthrobacter mysorens* (Srivastava *et al.*, 2012) , *A. brasilense – Rhizobium, A. brasilense – A. chroococcum – Klebsiella pneumoniae – R. meliloti, A. brasilense – R. leguminosarum,* and A. brasilense/Streptomyces mutabilis – A. chroococcum.

Microbes involving AM fungi and bacteria have also been suggested for improvement in both yield and quality. These include: *A. brasilense – G. fasciculatum* in wheat, strawberry (Amor *et al.*, 2008) and *A. brasilense – Pantoea dispersa* in sweet pepper, and *A. chroococcum – G. mosseae* in pomegranate (Aseri *et al.*, 2008). Later, we at ICAR-Central Citrus Research institute at put forward a microbial consortium, *Aspergillus flavus* MF113270, *Bacillus pseudomycoides* MF113272, *Acinetobacter radioresistens* MF113273, *Micrococcus yunnanensis* MF113274, and *Paenibacillus alvei* MF113275 developed through isolation, characterization, and evaluation of effective microbes from citrus rhizospheres (Srivastava *et al.*, 2019 : Srivastava and Sharma, 2025).

Response of Soil Microbial Inoculation in Acid Lime Nursery

The microbial response study was carried out over the acid lime seedlings at pre-evaluation stage (primary and secondary stages of nursery management) after its morphological and biochemical identification. In the experiment, the progressive response of multiple microbes of the microbial consortium was tested without addition of any inorganic fertilizers through soil inoculation, different microbes were inoculated into the soil (growing medium) on a month-old seedlings of acid lime.

Response of acid lime through soil inoculation: A nursery experiment was set up at CCRI Experimental Farm, Nagpur, to observe the progressive response of different microbes on germination rate of acid lime seeds and subsequent growth. Different treatments consisted of:T_1 (Control), T_2(*Ar, Acinetobacter radioresistens,* MF113273*),* T_3(*Ar, Acinetobacter radioresistens,* MF113273 *+ My, Micrococcus yunnanensis* MF113274) , T_4(*Ar, Acinetobacter radioresistens,* MF113273) *+ My, Micrococcus yunnanensis,* MF113274 *+ Bp, Bacillus pseudomycoides,* MF113272),T_5 (*Ar, Acinetobacter radioresistens,* MF113273)*+ My, Micrococcus yunnanensis,* MF113274 *+Bp, Bacillus pseudomycoides,* MF113272) *+ Pa, Paenibacillus alvei,* MF113275) and T_6 (*Ar, Acinetobacter radioresistens,* MF113273)*+ My, Micrococcus yunnanensis,* MF113274 *+ Bp, Bacillus pseudomycoides,* MF113272) *+ Pa, Paenibacillus alvei,* MF113275) *+Af, Aspergillus flavus,* MF113270) and replicated four times in a CRD experimental design. Microbial treatment as per treatment was applied to the soil over a month-old acid lime seedlings (100 mL) and after 8- days another 100 ml microbial treatment was applied as per the treatment. Response of these microbes was evaluated for changes in germination rate at every 10- days' interval (till 100 days), changes in available nutrient status of soil, leaf nutrient status and microbial status to quantify the magnitude of response with various treatments.

The microbes were observed inflicting response on both seed germination and seed viability index through synthesized microbial metabolites (Srivastava , 2012) . The significant response reported over the germination of acid lime seedlings at the various days of observation. The germination rate was reported as high as 79.8 % with treatmentT_6 at 100-days of observation with seed viability index of 3.20 followed by the treatment T_4, T_5, T_3, T_2 and T_1 respectively in a decreasing order (Table 1). The maximum rate of seed germination was reported within 30-days of observation amongst all the treatments. The seed germination percentage of the treatments T_4 and T_5 was on par with each other depicting the relatively similar response on the growth and development of the growing seedlings in response to added microbes.

Growth response in secondary nursery: Different growth parameters (Shoot parameters viz., shoot length, shoot weight, number of leaves, girth and plant and root parameters viz., root length and root weight) were recorded following the transfer of seedlings from primary nursery to secondary nursery. These growth parameters were significantly affected by treatments (Table 2). The shoot parameters observed higher with the treatment T_6 followed by the treatment T_5, T_4, T_3, T_2 and then control in a decreasing order. The shoot length of the treatments T_4, T_5 and T_6 was on par with each other However, root length and root weight was almost statistically on par with all the treatments, except control, indicating an active response on the root density of the seedlings under the respective treatments (Figs. 1 and 2).

Soil fertility changes: The soil properties like pH and EC were not influenced by any of the microbial inoculation treatments. While organic carbon showed some distinctive changes, which increased from a minimum of 0.10 g kg^{-1} with treatment T_1 to maximum of 0.50 g kg^{-1} with treatment T_6 within120 days of experiment. However, with available soil nutrients the macronutrients like $KMnO_4$-N, Olsen-P, NH_4OAc-K as well as micronutrients like DTPA-Fe, DTPA-Mn, DTPA-Cu and DTPA-Zn reported significant changes suggesting the fact that these microbes bring about the changes in available pool of micronutrients as well as their secondary function. As compared to the control or treatment T_1, maximum increase in the $KMnO_4$-N was observed with treatment T_4, Olsen – P with treatment

Table 1: Changes in germination percentage of acid lime seeds in response to different treatments involving various microbial inoculants

Treatment	Changes in germination percentage (days)										Seed viability index
	10	20	30	40	50	60	70	80	90	100	
T_1 (Control)	15.3	20.1	32.5	39.4	39.4	39.4	39.4	39.4	39.4	39.4	1.05
T_2 (*Ar*)	12.5	18.2	35.6	40.5	40.5	40.5	40.5	40.5	40.5	40.5	1.13
T_3 (*Ar+My*)	17.3	21.3	46.2	50.2	50.2	50.2	50.2	50.2	50.2	50.2	1.79
T_4 (Ar*+My +Bp*)	13.2	19.2	62.5	65.5	65.5	65.5	65.5	65.5	65.5	65.5	2.39
T_5 (*Ar+My +Bp+Pa*)	14.3	20.9	70.3	70.3	70.3	70.3	70.3	70.3	70.3	70.3	2.67
T_6(*Ar+My+Bp+Pa +Af*)	15.7	23.7	79.8	79.8	79.8	79.8	79.8	79.8	79.8	79.8	3.20
CD(*P=0.05*)	NS	1.8	2.8	6.1	5.3	5.4	5.9	5.8	6.1	6.4	-

Ar,My, Bp, Pa, Af, stand for *Acinetobacter radioresistens* (MF113273), *Micrococcus yunnanensis* (MF113274) *Bacillus pseudomycoides* (MF113272), *Paenibacillus alvei* (MF113275) and *Aspergillus flavus* (MF113270), respectively.

Note: Seed viability index was calculated at 100 days of germination as Germination percentage x average seedling length (mm)/100

Table 2: Growth response of acid lime seedlings in response to different microbial inoculations (period: 120- days period)

Treatments	Shoot parameters			Root parameters		
	Shoot length(cm)	Shoot weight(g)	No. of leaves/ plant	Girth (mm)	Root length (cm)	Root Weight (g)
T_1 (Control)	16.9	1.70	17	1.60	9.8	0.36
T_2 (*Ar*)	17.5	2.23	22	1.79	10.6	0.42
T_3 (*Ar+My*)	18.9	2.90	26	2.30	16.9	0.53
T_4 (Ar*+My +Bp*)	21.0	3.60	32	2.92	17.0	0.75
T_5 (*Ar+My +Bp+Pa*)	21.8	3.09	30	2.80	16.0	0.66
T_6 (*Ac+Pf +Bm+*Pa+*Af*)	22.7	3.72	34	2.75	17.5	0.79
CD(*P=0.05*)	0.40	0.23	03	0.10	0.72	0.04

Ar,My, Bp, Pa, Af, stand for *Acinetobacter radioresistens* (MF113273), *Micrococcus yunnanensis* (MF113274) *Bacillus pseudomycoides* (MF113272), *Paenibacillus alvei* (MF113275) and *Aspergillus flavus* (MF113270), respectively.

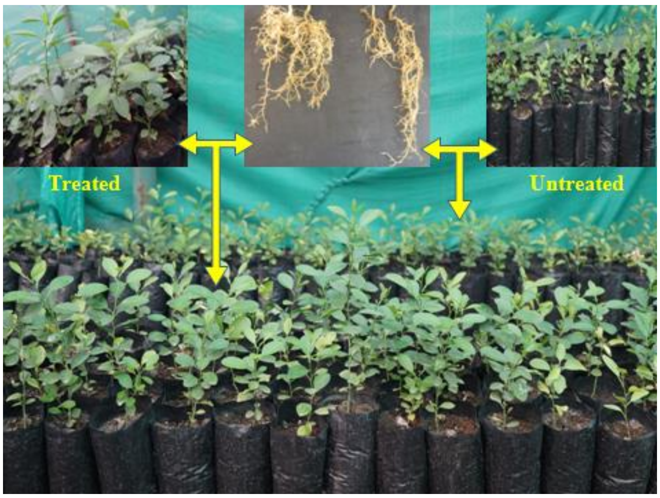

Fig 1: Response of microbial consortium in acid lime (clone NRCC-AL-8) on a commercial-scale use

Fig 2: Response of microbial consortium in acid lime (clone NRCC-AL-7) on a commercial-scale use

T_4 and T_6, NH_4OAc-K with treatment T_6, DTPA-Fe with treatment T_5, DTPA-Mn with treatment T_5, DTPA- Cu with treatment T_5 and T_6 and DTPA-Zn with treatment T_6.

Changes in soil microbial population: Changes in soil microbial population (Bacterial count as well as fungal count) were observed to be significantly affected by different microbial treatments. Among all the treatments maximum bacterial count observed with the treatment T_6 as 73 x 10^3cfu g^{-1} followed by treatments T_4 and T_3, both as 69 x 10^3cfu g^{-1} and 67 x 10^3 cfu g^{-1} with treatment T_2 in decreasing order. Similar response reported with fungal count changes in response to the given treatments. The treatment T_4 recorded fungal count as 36 x 10^3 cfu g^{-1} while the treatments T_2, T_5 and T_6 were statistically on par with each other except control with fungal count as 26 x 10^3 cfu g^{-1} soil (Table 3).

Population dynamics of component microbes: All the component microbes of added consortium showed pragmatic changes in their population over the period of 120 days of observation (Table 4). Highest population of the fungus *Aspergillus flavus* reported with treatment T_6 as34 x 10^3cfu g^{-1}soil. The similar response of increase in population of the fungus *Aspergillus flavus* recorded from initial population of 10x 10^3 cfu g^{-1} to 32 x 10^3 cfu g^{-1}soils with treatment T_4 and T_5. The *Bacillus pseudomycoides* population was also differentially similar with all the other treatments; however highest population of *Bacillus mycoides* reported with the treatment T_6 as 46 x 10^3cfu g^{-1} soil. Maximum population of *Paenibacillus polymyxa* reported with the treatment T_4 as 52 x 10^3cfu g^{-1}soil, *Acinetobacter radioresistens* from initial value of 14 x 10^3cfu g^{-1}soil to 50x 10^3cfu g^{-1} soil with treatment T_5 while the population of *Micrococcus yunnanensis* reported shifts from initial value of 25 x 10^3cfu g^{-1}soil to 55x 10^3cfu g^{-1}soil with treatment T_4 after 120 -days of inoculation.

Leaf nutrient composition: Concentration of micronutrients in leaves showed responses of varying proportion. The leaf micronutrients like Fe was reported maximum with treatment T_6 (90.2 ppm) followed by treatment T_4 (88.9 ppm). The concentration of Mn assessed out under the leaves of different treatments was relatively on par with all the treatments. However highest Mn concentration was reported with treatment T_4

(43.2 ppm). The zinc concentrations were differentially affected by the treatments. Among all the treatments zinc concentration were as high as 17.5 ppm with treatment T_5 followed by the treatment T_6 (13.9 ppm) while the treatments T_1, T_2, T_3, T_4 recorded 6.3 ppm, 7.2 ppm, 7.9 ppm and 12.8 ppm respectively.

Microbes-fertilizer interaction response

Microbes and chemical fertilizers often do not match for nutrient-use-efficiency. It is still debatable to arrive at conclusive opinion that both can go hand-in-hand, either in citrus nursery or even grown-up orhards. With this objective, five microbes of microbial consortium were tested *in-vitro* for their colony growth against varying concentration of conventionally used inorganic fertilizers. Five micro-organisms (*Aspergillus flavus* MF113270, *Bacillus pseudomycoides* MF113272, *Acinetobacter radioresistens* MF113273, *Micrococcus yunnanensis* MF113274, and *Paenibacillus alvei* MF113275) were further evaluated *in-vitro* against commercially used fertilizers (Urea, potassium dihydrogen phosphate, muriate of potash, iron sulphate, zinc sulphate, manganese sulphate and borax) to study their colony growth response against these fertilizers at six concentrations viz., 0, 100 ppm, 200 ppm, 400 ppm, 800 ppm and 1600 ppm (Table 5)

Response of *Micrococcus yunnanensis*: Maximum colony growth of microbe was observed covering 30.5 mm area at concentration of 200 ppm urea (N-source), thereafter with higher urea concentrations upto 1600 ppm, there was a concurrent reduction in colony growth upto 16.0 mm. While, the same magnitude of response was observed with 200 ppm potassium dihydrogen phosphate (P-source). The colony growth of *Micrococcus yunnanensis* was observed in an area of 21.3 mm followed by reduction in growth upto 18.5mm 1600 ppm concentration. On the other hand, *Micrococcus yunnanensis* responded more favourably with muriat of potash (K-source) upto 800 ppm concentration, displayed increase in colony growth from 12.6 mm at zero concentration to 25.6 mm at 800 ppm but significantly reduced to 22.3 mm at 1600 ppm concentration.

Micronutrients fertilizer sources showed that a microbe triggering the growth response possess some ability to exploit the nutrient source towards its metabolic activity. Ferrous sulphate was observed to increase the colony growth of *Micrococcus yunnanensis* from 14.6 mm at zero concentration to 21.5 mm at 100 ppm, but thereafter subsequent increase in concentration upto 1600 ppm induced consistent reduction in colony growth upto 16.2 mm. The same was response with respect to different concentrations of zinc sulphate (Zn-source), inducing colony growth as 24.0 mm at 100 ppm concentration, significantly higher as 16.3 mm at zero concentration, but higher concentrations continued to inhibit the colony growth upto 1600 ppm.

Response of *Paenibacillus alvei:* Maximum colony growth was observed covering 32.5mm area at concentration of 1600 ppm urea (N-source), increase in colony growth of *Paenibacillus alvei* was observed from 100 ppm onwards. Similarly, the same response was observed with 200 ppm potassium dihydroge phosphate (P-source). The colony growth of *Paenibacillus alvei* was observed maximum in the area of 30 mm at concentration of 1600 ppm. On the other hand, colony growth of *Paenibacillus alvei* was maximum at 100 ppm in an area of 22 mm, but significantly reduced to 200 mm at 800 ppm concentration. Ferrous sulphate was observed to show the maximum colony growth with an area of 25 mm at 1600 ppm concentration.

Similarly, concurrent increase was observed from 18 mm at zero concentration to 28 mm at 1600 ppm concentration of zinc sulphate (Zn-source). In manganese sulphate colony growth in area of 27 mm at concentration of 1600 ppm was observed maximum. The colony growth was more favourable at the concentration of 100 ppm with 24 mm area, but the higher concentration inhibited the colony growth upto 800 ppm. These observations lend steong support in favour of nutrient-microbe synergy (Srivastava *et al.*, 2002; 2008; Srivastava and Malhotra, 2017)).

Response of *Azotobacter chroococcum* : The maximum colony growth was observed covering 12.5 mm area at the concentration of 100 ppm, thereafter followed by the reduced in the response from 200 ppm to 1600 ppm in urea (N-source). In response against potassium dihydrogen phosphate, maximum colony growth was 16.6 mm at concentration of 400 ppm, but it was followed by concurrent decrease at 1600 ppm concentration with 10.3 mm. The response with respect to different concentrations of muriat of potash (K-source) including colony growth as 13.3 at zero concentration, but higher concentration inhibited the colony growth upto 1600 ppm. Similar response was shown by ferrous sulphate, where the maximum colony growth was 15.3 mm at zero concentration followed by concurrent decrease upto 800 ppm and no growth response at 1600 ppm concentration.

Zinc sulphate also showed the same response where the maximum response was observed at zero concentration with 15.6 mm area followed by lateral decrease till 11.3 mm at 400 ppm concentration and no response in 800 and 1600 ppm concentration. Similarly, no growth response was observed in 800 and 1600 ppm concentration of manganese sulphate. The maximum growth response was shown at zero concentration with 13.3 mm area. The last response was observed in borax, where there was no growth response at 400, 800 and 1600 ppm concentration. It showed maximum growth area of 13.6 mm at the zero concentration. These observations strongly suggest that an efficient microbe has some affinity with exogenous nutrient source, where the higher concentrations continued to inhibit the colony growth (Srivastava and Singh, 2008b; 2009b) .

Response of *Aspergillus flavus:* Highest colony growth of microbe was seen covering 41.3 mm at concentration of 1600 ppm of urea (Table 5). Further maximum growth was observed in potassium dihydrogen phosphate treatment of 1600 ppm concentration with the colony growth of 40.3 mm. While, same magnitude of response was observed with 1600 ppm muriate of potash (K-source). Escalating response was again observed in treatment with ferrous sulphate

with 39.0 mm area at the concentration of 1600 ppm. Zinc sulphate showed increasing growth from 27.6 mm of area of zero concentration to 38.6 mm of 1600 ppm concentration. Manganese sulphate also showed mounting response with the area of 40.0 mm at the concentration of 1600 ppm. No response against any of the concentration was noted against borax, it showed the colony growth of 28.3 mm at zero concentration). These observations established the fact that microbial consortium would serve a strong nutrient sink in field. Based on these observations, we put forward another concept, on these observations, we put forward another concept, on these observations, we put forward another concept, these observations, we put forward another concept, known as "rhizosphere hybridization" (Srivastava *et al*., 2015), in addition to better nutrient-use-efficiency (Srivastava and Singh , 2009a ; 2009b). According to this concept , we can introduce the rhizo-microbiome of any healthy crop into the rhizosphere of target crop to have more microbiome of much wider functional diversity coupled with better disease suppressiveness (Srivastava *et al*., 2007).

Microbiome manipulation for rhizosphere engineering

Artificially, the rhizosphere can be modified or reconstruct as per the need the of plant to enhance the physiological efficiency by rhizosphere engineering, rhizosphere hybridization (Cheke *et al*., 2018; Hota *et al*., 2020; Srivastava *et al*., 2025), creating an artificial environment suitable for the plant growth-promoting microorganisms (PGPMs) to surplus a protective layer against the pathogenic microbes (Rhizosphere fortification), or by various agronomic practices. Rhizosphere hybridization is new concept to modify the rhizosphere ecology to create an optimum environment for PGPMs to show the positive effect of plant agronomy (Srivastava *et al*., 2022). The concept of "rhizosphere hybridization" is therefore, advocated to harness the value-added benefit of nutrient-microbe synergy, besides

Table 3: Changes in soil microbial count in response to different treatments in primary nursery (period: 120 days)

Treatment	Changes in Bacterial count over time (x 10^3 cfu g^{-1} soil)						
	10	20 days	40 days	60 days	80 days	100 days	120 days
T_1 (Control)	31	35	39	41	50	56	60
T_2 (*Ar*)	35	38	42	48	53	59	67
T_3 (*Ar+My*)	30	32	37	40	56	65	69
T_4 (Ar+*My +Bp*)	39	38	40	57	60	65	69
T_5 (*Ar+My+Bp+Pa*)	42	45	45	52	57	61	65
T_6(*Ar+My+Bp+Pa +Af*)	37	39	40	47	62	70	73
CD(*P=0.05*)	03	02	02	04	03	03	06
Treatments	Changes in Fungal count over time (x 10^3 cfu g^{-1} soil)						
T_1 (Control)	12	12	14	17	20	22	26
T_2 (*Ar*)	15	16	21	24	27	31	34
T_3 (*Ar+My*)	10	10	13	20	22	26	30
T_4 (Ar+*My +Bp*)	19	17	23	25	29	34	36
T_5 (*Ar+My+Bp+Pa*)	12	15	20	29	32	37	34
T_6(*Ar+My+Bp+Pa +Af*)	14	17	19	24	30	31	35
CD(*P=0.05*)	03	02	04	04	06	0.8	07

Ar,My, Bp, Pa, Af, stand for *Acinetobacter radioresistens* (MF113273), *Micrococcus yunnanensis* (MF113274) *Bacillus pseudomycoides* (MF113272), *Paenibacillus alvei* (MF113275) *and Aspergillus flavus* (MF113270) respectively.
Source : Srivastava and Hu (2019)

Table 4: Changes in individual microbial count in response to different treatments (period: 120 days) in primary nursery.

Treatment	Changes in population of different microbes						
	Initial	20 days	40 days	60 days	80 days	100 days	120 days
	Changes in *Aspergillus flavus* population (x 10^3 cfu g^{-1} soil)						
T_1 (Control)	12	15	17	19	17	21	23
T_2 (*Ar*)	17	13	15	17	25	26	27
T_3 (*Ar+My*)	15	10	10	09	19	25	29
T_4 (Ar+*My* +*Bp*)	10	17	19	21	26	30	32
T_5 (*Ar+My+Bp+Pa*)	12	16	16	25	30	32	32
T_6 (*Ar+My+Bp+Pa +Af*)	17	15	17	20	29	30	34
CD(*P=0.05*)	02	02	02	03	04	04	04
	Changes in *Bacillus pseudomycoides* population (x 10^3 cfu g^{-1} soil)						
T_1 (Control)	09	12	15	21	28	32	32
T_2 (*Ar*)	12	16	19	25	25	35	37
T_3 (*Ar+My*)	10	13	17	31	39	42	43
T_4 (Ar+*My* +*Bp*)	17	19	21	28	32	40	45
T_5 (*Ar+My+Bp+Pa*)	21	25	25	27	35	39	42
T_6 (*Ar+My+Bp+Pa +Af*)	20	22	24	30	37	45	46
CD(*P=0.05*)	02	02	03	03	03	02	04
	Changes in *Paenibacillus alvei* population (x 10^3 cfu g^{-1} soil)						
T_1 (Control)	11	15	17	25	30	38	37
T_2 (*Ar*)	14	17	20	22	32	42	45
T_3 (*Ar+My*)	21	24	21	28	35	39	42
T_4 (Ar+*My* +*Bp*)	20	22	23	32	40	47	52
T_5 (*Ar+My+Bp+Pa*)	19	23	25	29	39	43	45
T_6 (*Ar+My+Bp+Pa +Af*)	17	20	27	32	41	48	50
CD(*P=0.05*)	02	02	02	03	02	03	04
	Changes in *Acinetobacter radioresistens* population (x 10^3 cfu g^{-1} soil)						
T_1 (Control)	05	07	09	15	19	23	25
T_2 (*Ar*)	17	15	17	21	25	27	30
T_3 (*Ar+My*)	12	14	16	28	32	39	43
T_4 (Ar+*My* +*Bp*)	10	18	21	31	39	45	49
T_5 (*Ar+My+Bp+Pa*)	14	17	19	26	35	46	50
T_6 (*Ar+My+Bp+Pa +Af*)	09	12	18	27	37	35	35
CD(*P=0.05*)	02	03	03	04	04	03	05
	Changes in *Micrococcus yunnanensis* population (x 10^3 cfu g^{-1} soil)						
T_1 (Control)	18	18	20	15	19	37	40
T_2 (*Ar*)	20	22	25	21	25	39	43
T_3 (*Ar+My*)	27	20	23	28	32	40	42
T_4 (Ar+*My* +*Bp*)	25	30	32	31	39	51	55
T_5 (*Ar+My+Bp+Pa*)	19	21	27	34	35	42	44
T_6 (*Ar+My+Bp+Pa +Af*)	20	18	21	29	37	39	42
CD(*P=0.05*)	02	03	03	04	04	03	03

Ar,My, Bp, Pa, Af, stand for *Acinetobacter radioresistens* (MF113273), *Micrococcus yunnanensis* (MF113274) *Bacillus pseudomycoides* (MF113272), *Paenibacillus alvei* (MF113275) *and Aspergillus flavus* (MF113270) respectively.
Source: Srivastava and Hu (2019)

providing dynamism to microbial consortium suiting to wide range of perennial fruits .

Our studies on response of different treatments involving rhizosphere soil of three perennial trees viz., *Ficus racemosa* L. (Umber tree), *Ficus benghalensis* L. (Banyan tree), and *Ficus religiosa* L. (Pipal tree) along with rhizosphere soil of healthy and highly productive sweet orange trees in sweet orange buddlings showed differential

Table 5: Fertilizer interaction response (measured by colony growth in mm) under controlled conditions

Fertilizer type	*Micrococcus yunnanensis*						
	Concentration (ppm)						
	Control	100	200	400	800	1600	CD (*P=0.05*)
Urea	14.0	20.5	30.5	21.6	22.0	16.0	1.41
KH_2PO_4	13.3	16.6	21.3	18.5	18.3	18.5	1.64
MOP	12.6	21.0	22.3	23.3	25.6	22.3	0.84
$FeSO_4$	14.6	21.5	20.3	16.0	16.0	16.2	0.94
$ZnSO_4$	16.3	24.0	19.0	20.6	19.0	17.5	1.10
	Paenibacillus alvei						
Urea	20	28	29.5	31	32	32.5	0.50
KH_2PO_4	21	23	24	26.5	25	30	1.20
MoP	19	22	21	21	20	21	1.40
$FeSO_4$	19	21	21	22	23	25	1.50
$ZnSO_4$	18	20	22	24	26	28	1.30
$MnSO_4$	18	21	21	25	24	27	2.10
Borax	20	24	22	22	20	21	1.80
	Acinetobacter radioresistens						
Urea	12	12.5	10.6	11.0	10.0	10.3	NS
KH_2PO_4	11.6	12.3	11.3	16.6	16.0	10.3	1.1
MoP	13.3	11.0	11.3	11.0	-	-	0.70
$FeSO_4$	15.3	13.6	13.3	12.3	10.6	-	1.10
$ZnSO_4$	15.6	13.3	11.0	11.3	-	-	1.14
$MnSO_4$	13.3	11.3	11.3	11.6	-	-	NS
Borax	13.6	12.6	11.6	-	-	-	NS
	Aspergillus flavus						
Urea	34.3	36.0	36.3	38.0	38.3	41.3	1.10
KH_2PO_4	26.3	27.6	30.0	33.6	34.6	40.3	0.80
MoP	28.3	28.6	29.6	34.3	37.6	40.3	1.04
$FeSO_4$	32.3	32.6	34.3	34.6	35.3	39.0	1.20
$ZnSO_4$	27.6	28.6	34.0	28.6	25.3	38.6	0.80
$MnSO_4$	28.3	30.6	33.0	35.3	38.3	40.0	1.10
Borax	28.3	-	-	-	-	-	

These fertilizers are commonly used in citrus fertilization programme

Source: Srivastava *et al.* (2015)

response in terms of agronomic parameters, changes in soil physical properties, and pool of plant available nutrients. However, hybridized rhizosphere of sweet orange and *Ficus racemosa* L. out-smarted the response over other rhizosphere hybridization treatments. These studies lend some support to the fact that inoculation of soil or crops with rhizospheric or endophytic microbes, respectively, can enhance the micronutrient contents in various plant tissues including roots, leaves, and fruits (Cheke *et al.*, 2018).

Conclusion remarks and way forward

These studies hence established that microbial consortium can be effectively retrofitted replacing conventionally used chemical fertilizers in nursery, considering very low nutrient requirement of such juvenile citrus plants (Kohli *et al.*, 1998; Srivastava , 2023; 2025). There is every possibility, we can further rationalize the use of function specific microbes as per growth stages of nursery plants with use of biochars (Agegnehu *et al.*, 2017; Mousavi *et al.*, 2023), mycorrhizal-mediated microbial consortium (Wu *et al.*, 2013; 2017), crop phenology-based fertigation (Shirgure *et al.*, 2001; Srivastava *et al.*, 2003) , speciality fertilizers (Srivastava and Pandey , 2021), orchard efficiency (Srivastava *et al.*, 2008a) , regular flowering (Srivastava *et al.*, 2000) and site-specific nutrient management (Srivastava *et al.*, 2006). However, no distinction in morphological or physiological growth behavior exists in nursery plants, right from growth in primary nursery to secondary nursery. And, morphologically, it is very difficult to identify such shifts in growth stages (Srivastava and Singh 2008a; 2008b).

Our studies also establish and advocate following other impotent issues : i. microbes can replace nutrients requirement of citrus nursery, considering absymally low nutrient requirement of nursery plants ii. microbial consortium is a far better choice than individual microbe(s); iii. liquid formulation of microbes is better than substrate-based inoculants, either individual microbe or consortium of microbes; iv. the quantity of microbial broth needs to be standardized for containerized citrus nursery versus field nursery; v. inoculation of citrus nursery plants with microbial consortium needs to be standardized depending upon substrates used (solarized soil versus soilless medium) ; vi. the treatment of microbial consortium (5ml/plant) reduced the rate of mortality of citrus nursery plants to bare minimum, once transplanted in new orchard site. This is an excellent piece of information, otherwise orchardists are fed up with high rate of mortality of citrus nursery plants; vii. treatment with microbial consortium provided an additional plant immune on account of biopriming effect of microbes, which eventually aided in far better withdrawal of nutrients from soil and ensured better plant health in ultimate terms; viii.the treatment with microbial inoculants individually or as microbial consortium has a strong promise to be integrated with fertigation to evolve a new concept called "biofertigation" for exclusively citrus nursery and ix. the use of microbial inoculants can be tailored in citrus nursery, depending upon contrasting growth stages (initiation, establishment and growth stages, though these stages are poorly differentiated and quite inter-changeable). With these concluding remarks, microbe s-mediate organic citrus nursery would pave away forward to more sustainable citrus industry.

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Next-generation onion (*Allium cepa*) breeding using molecular markers: progress and prospects--a review

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Abstract

Onion (*Allium cepa* L.) is an indispensable vegetable crop throughout the globe. It is a member of family Amaryllidaceae (formerly Alliaceae family) and is characterized by 16 somatic chromosomes and a massive 16.4 Gbp genome. Despite its economic significance, the pace of genetic improvement has historically been hindered by the complex and biennial life cycle, photosensitivity, high inbreeding depression and a lack of sufficient genomic resources. The shift from low-throughput markers like RAPD and RFLP to high-resolution and reliable marker systems like SSRs and SNPs has facilitated the construction of saturated linkage maps, leading to identification of QTLs for various traits like resistance to downy mildew, purple blotch, anthracnose and thrips, apart from tolerance to drought stress. Development of specialized mapping populations like doubled haploids has also enabled rapid fixation of heterozygosity and led to identification of responsible loci for different nutritional and bulb-quality traits. Deployment of molecular markers like *PsaO*, *AcPMS1* for rapid identification of Cytoplasmic-Genetic Male Sterility (CGMS) systems, is paving the way for hybrid seed production in a cost-effective manner. Thus, this review synthesizes the evolution of onion breeding, transitioning from conventional phenotypic selection to modern genomic-assisted improvement to overcome the genome complexities for accelerated development of high-yielding, multi-stress-resistant onion cultivars.

Key words: Abiotic stress, *Allium*, Biotic stress, Diseases, Marker-Assisted Selection, Pests, Quality

Onion (*Allium cepa* L.), a member of family Amaryllidaceae (formerly Alliaceae), is characterized by a diploid chromosome complement of 2n = 2x = 16 (Tripathi *et al.,* 2017; Mahala *et al.,* 2024). The primary center of origin is localized to Central Asia-encompassing modern-day Uzbekistan, Turkmenistan, Tajikistan, Afghanistan, Iran and Pakistan (Tripathi and Lawande, 2019). Found growing wild in these regions, *Allium vavilovii* has been identified as the closest wild progenitor to cultivated onion, while Mediterranean basin is recognized as the secondary center of origin.

Globally, onion cultivation is spanned in 5.719 million ha area, yielding a total output of 108.260 million tonnes. Production is heavily concentrated in Asia, which accounts for 64.70% of global volume, followed by Africa (15.70%), Europe (9.70%), and the USA (9.70%). India has emerged as the largest producer of onion followed by China, the USA, Egypt and Bangladesh (FAO STAT, 2024). India produces about 24.266 million tonnes of fresh bulbs from 1.54 million ha area (Anonymous, 2024). It is regarded as the most important export vegetable, contributing to 1.3 million tonnes worth ₹ 2,107.14 crore from India (NHRDF, 2021).

Since the early era of domestication of onion, human being invariably conducted selection leading to the development of different types with respect to bulb shape, size, colour, pungency, photoperiodic response etc. However, despite presence of appreciable variability, speed of onion improvement is not at the pace of other monocot taxa (McCallum, 2007, Varshney *et al.,* 2012). Systematic onion breeding stated with mass selection during the 19th century, followed by discovery of cytoplasmic male sterility (CMS), paving the way for development of F_1 hybrids (Brewster, 2008; McCallum *et al*., 2008). Presently, F_1 hybrids predominate in long-day season, whereas in short-day conditions, open-pollinated varieties predominate (Brewster, 2008).

The onion breeders following conventional breeding methods often face difficulties because of its high levels of cross-pollination and inbreeding depression, biennial life-cycle, photosensitivity and short seed viability. Genome sequencing poses challenge because of its large 16.4 giga base pairs (Gbp) genome size. Thus, there is an urgent need for deployment of genomic tools to facilitate rapid genetic improvement for yield, quality traits and multi-stress resistance in different genetic backgrounds.

Phenotypic selection of onion often leads to biased results due to the substantial influence of environment on trait expression. Moreover, it is also difficult to improve the traits which have low heritability. Marker-Assisted Selection (MAS) using tightly-linked markers, proves efficient in such cases. The MAS provide additional advantages like high accuracy, rapidity, early trait detection etc. Thus, this review collects the available

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information on progress made by identification and deployment of genomic resources for onion improvement and its prospects.

Genetic Resources

Availability of diverse germplasm is the most crucial factor for a successful breeding programme (Glaszmann *et al.,* 2010). Growing of onion in different agro-ecological zones over centuries led to development of a wide range of landraces, harbouring tremendous genetic variation (Brewster, 2008). However in the late 1980s, development of F_1 hybrids resulted in genetic erosion of those landraces in different countries. However, presently, a huge number of onion accessions are being maintained in different gene banks in the world (Table 1).

Table 1: Major onion gene banks in the world along with their germplasm holdings.

Institute	Location	Number of *Allium* accessions
European Cooperative Programme for Plant Genetic Resources	Germany	14400
Directorate of Onion and Garlic Research	India	2050
N.I. Vavilov Research Institute	Russia	1888
Warwick Crop Center	United Kingdom	1755
National Institute of Agro-biological Sciences	Japan	1352
US Department of Agriculture	USA	1304
Royal Botanic Gardens	United Kingdom	1100
AVRDC – The World Vegetable Center	Taiwan	1082

Despite the wide distribution, the knowledge of genome, population structure and genetic architecture are limited in onion (McCallum, 2007), necessitating genetic resource mining and accelerated genetic analysis (Baldwin *et al.,* 2012a). The genome of onion is very large (~16.4 Gbp) and very few ESTs are available in public domain (McCallum *et al.*, 2001; Kuhl *et al.*, 2004). Forming a partial onion BAC library, Jakše *et al.* (2008) reported genome composition, gene structure and very low gene density of one gene per 168 kb genome. The genomic data and genetic maps of onion and other economically important *Allium* species are available from an online genomic database named as *Allium Map* (https://allium.scinet.org.nz/) (McCallum *et al.*, 2012).

Mapping populations

Predominantly, early generation inbreds are used for development of the mapping populations in onion. The residual heterozygosity in such parental inbreds complicates both marker development and sequence analysis (Baldwin *et al.,* 2012b). In general, F_2 population is most used for molecular mapping owing to difficulties to maintain fertility and plant vigour, while developing Recombinant Inbred Lines (RILs). To overcome this, doubled haploid (DH) onion lines have been developed in both short- and long-day onions (Bohanec, 2009; Baldwin *et al.,* 2012b; Duangjit *et al.,* 2013) for mapping of quantitative and qualitative traits. However, major drawback of doubled haploid accessions is poor seed setting (Bohanec, 2002). In this context, Alan *et al.* (2003 and 2004) developed highly fertile doubled haploid lines from long-day US onion varieties, paving the foundation for better insights into onion genetics and genomics.

Genetic maps/ linkage maps

Linkage Map is a schematic representation of the relative locations of genetic markers present on chromosome of an organism as determined from the frequency of recombination between the marker pairs. The first genetic map of onion consisting of 12 linkage groups was developed by Bradeen and Havey (1995) by using 116 RFLP and RAPD markers in a population developed from BYG15-23 × AC43. To date, this map is regarded as a key reference genetic map of onion. Furthermore, Martin *et al.* (2005) placed 100 additional markers to the same intra-specific map for a better saturation with 14 linkage groups. This map predominantly contained dominant RFLP markers which implies that the large genome size of onion is mainly because of high levels of gene duplication.

Marker systems

RAPD

The RAPD (Randomly Amplified Polymorphic DNA) markers use 10 base long arbitrary primers (decamers) which amplify random segments of genomic DNA in a Polymerase Chain Reaction (PCR). Being random, this marker does not require any sequence information. RAPD had been deployed for characterization of onion germplasm comprising of both wild and cultivated genotypes by Maniruzzaman *et al.* (2010) and Sudha *et al.* (2019).

ISSR

The Inter Simple Sequence Repeat (ISSR) technique amplifies the genomic region falling between two Simple Sequence Repeats (SSRs) by using both anchored and non-anchored primers (Ziętkiewicz *et al.,* 1994). Smolik *et al.* (2007) analyzed genetic diversity of onion genotypes belonging to six different species using 16 ISSR markers. Mukherjee *et al.* (2013) used both RAPD and ISSR for diversity analysis of five *Allium* species and found ISSR to

Table 2: Linkage maps developed in onion along with the marker system utilized

Population type	Population derived from	Marker system employed	Reference(s)
F_2	NW-001 × NW-002	SNP	Jo *et al.* (2017)
F_2	BYG15-23 × AC43	InDel, SNP, SSR, RFLP	Martin *et al.* (2005)
F_2	BYG15-23 × AC43	RFLP, SSR and SNP	King *et al.* (1998); Duangjit *et al.* (2013)
F_2	*Allium cepa* × *Alliumroylei*	AFLP and SSR	Van Heusden *et al.* (2000), McCallum *et al.* (2012)
F_2	W202A × Texas Grano	SSR	McCallum *et al.* (2006, 2007)
F_2	Nasik Red × CUDH2150	SSR and SNP	Baldwin *et al.* (2012b)
F_2	OH1 × 5225	SSR and SNP	Duangjit *et al.* (2013)
F_2	DehyA × B5351C	SNP	Damon and Havey (2014)
F_2	*Allium cepa* × *A. fistulosum*	SSR, InDel, STS and RAPD	Tsukazaki *et al.* (2012)

be more polymorphic between *Allium cepa* and *A. sativum,* while RAPD markers showed better polymorphism within *A. porrum.* In overall, RAPD markers revealed more intraspecific diversity than ISSRs, whereas ISSRs showed more interspecific diversity than RAPD, proving its potential for diversity analysis.

SRAP

The Sequence Related Amplification Polymorphism (SRAP) is a simple marker designed for the amplification of Open Reading Frame (ORF). SRAP primers are generally 17-18 nucleotides long and consist of core sequences (13-14 base pairs) and nucleotides 'CCGG' in the forward primer and 'AATT' in the reverse primer.

RFLP

The Restriction Fragment Length Polymorphism (RFLP) involves a single restriction enzyme which generates fragments of different lengths from the same genomic region of different individuals/strains/lines or species, depending upon the presence or absence of recognition sites. Bark and Havey (1995) deployed this marker system to characterize onion inbred lines.

AFLP

The Amplified Fragment Length Polymorphism (AFLP) was developed by Zabeau and Vos (1993). This technique is a combination of restriction digestion and PCR. Ohara *et al.* (2005) constructed the first genetic linkage map of Japanese bunching onion (*Alliums fistulosum*) using 149 AFLP markers.

SSR

The term *Simple Sequence Repeat* (SSRs) was originally coined by Lilt and Luty (1989) to describe simple sequence fragments generated in PCR. This is also known as Short Tandem Repeat (STR) and Simple Sequence Length Polymorphism (SSLP). The multi-allelic nature and PCR-based detection of the SSR markers makes them more breeder friendly than the SNPs (Pal *et al.,* 2020). More than a hundred SSR markers were developed by Tsukazaki *et al.* (2008) from size-fractionated genomic DNA libraries. SSR makers have provided the greatest insights into onion diversity analysis (Khosa *et al.,* 2014). Mitrová *et al.* (2015) established a panel of 15 easy to score SSR markers which easily differentiated 16 commercial onion cultivars of Czech Republic, making them potential for future diversity analysis studies. Anwar *et al.* (2017) employed 16 SSR and three ISSR markers for diversity analysis in *Allium cepa* L., *A. sativum* L. and *A. kurrat* L. accessions. Ivchenko *et al.* (2017) used six SSRs for identification of onion varieties. While studying the genetic diversity of 16 onion genotypes, using RAPD, SSR and ISSR markers Kesraliker *et al.* (2017), concluded ISSR and SSR to be more reliable over RAPD. Singh *et al.* (2021) and Gupta *et al.* (2022) also deployed microsatellite markers for germplasm characterization and emphasized on these markers over the others for better reproducibility and preciseness.

Single Nucleotide Polymorphism (SNP)

Single Nucleotide Polymorphism (SNPs) are one of the most reliable marker systems owing to its preciseness and reproducibility. In recent times, the marker system is gaining importance with the reduction in the sequencing costs. This marker had been deployed for characterization of commercial cultivars by Labate *et al.* (2020), Lee *et al.* (2021) and Jeon *et al.* (2022).

QTL Mapping and Marker Assisted Breeding

Quantitative traits show continuous variation due to their polygenic inheritance and environmental influences. Polygenes produce small individual and cumulative effects on the phenotype. A quantitative trait locus (QTL) is a section of DNA which correlates with the variation of quantitative traits. Thus, QTL mapping employs an algorithm for detection of an association between a phenotypic trait and genetic marker(s).

Disease resistance

Downy mildew, caused by *Peronospora destructor*, is a major yield limiting factor, especially during the *rabi* season. The resistance is conditioned by two dominant genes. Introgression of the resistant genes have been made from *Allium royeli* with the help of tightly-linked AFLP markers are available (Scholten *et al.,* 2007; Khosa *et al.*, 2016). Eidlin *et. al.* (2021) carried out marker-assisted selection using DMR1 marker, linked to the resistant gene '*Pd*', to develop a maintainer line with downy mildew resistance.

Genetics of resistance to *Fusarium* basal rot varies from monogenic to polygenic, depending upon the resistant sources employed (Cramer, 2000, Galvan *et al.*, 2008, Taylor *et al.,* 2013). The resistance can also be precisely introduced in susceptible genotypes through CRISPR/Cas mediated genome editing (Pal *et al.*, 2025). Incomplete resistance (tolerance) to white rot (Hovius *et al.*, 2004) and resistance to pink root (Esfahani and Pour, 2008), *Stemphyllum* blight (Anitha *et al.,* 2011), black mold (Kamal *et al.,* 2012) and Iris Yellow Spot Virus (Bag *et al.,* 2014) has also been identified in onion or other alliums, which are to be deployed in future breeding programmes.

Among biotic stresses, purple blotch causes havoc crop loss ranging from 2.5 to 97% both in bulb and seed crops throughout the globe (Schwartz *et al.,* 2005; Kareem *et al.,* 2012; Tripathi *et al.,* 2013; Nanda *et al.,* 2016; Priya *et al.*, 2016). Chand *et al.* (2018) identified *ApR1* governing resistance to this disease in a population derived from Arka Kalyan (resistant parent) and Agrifound Rose (susceptible parent). They further identified one SSR, *i.e.*, AcSSR7 and one STS marker, *i.e.*, ApR-450 linked to the *ApR1* gene at 1.3 and 1.1 centi Morgan genetic distance, respectively. Recently, KASP based SNP (ApRsnip14, ApRsnip23), SSR (AcSSR7) and SRAP marker (ApR-450) have also been developed for marker-assisted introgression (Sahoo *et al.*, 2023) of purple blotch resistance.

Anthracnose disease, caused by *Colletotrichum gloeosporoides*, also poses serious threat to onion cultivation worldwide. Jayaswall *et al.* (2025) shortlisted a total of 131 differentially expressed genes, belonging to gene classes like Mitogen-activated protein kinases, *WRKY* and *MYB* transcription factors, R genes and transcriptional activators, putatively associated with resistance to anthracnose in wild species of onion. Scholten et al. (2016) identified a QTL (*Bs1*) on chromosome 6 for resistance to leaf blight disease caused by *Botrytis squamosa*. Kim *et al.* (2021) identified SCAR-OPAN-1 and SNP-3 HRM markers linked to gray mold disease resistance.

Epicuticular wax for insect resistance

Insects with piercing and sucking type mouth parts recognize their host plants by sensing the organic constituents of epicuticular waxes (Diaz-Montano *et al.,* 2011). Wide natural variation for the amount and types of epicuticular wax has been reported and reduced amount of epicuticular wax (glossy foliage) have been associated with thrips resistance (non-preference) in onion (Bag *et al.,* 2014; Damon and Havey, 2014). Recessive inheritance of reduced epicuticular waxes and found two QTLs each on chromosome 2 and chromosome 5, governing the production of hentriacontanone-16 (primary wax on leaves) and for production of several primary alcohols, respectively (Damon and Havey, 2014). SNP markers are being identified for marker-assisted selection for this trait (Bag *et al.,* 2014; Damon and Havey, 2014).

Drought tolerance

Caldwell *et al.* (2003) investigated soil moisture potential, leaf water potential, growth, photosynthesis, stomatal conductance, leaf transpiration, water use efficiency, membrane function, nature and accumulation of quaternary ammonium compounds in 21 days old onion seedlings which were subjected to various drought regimes by withholding irrigation. They concluded that the onion seedlings were able to tolerate drought by increasing water use efficiency. Though QTL mapping has not been followed in onion for drought tolerance, the related species, *viz. Allium fistulosum* and *A. munzii* and genotypes *viz*., Arka Kalyan, MST 42 and MST 46 have been reported to be drought-tolerant (Singh, 2010), necessitating their deployment in QTL mapping.

Bulb pigmentation

A total of 54 types of flavanoids determine the bulb colour by their relative proportion (Slimestad *et al.,* 2007). However, the major colour compounds are anthocyanin (red), flavonols (pale yellow) and chalcones (bright yellow) (Schwinn *et al.,* 2016). Clarke *et al.* (1944) reported three pairs of genes, *viz*. C, R and I, being involved in the development of pigment in the onion bulbs. *C-c* is a basic color factor and dominant C is necessary to produce any pigment. Consequently, all *cc* plants produce white bulbs. Dominant *R* gene in the presence of *C* produces red pigment and recessive *r* allele is responsible for the production of yellow pigment. The gene, *I* is inhibitory to the coloured bulbs and presence of dominant allele of *I* results in white bulbs. El-Shafie and Davis (1967) reported five genes *viz*., *I, C, G, L, R* (all with two alleles each) responsible for onion bulb colour. Khar *et al.* (2008) developed three different families segregating for bulb colours and identified a SSR marker on chromosome 6 linked to C locus. In cross B2246×B11159, they further reported that red bulbs versus yellow bulbs were controlled by DFR and a locus (*L2*) linked at 6.3 cM to ANS (anthocyanidin synthase). Hence, authors proposed that onions with yellow bulbs were independently selected for numerous times and thus yellow genotypes carry independent mutations in structural or regulatory genes controlling the production of red bulb colour. Earlier, Kim *et al.* (2004) identified a critical mutation within the chalcone isomerase (CHI) gene that reduces the amount of quercetin, giving rise to gold-coloured bulbs. Further, Kim *et al.* (2005) reported that the inactivation of *DFR* was responsible for colour difference between yellow and red onions and two recessive alleles of *ANS* gene were responsible for the development of pink bulb colour. Later, Park *et al.* (2013) designed functional CAPS marker for differentiating two *DFR-A* alleles *viz.*, DFR-A^{PS} (present in yellow onion) and DFR-A^{DEL} (present in red onion). Tsukazaki *et al.* (2012) developed a linkage map from a population derived from *Allium cepa* and *Allium fistulosum* consisting of 11 linkage groups and total marker coverage of 1040 cM. A QTL for bulb pigmentation was reported on chromosome 7.

Pungency

Bulb pungency is an important determinant of end use of onion bulbs and consumer acceptance. McCallum *et al.* (2007) identified a major QTL for pungency on chromosome 3 by using a $F_{2:3}$ mapping population derived from W202A (pungent type) × Texas Grano 438 (sweet type). Marker assisted transfer of the QTL could be followed for improving pungency into non-pungent genotypes.

Tearless onion

Lachrymatory factor synthase (*LFS*) gene catalyzes the production of the lachrymatory factor in onion bulbs. Masamura *et al.* (2012) mapped a candidate *LFS* gene on chromosome 5 of onion using a complete set of *A. fistulosum*–shallot (*A. cepa* L. *aggregatum* group) monosomic alien addition lines. Eady *et al.* (2008) produced tearless onion by RNAi mediated suppression of *LFS* gene which prevented the conversion of isoalliin into thiosulfinates.

Kato *et al.* (2016) developed non-pungent onions by irradiating seeds with neon-ion at 20 Gy. The level of lachrymatory factor production of the selected bulbs in M_4 generation had 7.5 times lower than those of normal onion, confirmed by the lower mRNA production for the gene coding for Allinase enzyme.

Bulb quality and nutritional traits

Substantial genetic variation for fructo-oligosaccharides, quercetin, epigallocatechin gallate and epicatechin gallate, total and essential amino acids, carbohydrates, total soluble solids, trisulphinates and total antioxidant activity occurs among onion cultivars (Insani *et al.* 2016). The parameters *viz.,* bulb colour, firmness, number of scales, number of growing points, neck thickness, total soluble solids (TSS), pungency and antioxidants are the most important quality determinants in onion among the consumer (Brewster 2008; Goldman 2011. Dry matter, TSS and pungency are positively correlated and show moderate to high heritability, indicating the possibility of simultaneous improvement through selection. However, development of less pungent onion with high dry matter becomes critical (Galamarini *et al.*, 2001; Mallor *et al.,* 2011).

Galmarini *et al.* (2001) used the genetic map of BYG15-23 × AC43 to carry out mapping of loci for pungency, dry matter and TSS. One gene (acid invertase-API89) on chromosome 3 and another gene (phloem unloading sucrose transporter- API66) on chromosome 5, both being responsible for high TSS and dry matter accumulation was reported. Furthermore, the acid invertase gene also elevated pungency, which suggests a possible pleiotropic effect of the gene on bulb composition. Havey *et al.* (2004) conducted further QTL analysis in the same genetic map with extensive analysis of bulb carbohydrates and again reported significant effects of the acid invertase region of chromosome 3 on bulb sucrose content. Three loci each on chromosomes 3,5 and 8 have been identified in a population of *A. cepa × A. royeli,* regulating accumulation of non-structural carbohydrates like fructose, fructan, glucose and sucrose (McCallum *et al.,* 2006; Raines *et al.,* 2009), of which *Frc* locus on chromosome 8 is the major determinant for fructan content. This result was later confirmed by Yaguchi *et al.* (2008) by using *Allium fistulosum* and *A. cepa* var. *aggregatum* monosomic alien addition lines. Although two markers *viz.*, ACM033 and ACABE58 have been reported to be linked with the *Frc* locus on chromosome 8, but more tightly linked markers are urgently needed for marker-assisted selection.

Male sterility

Male sterility is the most important genetically controlled pollination control mechanism economizing the hybrid seed production in onion. Cytoplasmic-genetic male sterility (CGMS) system, owing to its ease of maintenance and restoration through the *Ms* locus, is preferred in onion over the CMS. The *Ms* locus codes for penta-trico-peptide repeat (PPR) proteins, leading to restoration of fertility.

The CGMS system has been reported in different cytoplasmic backgrounds of onion like CMS-S (Jones and Emsweller 1936), CMS-T (Berninger 1965) and CMS-T like or CMS-Y cytoplasm (Kim 2014). The fertility restoration of CMS-S cytoplasm gets restored by the fertility restorer, *Ms* locus (Jones and Clarke 1943). However, restoration of fertility in CMS-T cytoplasm is governed by three genes i.e., two complementary genes and an independent gene (Schweisguth 1973) while in CMS-T like cytoplasm, it is governed by a single nuclear gene (Kim and Kim 2019). Go¨kce *et al*. (2002) and Martin *et al.* (2005), carried out marker-assisted selection using a codominant and PCR-based marker. Manjunathgowda and Selvakumar (2021) validated PsaO marker for its co-dominance and tight linkage to restorer of fertility, *Ms* locus. Thus, the PsaO is suitable rapid identification of desirable recombinants with respect to male fertility while handling segregating generations. Saini et al. (2015) utilized 5'cob and *orfA50*1 markers, originally reported by Sato et al. (1998) and Engeleke et al. (2003), to distinguish between the male sterile and male fertile cytoplasm of accessions in three different populations. Further, upon testing of two markers *viz*., OPT and PsaO-linked to *Ms* locus, *OPT* was proven better over the PsaO. Khar et al. (2022) used accD and MKFR markers for *cytotype* identification and AcPMS1 and AcSKP1 markers (Huo *et al.* 1015; Kim *et al.* 2015) to identify *Ms* locus in a diverse germplasm panel. While both accD and MKFR were equally potent for identification of cytoplasm, AcPMS1 was found more reliable to identify the *Ms* locus compared to AcSKP1. Manjunathagowda (2025) utilized two sets of PCR-based molecular markers linked to orf725, originally identified by Kim *et al.* (2009) and reported two short day and Indian accession *viz.* DOGR-MG-2 and DOGR-MG-70 carrying CMS-S cytoplasm.

CONCLUSION AND FUTURE PROSPECTS

Onion is an important vegetable cum spice crop with great impact on the economy across the globe. Though several attempts and significant progress have been made for the genetic improvement, the yield potential is many-a-times affected adversely due to the frequent occurrence of several biotic and abiotic stresses. Moreover, there is differences in the consumer appealing traits for specific end use. Thus, there is an urgent need to develop trait-specific varieties with multiple disease resistance, wider adaptability for varied agro-climatic conditions and suitability for various end use including processing into different products.

Keeping in view the large genome size of onion, there is an urgent need to develop more polymorphic and codominant markers to study genetic variation among the accessions. To meet the increasing demand, development of high-yielding varieties and hybrids in a short span can be achieved by through marker-assisted selection. For this, identification of tightly linked molecular markers and/or identification of candidate genes and functional markers within the gene(s) is urgently needed. Candidate gene association mapping for fructan content and bulb pungency is also required. Mainkar et al. (2023) standardized the genome editing protocol in onion and edited the *AcPDS* gene, paving the way for precise modification of other desirable genes. Keeping in view the dynamic and diverse market segments, less pungent and tearless onion need to be developed through genome editing, apart from development of multi-stress resistant varieties.

DECLARATION

The authors declare that they do not have any conflict of interest.

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Integrated use of plant nutrients for higher quality yield of ginger *(Zingiber officinale)* in midhills of Nepal

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Abstract

A study was carried out to assess the effectiveness of integrated plant nutrient management (IPNM) on ginger *(Zingiber officinale* Roscoe) yield and quality in two midhill sites: Bhotechaur (Sindhupalchok) and Anekot (Kavre), during 2023 and 2024, in a randomized complete block design with eight treatments combining several types of *in- situ* composts, chemical fertilizers, and micronutrients (Zn and B). Nutrient management treatments had no significant effect on final plant population (p>0.05) or rhizome yield (p>0.05) at either sites. However, notable yield differences were observed numerically. T_7 (multimix compost @ 20 tonnes/ha) produced the highest rhizome yield at both sites (24.86 tonnes/ ha at Anekot and 49.32 tonnes/ ha at Bhotechaur), while T_1 (control) and T_2 (100% NPK + Zn/B) were consistently the lowest performers. Quality parameters (crude fiber and oleoresin) were significantly influenced by treatments, though effects were highly site specific. At Anekot, T_4 (50% NPK + fine compost + Zn/B) produced the lowest crude fiber content (13.43%), while T_3 (50% NPK + coarse compost + Zn/B) yielded the highest oleoresin content (0.61%). Integrated nutrient management significantly improved ginger quality by reducing crude fiber and improving oleoresin content, despite inconsistent responses to yield across locations. For mid hill regions of Nepal, application of 50% recommended NPK combined with 10 tonnes/ha quality compost supplemented with zinc and boron is recommended for optimal quality and yield.

Key Words: Crude fiber, IPNM, Oleoresin, Rhizome, Quality, Compost, Midhills, Nepal

Ginger (*Zingiber officinale* Roscoe) is a high-value spice crop of significant economic importance in global agriculture. Apart from culinary applications, ginger is also valued for its bioactive compounds, including gingerols, shogaols, and essential oils, contributing to its widespread use in traditional medicine, pharmaceutical formulations, and functional food products (Ali *et al*., 2024). Nepal is a major global producer of ginger, ranking 4th, with a production of 3,09,533 tonnes in 2023 (FAO, 2023), thereby earning foreign exchange over 1.22 million in 2023 by exporting over 23 thousand tonnes of fresh as well as crushed/ ground ginger (MOALD, 2023). Nepal's midhill regions, ranging from 900 to 2,000 m in altitude characterized by warm and humid weather coupled with well-distributed rainfall, provide conducive agroclimatic conditions for ginger production (Joshi and Khanal, 2021).

Despite this favorable resource base and huge export potential, ginger productivity in Nepal however remains low, averaging about 12-13 tonnes/ha against a potential yield of about 24-25 tonnes/ha (Khatiwada and Yadav, 2022). One of the fundamental challenges limiting crop productivity in Nepal's midhill regions is critically low soil nutrient stocks, with widespread deficiencies in nitrogen, phosphorus, and several micronutrients (Shrestha *et al*., 2018). IPNM combining the application of organic nutrient sources with chemical fertilizers has proved highly effective in improving ginger growth, yield, soil physico-chemical properties, economic returns, and the benefit: cost ratio of ginger production (Adekiya *et al*., 2020; Yanthan *et al*., 2010). These studies emphasize the judicious combination of organic, inorganic, and biological nutrient sources in balanced combinations tailored to specific edaphic, climatic, and agronomic conditions to maintain soil fertility, improve nutrient-use efficiency, and enhance crop productivity (Kumar *et al*., 2024).

In Nepal, studies evaluating the impacts of IPNM on ginger are limited, and the previous efforts to enhance ginger productivity have often focused on singular approaches, such as the application of chemical fertilizers or organic manures in isolation. While specific nutrient recommendations exist for ginger, the practical limitations faced by Nepalese farmers, particularly regarding the unavailability and cost of chemical fertilizers, render these solutions partially effective or unsustainable. On the other hand, *in-situ* composting, which involves management of agricultural waste on the site of production, is considered an economic method of nutrient recycling, as it can reduce costs involved in collection and transportation (Muzamil

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et al., 2022). *In-situ* composting could offer significant benefits for resource-constrained smallholder farmers in Nepal's mid hills by reducing input costs while promoting sustainable agricultural practices.

Evidence on the combined effects of *in-situ* composting, chemical fertilizers, and micronutrients in the midhills of Nepal is limited, particularly regarding their impact on both yield and quality parameters. Therefore, experiment was conducted to evaluate the effects of IPNM by combining *in-situ* compost with chemical fertilizers and micronutrients on yield and quality at two distinct midhill locations, representing Bhotechaur (Sindhupalchok) and Anekot (Kavre) of Bagmati province, Nepal.

MATERIALS AND METHODS

Experimental design

Field experiments were conducted during 2023 and 2024 at two midhill locations in Nepal, viz. Bhotechaur, Sindhupalchok (1380 masl) and Anekot, Kavre, (1255 masl), both known for their favorable conditions for ginger production. The experiments were conducted using the native ginger landrace, widely grown and well-adapted to these mid-hill environments. The soil parameters of the experimental sites were: 4.68 pH, 2.05 % organic matter , 0.20% total N , 20 .0 kgha^{-1} P_2O_5 and 221.82 kgha^{-1} K_2O at Bhotechaur (Sindhupalchok). Whereas at Anekot (Kavre), these properties were observed as: 5.8 pH, 2.70 % organic matter, 0.13% total N, 29.7 kgha^{-1} P_2O_5, and 125.38 kgha^{-1} K_2O. As many as eight IPNM-treatments with three replications at each site were tested under randomized complete block designs. Each plot measured 9 m^2, accommodating 100 ginger plants with a spacing of 30 cm × 30 cm apart. Standard agronomic practices for ginger cultivation were followed uniformly across all treatments.

Treatments formulations

Compost formulation: Three different quality compost formulations were prepared at the National Commercial Crop Research Centre as part of IPNM:

i. Quality compost (Fine-C1), prepared by thoroughly mixing 800 kg fresh cow dung, 50 kg mustard cake, and 150 kg wood ash. The mixture was finely ground, sieved, and well packaged before use.

ii. Quality compost (Course-C2), prepared in two layers. Layer 1 consisted of 150 kg of *Sesbania bispinosa* (green biomass), 10 kg of sawdust, 50 kg of wood ash, 50 kg of cow dung, and 10 kg of mustard cake. Layer 2 consisted of 10 kg of sawdust, 150 kg of cow dung, 10 kg of mustard cake, 1 drum of paddy husk and 100 kg of wood ash.

iii. Quality compost (Multimix-C3), formulated by mixing 100 kg each of cow dung, Sesbania green leaves, and Tithonia, along with 30 kg mustard cake, 50 kg wood ash, and 10 kg sawdust to enhance nutrient diversity and microbial activity.

Fertilizer sources

Chemical nutrient supplementation was provided using standard chemical fertilizers. Urea and Diammonium phosphate (DAP) served as sources of nitrogen (N) and phosphorus (P_2O_5), and muriate of potash (MOP) was used for potassium (K_2O). The required micronutrients were supplied through zinc carbonate (Zn-52%) and boric acid (B-17.5%).

Treatment combinations

The experiment included eight treatment combinations integrating chemical fertilizers, 3 compost types, and micronutrients (Zn and B) as : T_1(control as no input),T_2 (recommended dose of NPK at 100:50:75 kgha^{-1} + Zn at 5 kgha^{-1}), T_3 (NPK at 50:25:37.5 kgha^{-1} + Zn/B at 5 kgha^{-1} + coarse compost at 10 tonsha^{-1}), T_4 (NPK at 50:25:37.5 kgha^{-1} + Zn/B at 5 kgha^{-1} + fine compost at 10.0 tonsha^{-1}) ,T_5 (quality compost–1 as fine at 20.0 tonsha^{-1}), T_6 (quality compost–2 a Coarse at 20.0 tonsha^{-1}),T_7 (quality compost–3 as multimix at 20.0 tonsha^{-1}) , and T_8 (fine, coarse, and multimix compost at10.0 tons ha^{-1} each + Zn/B at 5 kg ha^{-1}).

Data collection and analysis

Observations were recorded on plant population, yield, and quality parameters of ginger (crude fiber and Oleoresin content), following standard procedures. Data were collected from selected plants from each plot to ensure representative sampling. At harvest, data on fresh rhizome weight per plant, rhizome yield per plot and total rhizome yield (tonsha^{-1}) were recorded.

For quality parameters, representative rhizome samples were analyzed for oleoresin and crude fiber content at the National Food Research Centre, Khumaltar, Lalitpur. Crude fiber content was estimated by acid-alkali digestion and oleoresin by high-performance liquid chromatography (FSSAI, 2016).

All statistical analyses were performed with software R (version 4.3.1) using the packages tidyverse, ggplot2, agricolae, and patchwork. Treatment effects were tested by ANOVA, and mean separation was carried out using Fisher's LSD at $\alpha = 0.05$.

RESULTS AND DISCUSSION

Effect on yield and yield- attributing characters

Plant population: Nutrient management treatments had no significant effect on final plant population of ginger, either within individual sites (Figs. 2 and 3) or data pooled across sites (Fig. 1). In the pooled analysis, the experimental site had a highly significant effect on plant population ($p<0.001$), whereas treatment ($p=0.551$) and the treatment x site interaction ($p=0.481$) were not significant. This indicated that plant establishment and survival were mainly influenced by site-specific conditions rather than nutrient management treatments. At Anekot (Site A), treatment effects on the final plant population were statistically non-significant ($p= 0.289$), despite the numerical differences among treatments (Fig. 2). However, effect of replication was significant ($p=0.025$) suggesting the presence of within-site variability influencing plant survival. Similarly, at Bhotechaur (Site B), no significant treatment effect ($p=0.972$) was observed on final plant population (Fig. 3). When the data were averaged across two sites, treatment effects remained non-significant ($p=0.709$) (Fig. 4). Across treatments, the average plant population ranged from ~54 plants (T_2, 100% NPK + Zn/B) to ~61 plants (T_7, multi-mix compost at 20.0 tonsha^{-1}), with all treatments belonging to a single statistical group, confirming the absence of treatment-induced differences. In summary, final plant population was largely independent of nutrient management treatments, with plant survival primarily governed by site-specific factors, indicating that the establishment of ginger and its survival were largely independent of nutrient source under the conditions of this study. The observed reductions in final plant stand from the initial 100 plants per plot are likely attributable to environmental conditions and biotic or abiotic stresses, rather than treatment effects.

Response on yield : The interaction effect between treatment and site was non-significant ($p=0.937$). There was no significant effect of treatments on yield at individual sites, and this response was consistent across both locations (Figs. 2 and 3). This indicates that, at either site, yield remained statistically similar across all treatments. At Anekot (Site A), the treatment effects on yield were not significant ($p=0.20$) (Fig. 2). This indicated that variation among replications contributed to differences in yield. While at Bhotechaur (Site B), no significant differences among treatments ($p=0.20$) were observed (Fig. 3). Treatment as T_7 (multi-mix compost at 20 tonsha^{-1}) had the highest yield at both sites (24.86

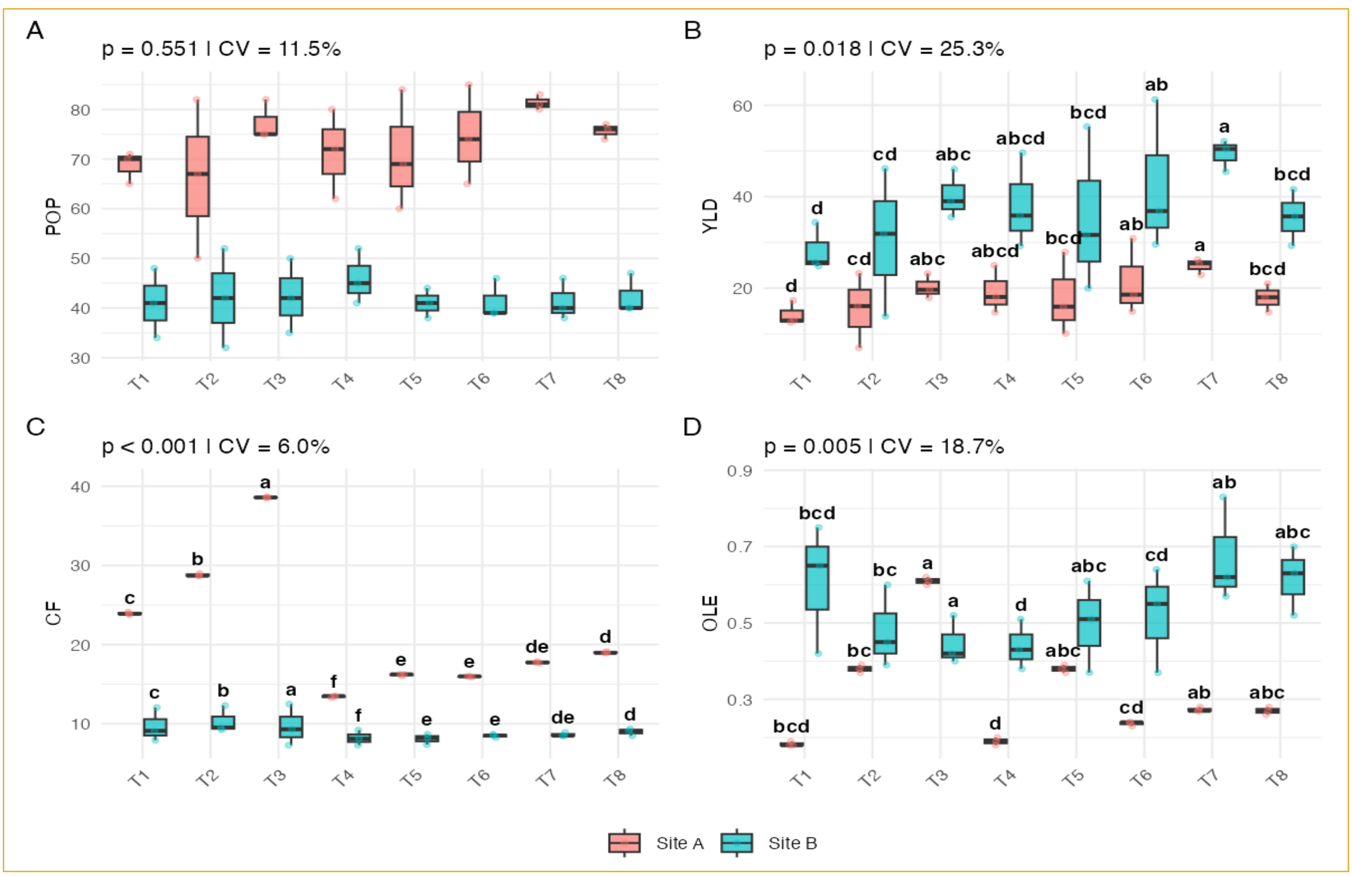

Fig. 1: Pooled analysis of treatment effects on plant population (A), yield (B), crude fiber content (C), and oleoresin content (D) across two locations using a combined RCBD model with site and treatment interaction terms. Colors indicate sites. Boxplots and points show replicate-level variation within each site. LSD group letters are based on the pooled treatment effect when significant (α = 0.05).

tons ha^{-1} at A and 49.32 tons ha^{-1} at B). These observations aligned with the established principle that when humus and organic matter are available, ginger grows well and has a favorable relationship with yield (Divyashree *et al.*, 2022). Similarly, Srinivasan *et al.* (2018) stated INM reduced nitrogen losses via nitrification, leaching, volatilization, and N_2O emissions while improving inorganic nutrient use efficiency, leading to higher rhizome yield. Treatments T_1 (control, 14.25 tonsha^{-1}A, 28.28 tonsha^{-1}) and T_2 (100% NPK + Zn/B, 15.44 t/ha-A, 30.63 t/ha-B) were consistently the lowest performers. Although, the effects of treatments on the yield were statistically non-significant, the use of organic fertilizers such as multi-mix compost and the integrated use of organic and chemical fertilizers demonstrated higher rhizome yield over chemical fertilizers alone.

The enhanced performance of organic nutrients was attributed to improved soil structure, slow release of nutrients, and water retention (Abdou *et al.*, 2023), which enhanced the rhizome growth. Amala *et al.* (2023) also reported optimal performance achieved even at reduced (50%) RDF levels. These results also agreed with Noor *et al.* (2008), who reported that integrated use of zinc and boron along with recommended doses of NPK, produced significantly higher ginger yield. Similarly, Shaikh *et al.* (2010) also documented that the recommended dose of fertilizer combined with farmyard manure favorably influenced the yield and uptake of nutrients of ginger.

Collectively, the data from Anekot and Bhotechaur confirm that the combined effect of organic and inorganic fertilizers in INM strategies is a useful approach to acquire higher yield. These results demonstrate two viable paths toward the sustainable intensification of ginger production, one fully organic and one integrated. From a sustainability perspective, the success of treatments incorporating reduced chemical fertilizers (50% NPK) demonstrated the potential for minimizing synthetic input use while maintaining high productivity, as indicated by the finding that integrated nutrient management systems produced higher values for a variety of economic factors in comparison to organic and conventional nutrient management (Divyashree *et al.*, 2022).

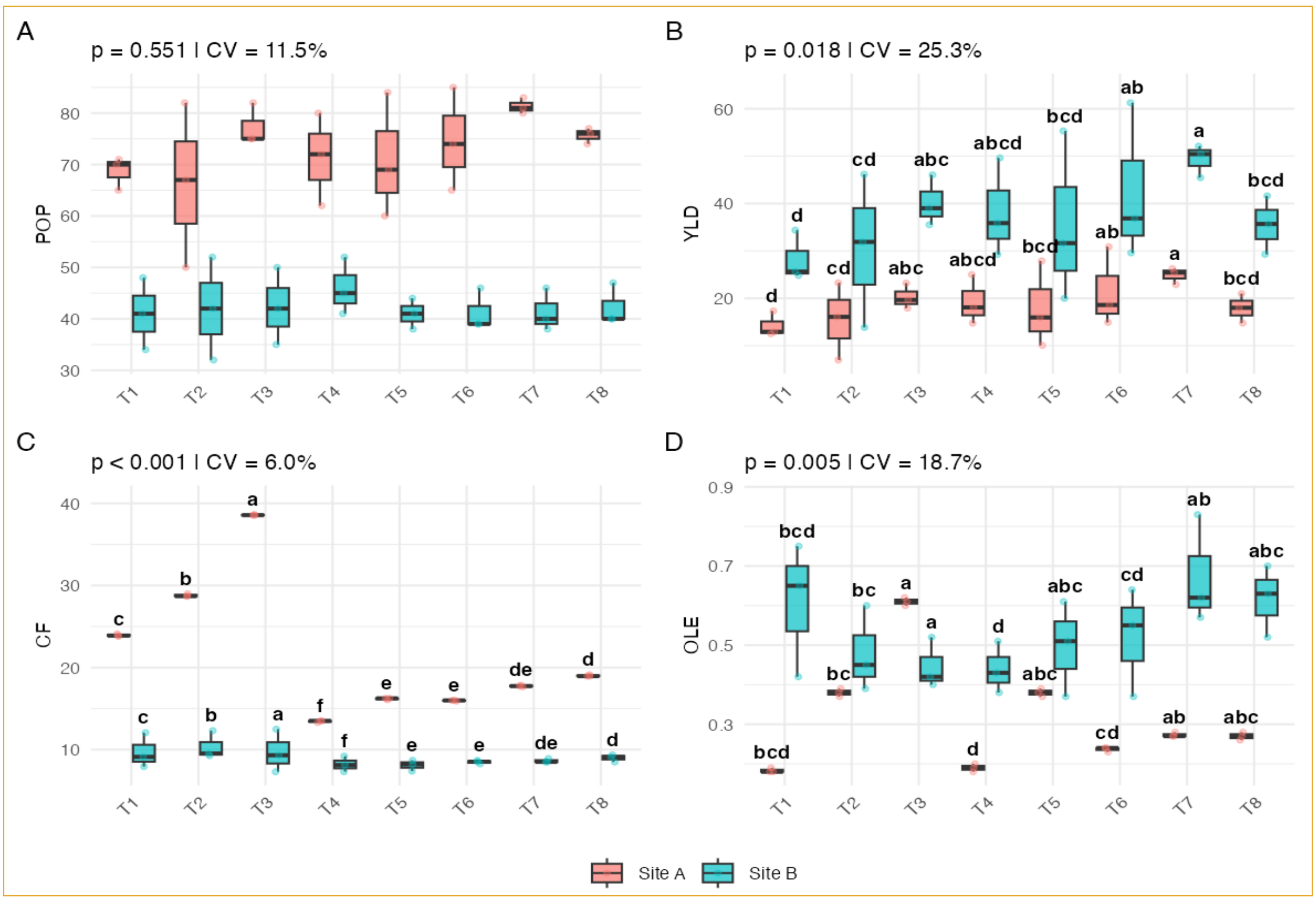

Fig. 2: Treatment effects on plant population (A), yield (B), crude fiber content (C) and oleoresin content (D) at Anekot, Sindhupalchok (Site A). Boxplots, jitter points, and whiskers show replicate variability within treatments. Letters above treatments indicate significant differences based on the LSD test at α=0.05 when the treatment effect was significant.

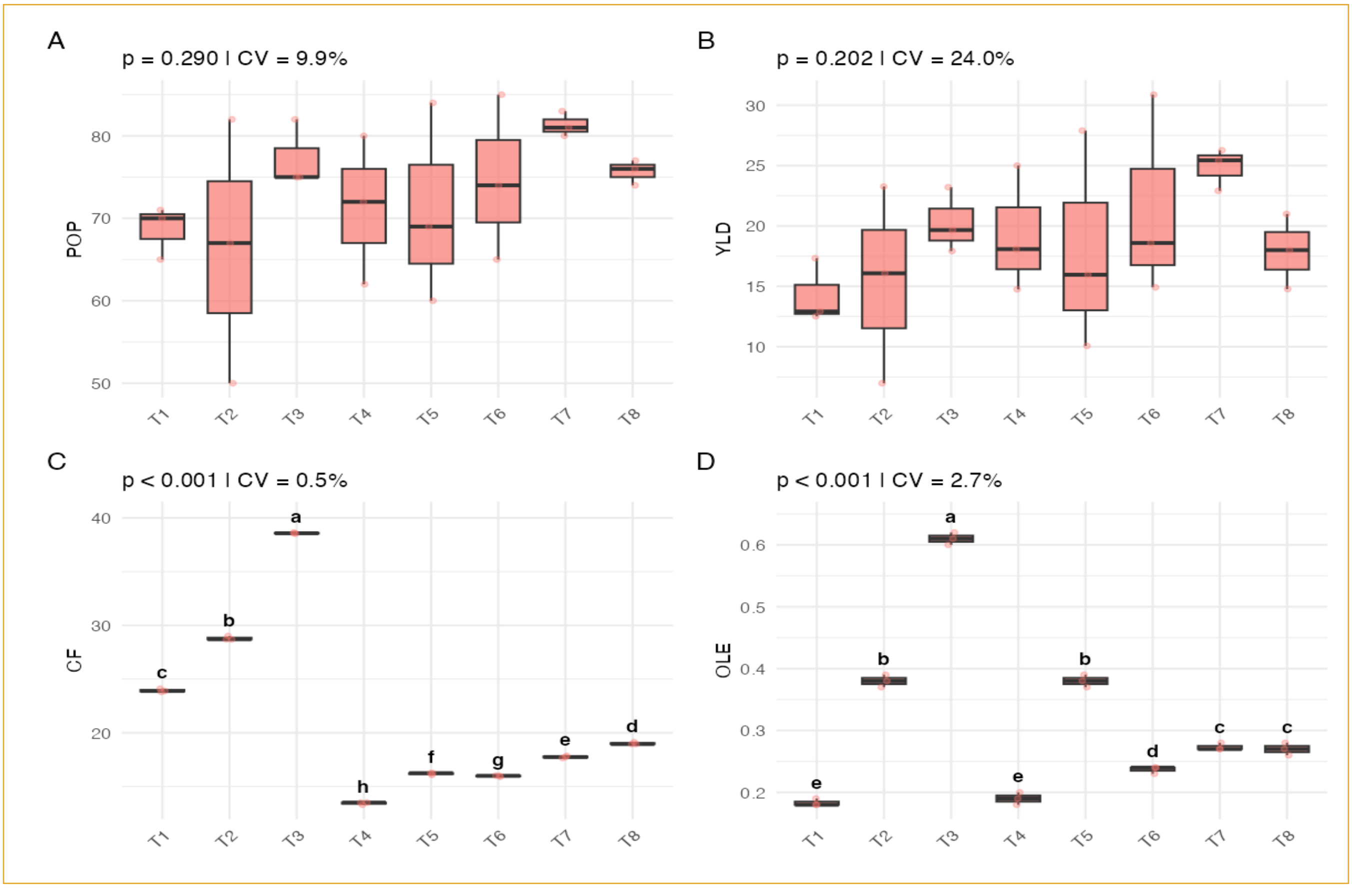

Fig. 3: Treatment effects on plant population (A), yield (B), crude fiber content (C) and oleoresin content (D) at Bhotechaur, Sindhupalchok (Site B).

Effect on quality parameters

Crude fiber content : The crude fiber content of rhizomes was significantly influenced by the treatment, but this effect was dependent on the experimental site, as indicated by a highly significant treatment x site interaction ($p < 2 \times 10^{-16}$). At Site A, the treatments had a strong and significant effect on the crude fiber content ($p<0.001$), showing that the treatments greatly affected fiber content of the ginger rhizomes (Fig. 2). Lowest fiber contents (13.43%), a desirable quality trait, was observed in T_4 (50% NPK + fine compost + Zn/B) followed by treatment T_6 (coarse compost only) of 15.97%, T_5 (fine compost only) of 16.19% and T_7 (multi-mix compost only) of 17.75%. Treatment T_3 (50% NPK + coarse compost + Zn/B) resulted in the highest crude fiber content of 38.58% which is undesirable ginger quality. In contrast, no significant differences among treatments were detected at Site B ($p=0.373$) (Fig. 3). Treatment T_4 numerically produced one of the lowest crude fiber values (8.21%), although differences among treatments were not statistically significant. These outcomes aligned with independent reports confirming that the fiber content was significantly reduced in organic nutrient management system (Divyashree *et al.*, 2022). When averaged across both sites, the overall treatment ranking was dominated by the strong response observed at Site A (Fig. 4). Treatment T_4 consistently yielded the lowest mean crude fiber content (10.82%), while T_3 promoted the highest fiber accumulation (24.14%). This aligns with findings from Li *et al.* (2021) who reported that balanced nutrition reduces lignification and improves tissue quality through optimized carbohydrate partitioning. In summary, while the expression of treatment effects on crude fiber was highly site-specific, the use of integrated nutrient sources along with micronutrients emerged as the best and most effective treatment for minimizing crude fiber content, a key quality trait for ginger. This suggested that the combined application of compost with reduced NPK and micronutrients created optimal conditions for secondary metabolite synthesis, potentially through enhanced phosphorus availability and microbial symbiosis as stated by (Abdou *et al.*, 2023).

Oleoresin content: Oleoresin content of ginger rhizomes was significantly influenced by treatment, but this effect was strongly dependent on the experimental

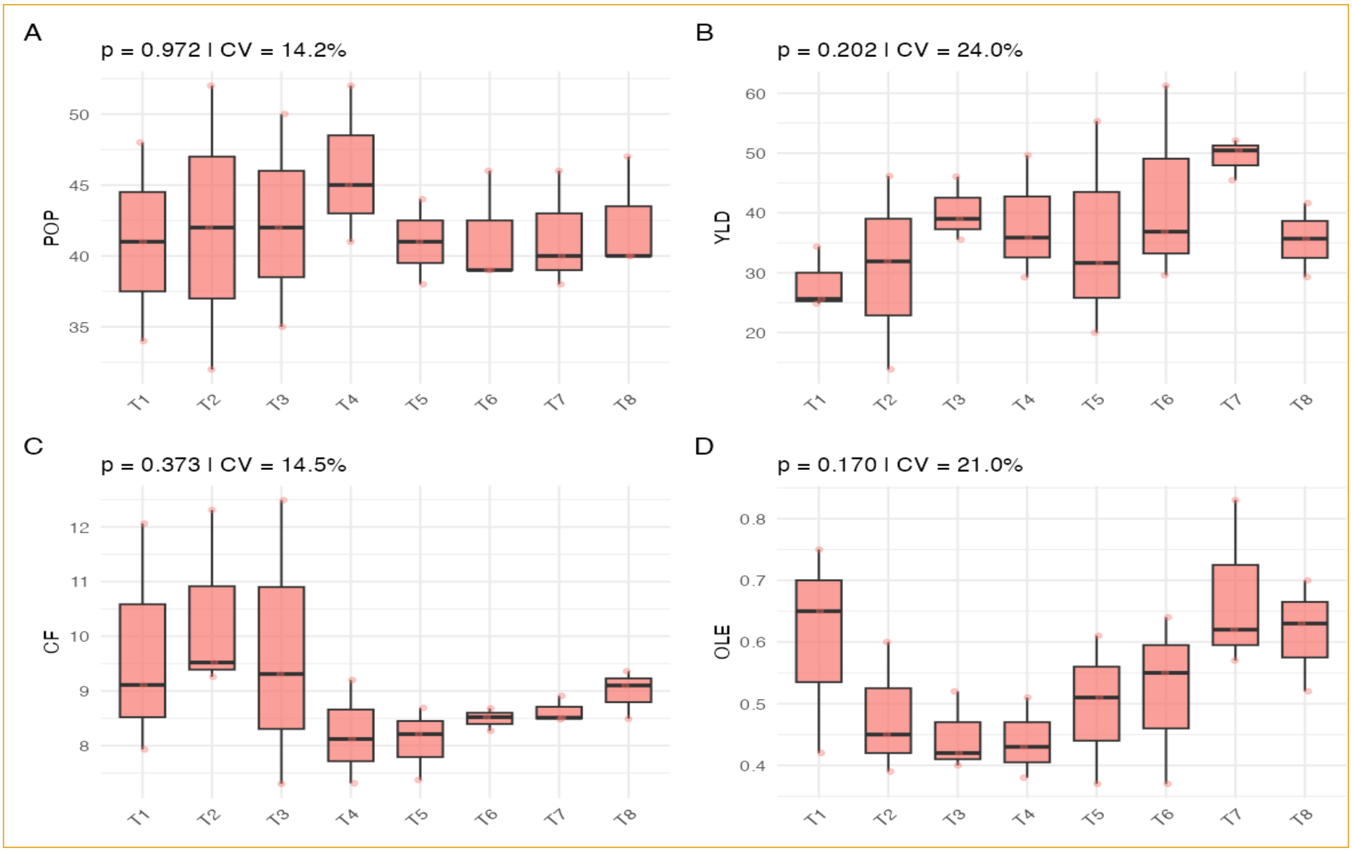

Fig. 4. Treatment performance averaged across Site A and Site B for plant population (A), yield (B), crude fiber content (C), and oleoresin content (D). Boxplots summarize the replicate variation of averaged values. Significance groupings (LSD, α = 0.05) are shown where applicable.

site, as indicated by a highly significant treatment × site interaction ($p = 8.68 \times 10^{-6}$) (Fig. 1). At Site A, treatment effects were extremely significant ($p<2 \times 10^{-16}$), with very high experimental precision (CV = 2.73%), indicating strong treatment differentiation (Fig. 2). Treatment T_3 (50% NPK + coarse compost + Zn/B) produced the highest oleoresin content (0.61), followed by T_2 (0.380%) and T_5 (0.380%), whereas T_1 and T_4 recorded the lowest values (0.183% and 0.190% each). In contrast, no significant differences were observed at Site B (p=0.170) (Fig. 3). Here, treatment T7 numerically recorded the highest oleoresin (0.67), while T_3, the top performer at Site A, recorded a comparatively lower oleoresin content at Site B (0.447%). These findings align with past reports that although the essential oil content was relatively higher in INM in comparison to conventional and organic nutrient management, there was very little variation in oleoresin and fiber contents among the different treatments (Srinivasan *et al.*, 2018).

When averaged across both sites, overall treatment means were heavily influenced by the strong response at Site A (Fig. 4). Treatment T_3 emerged as the top performer (0.530%), maintaining the highest overall average, while T_4 remained the lowest (0.320%), consistently producing the lowest oleoresin. The stronger response across all measured parameters at Bhotechaur suggests a highly responsive and biologically active soil environment. These observations aligned with the understanding that beneficial rhizosphere microorganisms can boost plant growth via multiple regulatory biochemical pathways, including manipulating the plant hormonal signaling, preventing pathogenic microbial strains and increasing the bioavailability of nutrients (Bargaz *et al.*, 2018). According to Sidhu and Shekon (2000), improvements in crop quality under integrated nutrient management arise from balanced macro and micronutrient supply and enhanced soil physicochemical and biological conditions that promote root proliferation and nutrient utilization.

CONCLUSION

The results clearly demonstrated that IPNM, combining compost, chemical fertilizers, and micronutrients, significantly influenced ginger quality attributes at both experimental sites, indicating that nutrient source and its balance play a decisive role in determining ginger quality. For yield responses, treatments incorporating organic and inorganic nutrient sources consistently outperformed the control and the treatment with only chemical sources across both sites, reflecting the benefits of nutrient integration. The mid hills of Nepal, having similar edaphic and biophysical conditions like Bhotechaur and Anekot, are suggested to use 50% of the recommended NPK dose plus 10 $tons ha^{-1}$ quality enhanced compost with Zn and B supplements as a soil application for better quality and yield of ginger rhizome. Additionally, farmers opting for organic farming are advised to apply quality enhanced compost at least 20 $tons ha^{-1}$ to obtain a better rhizome yield.

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Effect of NAA on fruit drop and quality attributes of litchi (*Litchi chinensis*)

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Abstract

The study envisaged the effect of synthetic auxin on fruit drop, cracking, yield and quality attributes of litchi (*Litchi chinensis* Sonn.) cultivars, *viz*. Dehradun and Calcuttia in sub mountaneous zone of Punjab (India). The experiments were conducted at four locations for both cultivars, during 2021 and 2022. The different concentrations of NAA (10, 20, 30, 40 ppm) along with the control (water spray) were applied 10 days after fruit setting (DAFS). The pooled data of two years revealed that application of NAA (20 ppm) drastically reduced fruit drop, cracking index, enhanced fruit retention and quality attributes, i.e. fruit weight, pulp: stone ratio, TSS and total sugars. Pericarp anthocyanin and; a^* and *hue* angle (h^*) colour coordinates were also improved with NAA treatments as compared to the control. Fruit yield/plant was better with NAA and it ranged from 5.67 to 19.55 % in 'Dehradun' and 7.40 to 17.33 % in 'Calcuttia' cultivar over the control. The application of NAA (20 ppm) effectively reduced the incidence of fruit drop, cracking, improving fruit retention and juice quality.

Key words: Auxin, Fruit drop, Cracking, Colour development, Fruit setting

Litchi (*Litchi chinensis* Sonn.) is well known for its dietic attributes such as higher content of sugars, vitamins, organic acids, phenolic acids, phytochemicals, anthocyanin, minerals etc. It provides good return to growers of Punjab (Singh *et al*., 2022). It is gown in 4,518 ha with a total production of 74,974 MT and productivity of 16.60 tonnes/ha (MA&FW, 2024-25). The litchi-based cropping system is affected with physiological disorders, fruit drop and cracking (Lal *et al*., 2017). Although, litchi tree flowers profusely but fruit setting in specific litchi cultivars varies from 1 to 50 per cent, which is far higher than fruits retained till maturity (Nath *et al*., 2021). Around 90-95 per cent fruit drop occurs up to third week after pollination (Gharge *et al*., 2025). The initial wave of fruit drop is mainly due to improper pollination, poor fertilization, and embryo abortion but fruit drop, during later fruit developmental stages, is ascribed to the lack of assimilates, irrigation, environmental factors, insect attack and hormonal imbalances (Lal *et al*., 2021). The higher level of growth promoters and lower abscisic acid (ABA) content are prerequisite for fruit set and afterward for fruit growth, development, and retention (Yuan *et al*., 2002). The foliar application of growth regulators is effective in improving fruit yield and quality (Hitesh *et al*., 2025; Piyadarshi and Hota, 2021). Lower ratio of growth promoters to ABA and ethylene concentrations may induces the formation of pre-mature abscission layer in pedicel that results in more loss of fruit productivity (Yuan and Carbaugh, 2007). Keeping in view physiological fruit drop in litchi, present investigation was intended to compare the efficacy of NAA in management of physiological disorders and fruit quality parameters.

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MATERIALS AND METHODS

The experiment was laid out in sub-mountane regions of Punjab (encompassing districts Hoshiarpur and Pathankot) situated between 30°9' to 32°05' N and 75°32' to 76°12' E, and 32°21' N and 75°31' E to 75°46' E, respectively during 2021 and 2022 on both uniform and healthy plants of 'Dehradun' and 'Calcuttia' litchi cultivars. The experimental plants were sprayed with NAA (Naphthalene acetic acid) (10, 20, 30, 40 ppm) after 10 days fruit set. The spray solution was prepared by dissolving the calculated dose of NAA (Lab. grade) in alcohol; then final volume in water along with spreader Tween 20 (0.01%). The control trees were sprayed with plain water (10 litres/plant). The cultural practices were adopted as per 'Package of Practices for Cultivation of Fruit Crops', Punjab Agricultural University, Ludhiana (2025).

Fifteen terminal flowering shoots / plant were randomly selected in each direction of tree canopy and tagged for recording various observations. The data on fruit retention, marketable yield and quality parameters were assessed and pooled estimates for four locations for each cultivar was worked out. Number of fruits in each tagged panicles were counted after fruit set before the plants sprayed and the percentage was worked out after counting the final fruit set about one week prior to harvesting. Fruit yield was estimated by weighing all fruits during harvesting and was expressed in kg/plant. Twenty fruits plant were harvested at physiological maturity for computing their average physico-chemical attributes. The fruits were weighed using digital balance and the average weight was expressed in 'g'. Fruit size (length, breadth, thickness) was measured with digital Vernier's Calipers' and expressed in 'mm'.

The pulp/stone was calculated based on fruit pulp weight and seed weight. Fruit cracking index (CI) was determined by counting total number of cracked fruits/ panicle, dividing it with total number of fruits and percentage was worked out (Haq and Rab, 2012). TSS (%) of fruit juice was recorded using digital hand refractometer (Atago, Japan) at 20°C with necessary correction factor, while acidity per cent was determined by titrating fruit juice with N/10 NaOH using phenolphthalein (1%) as an indicator. The acidity was expressed in maleic acid content. Juice sugars were analyzed as per (AOAC, 2005). Hue angle was determined by measuring the colour coordinates (a^* and b^* values wherein a^* value depicts the proportion of red to green colour, and b^* value specifies the proportion of yellow to blue colour) on each of 20 fresh fruits using ColorFlex EZ, (Hunter Associates Laboratory, Inc, USA). The hue angle was calculated by:

$$\text{Hue angle } (h^{\circ}) = \arctan (b^*/a^*)$$

Pericarp anthocyanin content was determined as per spectroscopy method suggested by Ranganna (1991). The technique involves extraction of anthocyanin using ethanolic HCl and measurement of colour at 535 nm wavelength against ethanolic HCl (used as blank).

The randomized block design (RBD) was followed with both experiments. Each experiment included four treatments and one control. Each treatment included three replications and one plant was taken as a unit. The experiment was conducted at four different locations (village Mamun, Nangal in Pathankot and Gangian, Darapur in Hoshiarpur) for each cultivar in litchi growing regions. The pooled data for different locations were analyzed with SAS package from SAS Institute Inc. (version 93, USA). To compare the variations in treatment means, the least significant difference method was employed as a post hoc analysis.

RESULTS AND DISCUSSION

Significant higher fruit retention in comparison with other treatments including the control was noted in plants sprayed with NAA (20 ppm) and values were 32.7 and 30.5% in Dehradun and Calcuttia cultivars, respectively; wherein, control had the values of 20.88 % and 22.62 %, respectively (Table 1). According to Liu (1986), levels of naturally occurring auxins are drastically reduced in litchi fruits during embryo development after 4-5 to weeks of fertilization. As a result, disintegration of cell wall and middle lamella of abscission zone occurs in pedicel due to more activity of hydrolytic cellulase and polygalacturonase enzymes (Kaur *et al.*, 2021). The spray of NAA improves the endogenous auxin content of fruits which subsequently, promotes fruit retention. Mostafa and El-berry (2020) also found that auxins promote carbohydrates mobilization from leaves to developing fruit lets and enhanced fruit retention per cent. Hence, balance between plant growth promoters and inhibitors control fruit abscission. These findings corroborate the results of Ghosh *et al.* (2012).

Table 1: Effect of NAA on physical attributes of litchi cultivars, pooled data for four locations

Treatment	Fruit retention (%)	Fruit yield (kg/ plant)	Fruit weight (g)	Fruit length (mm)	Fruit breadth (mm)	Fruit thickness (mm)	Pulp weight (g)	Pulp: stone ratio	Fruit cracking index (%)
Dehradun									
NAA 10 ppm	25.05^{c}	79.70^{bc}	20.40^{b}	33.8^{a}	31.2^{a}	29.6^{a}	15.33^{a}	4.16^{b}	12.18^{b}
NAA 20 ppm	32.70^{a}	90.15^{a}	22.78^{a}	35.9^{ab}	32.7^{a}	31.9^{ab}	15.99^{a}	4.63^{a}	6.43^{d}
NAA 30 ppm	29.63^{b}	86.43^{ab}	20.80^{b}	35.00^{ab}	31.8^{a}	30.5^{ab}	15.45^{a}	4.13^{b}	6.22^{d}
NAA 40 ppm	28.50^{b}	82.80^{abc}	20.70^{b}	34.9^{b}	32.0^{a}	30.2^{bc}	15.52^{a}	4.10^{b}	9.16^{c}
Control	20.88^{d}	75.45^{c}	19.00^{c}	31.4^{c}	30.8^{a}	28.1^{c}	14.27^{b}	3.92^{b}	19.45^{a}
Calcuttia									
NAA 10 ppm	26.28^{c}	87.40^{ab}	20.90^{b}	34.0^{a}	32.5^{a}	29.9^{a}	14.47^{bc}	5.07^{b}	6.21^{b}
NAA 20 ppm	30.50^{a}	95.48^{a}	23.93^{a}	37.1^{a}	34.2^{a}	33.7^{a}	15.87^{a}	5.55^{a}	3.04^{d}
NAA 30 ppm	28.98^{b}	91.58^{a}	21.30^{b}	36.5^{a}	33.9^{a}	33.0^{a}	15.13^{ab}	5.08^{b}	2.92^{d}
NAA 40 ppm	27.46^{c}	90.53^{a}	21.05^{b}	36.2^{b}	34.3^{a}	32.7^{b}	15.26^{ab}	4.98^{b}	4.35^{c}
Control	22.62^{d}	81.45^{b}	19.40^{c}	32.7^{b}	29.5^{b}	28.8^{b}	13.62^{c}	4.76^{c}	9.59^{a}

Least square means with same denotation are not significantly different ($p \leq 0.05$)

In Dehradun and Calcuttia cultivars, plants sprayed with NAA (20 ppm) significantly improved fruit yield by 19.55 and 17.33 per cent, respectively over the control. Fruit yield of Dehradun and Calcuttia cultivars varied from 79.7 to 90.15 kg/plant and 87.40 to 95.48 kg/plant, respectively, compared to the control (75.45 and 81.45 kg/plant, respectively). Arunadevi *et al.* (2019) also reported that exogenous applications of NAA significantly improved fruit retention due to balance between internal concentration of auxins that inhibits the degradation of abscission layer which may result in production of higher fruits. The application of NAA may stimulate auxins that enhances source-sink relationship which in turn had higher fruit set and fruit retention (Sahay *et al.*, 2018), reduced fruit drop and substantially improved fruit yield over the control.

The trees sprayed with NAA (20 ppm) registered highest fruit weight of 22.78 g and 23.93 g in Dehradun and Calcuttia, respectively compared to other treatments (Table 1). The enhancement in fruit weight ranged from 7.37 to 25.95 % with NAA treatments compared to the untreated plants in both the litchi cultivars. Fruit size was also significantly highest with foliar spray of NAA (20 ppm) in both the cultivars except fruit breadth of Dehradun over the control. In Dehradun, significantly highest pulp weight of 15.99 g was registered with NAA (20 ppm), followed by 15.52 g in NAA (40 ppm), 15.45 g in NAA (30 ppm) and 15.33 g in NAA (10 ppm) than lowest (14.27 g) in the control. It ranged from 15.87 g in NAA (20 ppm) to 13.26 g in the control in Calcuttia cultivar. Pulp: stone ratio was also significantly influenced with the application of NAA (20 ppm) than the rest of treatments; however, NAA (10, 30 and 40 ppm) treatments were statistically non-significant with each other in both litchi cultivars. An increment in pulp:stone of 18.11 per cent in Dehradun and 16.6 per cent in Calcuttia litchi cultivar was noted with NAA (20 ppm) treatment over the control, followed by NAA (10 ppm) in Dehradun and NAA (30 ppm) in Calcuttia.

The application of NAA increases the concentration of auxins within the fruits that promotes the fruit growth and development, which in turns improves fruit weight. NAA plays significant role in reduction of pericarp fruit cracking index in both litchi cultivars. The susceptibility to cracking intensity in fruits of Dehradun cultivar was more, hence cracking index (CI) was registered maximum in untreated fruits of Dehradun (19.45 %) than Calcuttia (9.59 %). The CI was minimum in Dehradun and Calcuttia cultivars with application of NAA (30 ppm) to the tune of 6.22 and 2.92%, respectively and these values were statistically at par with NAA (20 ppm) and values were 6.43 and 3.04 %, respectively. According to Sahay *et al.* (2018), application of NAA may stimulate auxins that enhance source-sink relationships resulting in higher percentage of fruit setting, retention and lesser cracking.

Auxin application improves the water uptake by increasing the osmotic pressure of cell sap which, may reduce fruit cracking (Saraswat *et al.*, 2006). Gill and Bal (2009) also reveal that foliar spray of NAA at pea size stage substantially enhances the inherent physiology of leaves and translocation of photosynthates during ber fruit development. The susceptibility of litchi fruits to cracking may be associated with the deterioration in IAA level in fruits which in turn decreases the levels of *LcXET1* (gene involved in cell wall metabolism) in pericarp; however, NAA application significantly enhanced levels of LcXET1 mRNA in pericarp of treated fruits which, resulted in reduced fruit cracking as reported by Lu *et al.* (2006). Similarly, Kaur *et al.* (2024) observed that foliar application of NAA (10, 20, 30 ppm) significantly reduced fruit splitting by 5.69, 5.27 and 6.16 per cent as compared to 13.25 per cent in the untreated Daisy mandarin grown under central zone of Punjab.

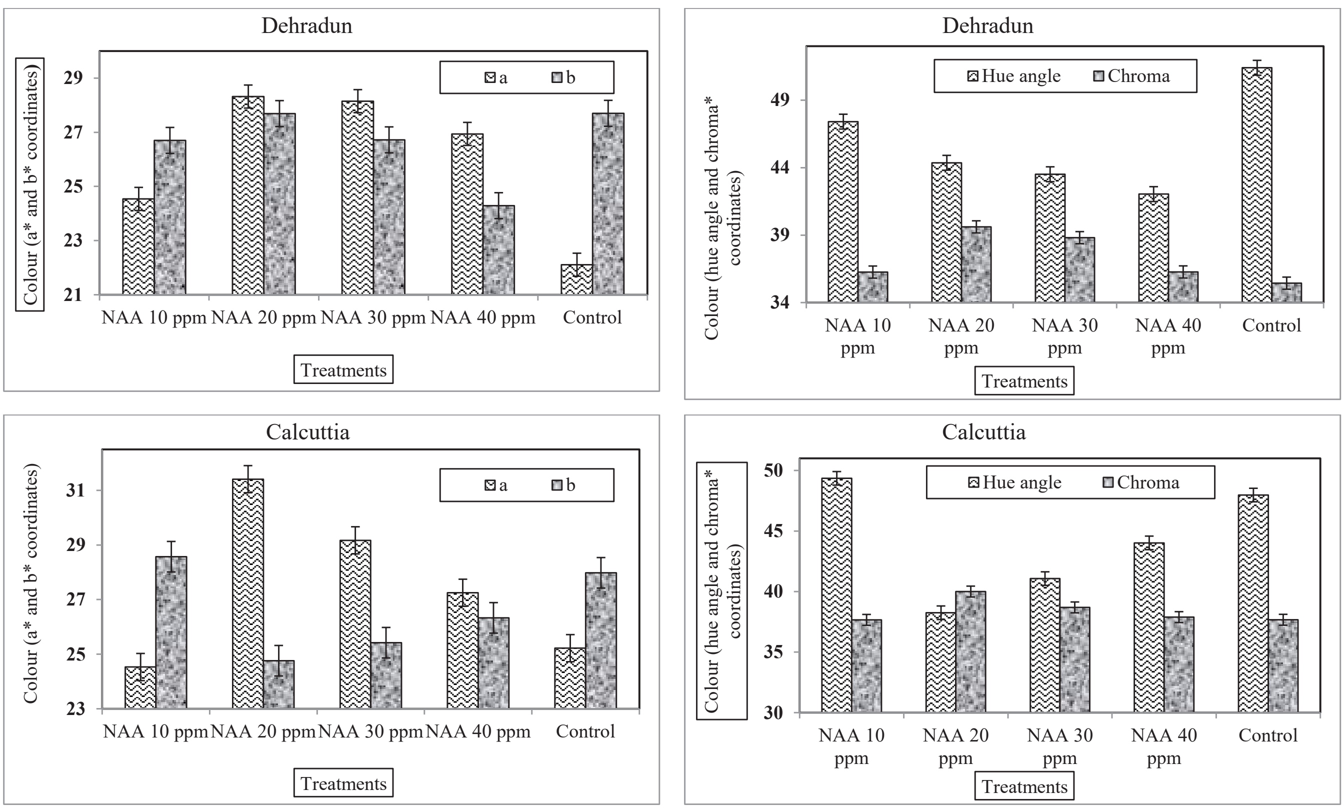
Apart from physical quality parameters, foliar sprays of auxins also remarkably produced fruits of better-quality attributes (Table 2). TSS and TSS:acid among NAA treated plants ranged from 18.78 to 19.83% and 30.55 to 37.3 % in Dehradun and 20.38 to 22.25% and 28.03 to 33.2 % in Calcuttia cultivar, respectively. Overall, foliar application of NAA significantly improved TSS, total sugars content and TSS:acid. A significant influence of NAA was also observed in different proportion of sugars content of fruit pulp. The highest percentage of reducing sugars was witnessed with treatment of NAA (20 ppm) (11.55 % in Dehradun and 12.20 % in Calcuttia) than rest of treatments. Similarly, maximum total sugars in both cultivars were also observed considerably with NAA treatments compared to the control. This may be attributed that auxin promotes rapid mobilization of photosynthates, minerals in developing fruits and increases cell membrane permeability that accelerates the breakdown of organic acids (stored in cell vacuole). Kaur *et al.* (2024) also confirmed that NAA as plant growth regulator enhanced fruit quality of Daisy mandarin.

The effect of auxins on pericarp a^*, b^* and hue angle coordinates depicted the degree of redness to greenness and yellowness to blueness and saturation of redness, respectively. It is obvious that 'a^*', 'b^*' and hue angle (h^*) coordinates were substantially influenced with NAA treatments (Fig. 1). In Dehradun, significantly higher 'a^*' coordinate was registered with NAA 20 ppm (28.32), followed by NAA 30 ppm (28.15) and NAA 40 ppm (26.94) than the rest of treatments. However, coordinate values of 'b^*' ranged from 24.29 to 27.69 in plants treated with NAA

Table 2: Effect of NAA on fruit chemical attributes of litchi cultivars, pooled data for four locations

Treatment	TSS (%)	T.S.S/acid	Reducing sugars (%)	Total sugars (%)	Anthocyanin (mg100 g^{-1}pericarp)
			Dehradun		
NAA 10 ppm	18.78^{c}	30.55^{d}	10.86^{b}	14.18^{bc}	13.50^{b}
NAA 20 ppm	19.83^{a}	37.3^{a}	11.55^{a}	14.86^{a}	13.15^{c}
NAA 30 ppm	19.40^{b}	35.0^{b}	11.11^{ab}	14.40^{b}	14.48^{a}
NAA 40 ppm	19.03^{c}	33.9^{c}	11.00^{ab}	14.51^{b}	15.00^{a}
Control	18.33^{d}	28.9^{d}	10.22^{c}	14.0^{c}	11.33^{d}
			Calcuttia		
NAA 10 ppm	20.65^{b}	28.90^{bc}	10.86^{bc}	14.71^{b}	20.33^{ab}
NAA 20 ppm	22.25^{a}	33.20^{a}	12.20^{a}	15.74^{a}	21.85^{a}
NAA 30 ppm	21.08^{b}	29.83^{b}	11.32^{bc}	14.98^{b}	20.13^{ab}
NAA 40 ppm	20.38^{b}	28.03^{c}	11.44^{ab}	15.00^{b}	19.73^{b}
Control	19.25^{c}	25.48^{d}	10.63^{c}	14.45^{b}	16.81^{c}

Least square means with same denotation are not significantly different ($p \leq 0.05$)



Vertical bars symbolize ±SE of means for 3 replicates.

Fig. 1 Effect of NAA treatments on pericarp colour in litchi cultivars

treatments. Likewise, '*b**' coordinate was observed the higher under the control in both cultivars depicting more yellowish pericarp colour.

Foliar application of NAA causes significant reduction in hue angle coordinate and notable enhancement of pericarp anthocyanin content. These findings corroborated with those of Balbontín *et al.* (2018) and they noted a decrease in hue angle with increase in anthocyanin content among sweet cherry subjected to different hormonal treatments. Dutta *et al.* (2011) also found the impact of PGRs on anthocyanin content of litchi fruits. The chroma value indicates the quantitative measure for colourfulness or purity of colour. The results indicated higher chroma value in NAA treated fruits over the control. Pericarp colour gradually changed from green to pink and finally red during fruit maturation. These transitions are not uniform across fruit surface which might be responsible for the comparative decrease in chroma values. Plant bio-regulators effectively improved fruit yield and quality due to better supply of nutrients, water and other compounds required for their growth and development that in turn produced fruits of superior size, quality and ultimately greater yield (Pandey, 1999).

CONCLUSION

Thus, application of NAA (20 ppm) after 10 days from fruit setting significantly managed physiological disorders which led to more fruit production and improved physical and chemical quality attributes of Dehradun and Calcuttia litchi cultivars growing under sub tropical climatic conditions of North India.

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Substitution response of nutrient sources on fruit yield, quality and soil fertility changes in Nagpur mandarin (*Citrus reticulata*)

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Abstract

Studies were caried out to find out the substitution response of chemical fertilizers with organic manures and biofertilizers in different proportions on quality production and soil fertility on black clay soils-growing Nagpur mandarin (*Citrus reticulata* Blanco), at Dr Punjabrao Krishi Vidyapeeth, during 2021-23. There was maximum fruit yield through balanced nutrition. The highest fruit yield was observed with the application of 100 % NPK + FYM (50 kg /tree) + biofertilizers + S + Zn + Fe + B as one combination of integrated nutrient management. Additionally, all other integrated nutrient management treatments showed promising results, with regard to fruit quality, juice recovery percentage, total soluble solids and TSS: acid ratio, compared to only chemical fertilizers. The integrated use of organic manures, biofertilizers and chemical fertilizers improved fertility status of soil, represented by available nitrogen, phosphorus, potassium and sulphur compared to individual application of organic and inorganic fertilizers. These studies showed the role of integrated nutrient use for quality production of Nagpur mandarin grown on black clay soils of central India.

Key words: Nutrient, Yield, Quality, Soil fertility status, Organic manure, Biofertilizers

Nagpur mandarin (*Citrus reticulata* Blanco) is an important citrus crop grown in India, covering 12.4 % of the total area under fruit crops. In Maharashtra, it is grown in 1.48 lakh ha with a production of 8.75 lakh tonnes and average productivity of 10-14 tonnes/ha, which is low compared to other citrus cultivars (Srivastava , 2013). Citrus crops are relatively nutrient-demanding (Srivastava and Hu, 2019 ; Srivastava , 2023) and highly responsive to applied nutrients in the form of fertilizers, particularly nitrogen, which is a critical input, involved in plant metabolism and growth, and in different biochemical processes (Srivastava *et al*., 2015 : Asthir *et al*., 2017). Enhanced yields with improved fruit quality are often obtained with integrated application of fertilizers, in right combinations of different nutrient source such as chemical fertilizers, organic manures and biofertilizers (Srivastava *et al*., 2019), but such responses are highly fluctuating depending upon nature and properties of soil facing the kind of soil fertility constraints (Srivastava and Sharma, 2025). The deficiency or excess of any nutrient under such conditions can lead to a reduction in crop yield coupled with inferior fruit quality (Srivastava and Malhotra, 2017), thus, judicious application of fertilizers including macronutrients, micronutrients and organic sources is essential for increasing the productivity as well as quality of mandarin (Srivastava and Singh, 2001; 2009). Therefore, studies were carried out to find out the right combination of chemical fertilizers, organic manure and biofertilizers to get high fruit yield, quality and soil fertility changes in Nagpur mandarin .

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MATERIALS AND METHODS

The experiment was conducted at Regional Fruit Research Station, Katol, Dr Panjabrao Deshmukh Krishi Vidyapeeth, Nagpur (Maharashtra) during 2021–23. Eleven-year-old Nagpur mandarin trees grafted on rough lemon rootstock (*Citrus jambhjri* Lush) spaced at 6 m x 6 m raised on black clay soil were employed. The experiment was designed in a randomized block design, with nine treatments, each replicated three times. The treatments comprised : T_1- only FYM (50 kg /tree) + biofertilizers, T_2- 100% recommended dose of NPK, T_3- 100% recommended dose of N only + FYM (50 kg/ tree) + biofertilizers, T_4-100% recommended dose of N and P + FYM (50 kg/ tree) + biofertilizers, T_5- 100% NPK + FYM (50 kg /tree) + biofertilizers, T_6- 100% NPK + FYM (50 kg/ tree) + biofertilizers + S, T_7- 100% NPK + FYM @ 50 kg/ tree + biofertilizers + S + Zn, T_8- 100% NPK + FYM (50 kg /tree) + biofertilizers + S + Zn + Fe and T_9- 100% NPK + FYM (50 kg /tree) + biofertilizers + S + Zn + Fe + B.

The recommended doses of fertilizers for Nagpur mandarin was observed as N 800 g- P_2O_5 300 g- K_2O 600 g -S 100 g/ tree . biofertilizers 500g VAM (*Glomus mosseae*) + 100 g phosphate solubilizing bacteria *(Bacillus megaterium)* + 100 g *Azospirillum* (*Azospirillum lipoferum*) + 100 g *Trichoderma* (*Trichoderma viride*) tree^{-1} mixed with FYM at 50 kg tree^{-1} were applied 15 days before imposition of all the treatments flowering and fruit setting stages in two equal splits. Micronutrients (Zn 0.50%, Fe 0.50% and B 0.10%)/ tree were applied in two foliar sprays in July, August and September. Nutrients (organic, inorganic and biofertilizers) were applied in a ring that covered an area of 90- cm away from periphery of tree trunk and later covered periphery of the trees with soil to facilitate easy distribution of applied fertilizers inputs.

The weight of five fruits was recorded on electronic balance and the results were expressed as weight in grams per fruit. Fruit size in terms of length (from calyx end to tip of styler end) and breadth of fruits was measured with the help of digital Vernier Calliper and the average values for length and breadth of fruits were expressed in centimeter (cm).The fruit juice was extracted by hand operated extractor. The juice per cent was calculated from juice content and total weight of fruit. The total soluble solids content in fruits was determined by Erma Hand Refractometer (0 -32 °Brix). The titratable acidity was calculated in terms of acidity on basis of one ml of 0.1 N NaOH equivalent to 0.0067 g of anhydrous ascorbic acid by using the formula of Ranganna (1987). Ascorbic acid content of fruits was also determined as per the method suggested by Ranganna (1987).

The soil samples from the zone of maximum feeder root concentration at a depth of 0–20 cm and at a distance of 110–125 cm from trunk were collected by using a soil auger during the initial phase (December 2020) and further soil samples were collected treatment-wise after harvesting of fruits . The soil samples were dried in shade, gently ground with a mortar and pestle and sieved through a 2- mm sieve to obtain a homogeneous sample. For determination of organic carbon, soil samples were passed through a 0.5- mm sieve. These samples were stored in polythene bags and were subsequently analyzed. Soil pH and EC were determined as per the methods of Jackson (1973) . Calcium carbonate was estimated using the rapid titration method and soil organic carbon were determined according to the wet oxidation method (Jackson ,1973). Available N and P were estimated using the alkaline K permanganate method and Olsen's method (Jackson, 1973). Available K was extracted in 1 N neutral normal ammonium acetate using a flame photometer (Jackson, 1973) and available S was determined by turbidity developed by barium chloride and measured spectrophotometrically at 420 nm wavelength (Chesnin and Yein, 1951) .

The results were statistically analyzed and appropriately interpreted as per the methods of Gomez and Gomez (1984) . Appropriate standard error and critical differences at 5% level were worked out.

RESULTS AND DISCUSSION

The rows treated with 100 % NPK + FYM (50 kg/ tree)+ biofertilizers + S + Zn + Fe + B (T_9) produced the highest number of fruits (561 fruits /tree), followed by treatment 100 % NPK + FYM (50 kg/ tree) + biofertilizers + S + Zn + Fe (T_8) recording 531fruits/ tree and 540 fruits /tree with treatment involving 100 % NPK + FYM (50 kg/ tree) + biofertilizers + S + Zn (T_7) (Table 1). These treatments were found on a par with each other but significantly superior to rest of the other treatments. In contrast, lowest number of fruits was observed with treatment carrying only FYM (50 kg/ tree) +biofertilizers (T_1) (315).Our results align with earlier observations made by Nurbhanej *et al.* (2016) , demonstrating more fruits due to inorganic fertilizers alone, and in combination with organic sources further facilitated sustained nutrient supply and nutrient-use of applied nutrients through improved microbial activity involved in nutrient transformation and fixation.

The average fruit weight was significantly higher with treatment 100 % NPK + FYM (50 kg/tree) + biofertilizers + S + Zn + Fe + B (T_9) with 113.74 g as pooled mean. This was followed by all other treatments using integrated supply of nutrients, excluding the inorganic nutrient source , superior to other treatments of integrated nutrients-supply. Conversely, a significantly lower average fruit weight was observed with the treatment with only FYM (50 kg/ tree) + biofertilizers (T_1) registering fruit weight of 106.59 g. Application of inorganic NPK along with biofertilizers resulted in a higher rate of photosynthesis conditioned by optimum soil fertility, eventually led to higher carbohydrate accumulation in fruits, and thereby causing improvements in fruit size and weight (Srivastava *et al.*, 2021). Regarding yield, significantly higher yields in terms of tree/kg and tonnes/ha were obtained with combined application of 100 % NPK + FYM (50 kg/ tree) + biofertilizers + S + Zn + Fe + B (T_9) as 63.93 kg /tree and 17.71 tonnes/ha , respectively.

These treatments were on par with each other and significantly superior to individual application of either organics (33.53 kg/tree and 9.29 tonnes/ha) or inorganic fertilizers (55.76 kg /tree and 15.44 tonnes/ ha), also significantly superior to remaining treatments. The integrated use of chemical fertilizers with organic materials added a significant quantity of organic matter to the soil, resulting in higher fruit yield. Similar results were earlier reported (Srivastava, 2023 ; Srivastava *et al.*, 2015), advocating maximized fruit yields with chemical fertilizers in combination with farmyard manure and biofertilizers (Srivastava and Sharma , 2025) .

Fruit quality parameters, including juice recovery, total soluble solids (TSS) and TSS: acid ratio, were

Table 1: Response of different integrated nutrient management treatments on fruit yield-attributing parameters of Nagpur mandarin (pooled data: Two seasons)

	Treatment	Number of fruits /tree	Average fruit weight (g)	Fruit yield (kg/ tree)	Fruit yield (tonnes/ ha)
T_1	FYM (50 kg/ tree) + biofertilizers	315	106.59	33.53	9.29
T_2	100 % recommended dose of NPK	501	111.10	55.76	15.44
T_3	100 % recommended dose of N only + FYM (50 kg /tree) + biofertilizers	384	108.78	41.72	11.56
T_4	100 % recommended dose of N and P + FYM (50 kg/ tree) + biofertilizers	461	111.02	51.24	14.19
T_5	T_2 + FYM @ 50 kg/ tree + Biofertilizers	523	112.17	58.78	16.28
T_6	T_2 + FYM (50 kg /tree) + biofertilizers + S	528	112.09	59.07	16.36
T_7	T_2 + FYM (50 kg /tree) + biofertilizers + S + Zn	540	113.62	61.47	17.03
T_8	T_2 + FYM (50 kg/ tree) + biofertilizers + S + Zn + Fe	551	113.65	62.57	17.33
T_9	T_2 + FYM (50 kg/ tree) + biofertilizers + S + Zn + Fe + B	561	113.74	63.93	17.71
	SE m (±)	13.08	2.13	1.83	0.51
	CD at 5 %	39.21	NS	5.48	1.52

significantly influenced by different nutrient management treatments (Table 2). However, influence of different treatments on fruit height, width, volume, peel thickness, acidity and ascorbic acid content showed non-significant response. The highest fruit height (5.92 cm), width (6.56 cm) and volume (171.15 cm^3) were observed with treatment carrying combination of 100 % NPK + FYM (50 kg /tree) + biofertilizers + S + Zn + Fe + B (T_9). While, lowest was recorded with treatment involving only organic inputs (T_1), though a slight improvement in fruit height was observed with treatment involving combined application of organic manure, biofertilizers and inorganic fertilizers along with foliar sprays of micronutrients, specifically in 100 % NPK + FYM (50 kg/tree) + biofertilizers + S + Zn + Fe + B . Previously, Hazarika and Aheibam (2019) reported an increase in fruit height due to the integration of organic and inorganic sources. Additional findings from Bakshi *et al.* (2018) indicated that application of organic, inorganic and biofertilizers significantly improved fruit width.

Improved fruit quality was observed, as indicated by a decrease in fruit juice acidity and ascorbic acid content, as well as peel thickness. The lowest fruit juice acidity (0.72 %) and ascorbic acid content (34.89 mg 100 /mL) were recorded with the treatment combining organic manure, biofertilizers and foliar sprays of micronutrients, specifically with treatment, 100 % NPK + FYM (50 kg / tree) + biofertilizers + S + Zn + Fe + B (T_9), compared to treatments using only inorganic fertilization. The lowest peel thickness (3.28 mm) was observed with the treatment with carrying FYM (50 kg/ tree) + biofertilizers (T_1) . These results are in agreement with the finding of Kumar *et al.* (2017). Fruit juice recovery significantly increased with different nutrient management practices. The highest juice recovery of 46.77% was observed with treatment carrying 100 % NPK + FYM (50 kg /tree) + biofertilizers + S + Zn + Fe + B (T_9) , followed by 46.33% with 100 % NPK + FYM (50 kg/ tree) + biofertilizers + S + Zn + Fe (T_8) and 46.07% with 100 % NPK + FYM (50 kg/ tree) + biofertilizers + S + Zn (T_7) . However, lowest juice recovery of 43.82% % was observed with the treatment with carrying FYM (50 kg /tree) + biofertilizers (T_1). Ennab (2016) observed the highest juice recovery (48.14 %) with combined use of farmyard manure and biofertilizers with a dose of NPK . A similar finding was also reported by Srivastava *et al.* (2019) highlighting significant increase in juice content of mandarin fruits with combination of 75 % RDF (recommended doses of fertilizers) and 25 % RDF-equivalent vermicompost and microbial consortium, which significantly improved different fruit quality parameters over 100 % RDF alone.

The highest total soluble solids of 9.53% in fruit juice was recorded with the treatment with 100 % NPK + FYM (50 kg /tree) + biofertilizers + S + Zn + Fe + B (T_9) , followed by 9.36% with 100 % NPK + FYM (50 kg/tree) + biofertilizers + S + Zn + Fe (T_8) (9.36), at par with 9.33% with 100 % NPK + FYM (50 kg/tree) + biofertilizers + S + Zn (T_7) . These treatments were on par with each other, but significantly superior to other treatments. The lowest fruit TSS of 8.72% was observed with only FYM (50 kg/ tree) + biofertilizers (T_1) , aligning these results with previously reported by Kumar *et al.* (2017).

The maximum soil pH was recorded with treatment carrying 100 % NPK (T_2) (Table 3). Such treatment response of FYM is accountable to acidifying effect of urea and organic acids produced during the course of the decomposition of organic amendments, a well-known fact established through a large number of experiments . While, electrical conductivity indicating total soluble salts

Table 2 : Response of different integrated nutrients-supply treatments on fruit quality of Nagpur mandarin (pooled data: two seasons)

	Treatment	Fruit height (cm)	Fruit width (cm)	Fruit volume (cm^3)	Peel thickness (mm)	Juice recovery (%)	Ascorbic acid (mg 100/ mL)	TSS (°Brix)	Acidity (%)
T_1	FYM (50 kg/ tree) + biofertilizers	5.65	6.40	168.39	3.28	43.82	35.26	8.72	0.75
T_2	100 % recommended dose of NPK	5.68	6.42	170.07	3.44	44.90	35.33	8.77	0.76
T_3	100 % recommended dose of N only + FYM (50 kg /tree) + biofertilizers	5.73	6.37	168.59	3.36	44.14	35.21	8.78	0.75
T_4	100 % recommended dose of N and P + FYM (50 kg/ tree) + biofertilizers	5.72	6.44	170.19	3.36	44.61	35.22	8.94	0.74
T_5	T_2 + FYM (50 kg/ tree) + biofertilizers	5.78	6.50	170.41	3.35	45.58	35.31	9.14	0.74
T_6	T_2 + FYM (50 kg/ tree) + biofertilizers + S	5.75	6.52	170.10	3.37	45.66	35.23	9.18	0.75
T_7	T_2 + FYM (50 kg/ tree) + biofertilizers + S + Zn	5.83	6.53	171.86	3.34	46.07	34.96	9.33	0.74
T_8	T_2 + FYM (50 kg/ tree) + biofertilizers + S + Zn + Fe	5.87	6.59	171.05	3.34	46.37	35.00	9.36	0.73
T_9	T_2 + FYM (50 kg/ tree) + biofertilizers + S + Zn + Fe + B	5.92	6.56	171.15	3.31	46.77	34.89	9.53	0.72
	SE m (±)	0.22	0.31	1.86	0.02	0.53	0.16	0.16	0.01
	CD at 5 %	NS	NS	NS	NS	1.59	NS	0.48	NS

in the soil did not change much due to various treatments, it ranged narrowly between 0.25 to 0.28 dS/m. The minimum $CaCO_3$ in soil was observed in organic manure-incorporated treatments and maximum was recorded in treatment 100 % NPK (T_2). The organic acids released during decomposition of organic manures reacted with $CaCO_3$ to release CO_2, thereby reducing the concentration of $CaCO_3$ in the soil .

Significantly highest available nitrogen of 312.2 kg/ ha in soil was recorded with treatment receiving 100 % NPK + FYM (50 kg/tree) + biofertilizers + S + Zn (T_7), on par with treatments, viz. 100 % NPK + FYM (50 kg/tree) + biofertilizers + S + Zn + Fe (T_8) , 100 % NPK + FYM (50 kg/ tree) + biofertilizers + S (T_6) , 100 % NPK + FYM (50 kg/ tree) + biofertilizers + S + Zn + Fe + B (T_9) and 100 % NPK + FYM (50 kg/ tree) + biofertilizers (T_5) , but significantly superior over treatment 100 % recommended dose of N only + FYM (50 kg/tree) + biofertilizers (T_3) (298.5 kg/ ha) and 100 % NPK + FYM (50 kg/tree) + biofertilizers (T_2) (294.8 kg/ha). The availability of organically bound nitrogen through transformation in soil to plant mainly depended on the population of microorganisms, influenced by application of inorganic fertilizers and organic manure, as reported (Srivastava *et al.*, 2021).

The available phosphorus in soil after harvest of Nagpur mandarin was noticed highest (20.80 kg/ha) with treatment. 100 % NPK + FYM (50 kg/tree) + biofertilizers + S (T_6) (), on par with treatments like 100 % NPK + FYM (50 kg/tree) + biofertilizers (T_5) , 100 % NPK + FYM (50 kg/tree) + biofertilizers + S + Zn + Fe + B (T_9) , 100 % NPK + FYM (50 kg/tree) + biofertilizers + S + Zn (T_7) , 100 % NPK + FYM (50 kg/tree) + biofertilizers + S + Zn + Fe (T_8) and 100 % recommended dose of N and P + FYM (50 kg/tree) + biofertilizers (T_4) and 100 % NPK (T_2) , but significantly superior over treatment 100 % recommended dose of N only + FYM (50 kg/tree) + biofertilizers (T_3) . Considerable improvement in available P status was observed due to residual effect of applied fertilizers and mineralization of organic sources coupled with solubilization of nutrients from the native sources during the process of decomposition (Srivastava, 2023).

Several other studies reported an increase in soil available P content with the application of FYM + inorganic fertilizers (Thakur *et al.*, 2009) and vermicompost + biofertilizers + inorganic fertilizers (Nakade *et al.*, 2021). The available potassium in soil after the harvest of Nagpur mandarin was noticed to be the highest (364.0 kg/ ha), with treatment involving 100 % NPK + FYM (50 kg/ tree) + biofertilizers + S + Zn + Fe + B (T_9) and significantly superior (344.6 kg/ha) over treatment 100 % NPK (T_2) or 100 % recommended dose of N and P + FYM (50 kg/tree) + biofertilizers (T_4) and 100 % Recommended dose of N only + FYM (50 kg/tree) + biofertilizers (T_3) (341.7 kg/ ha).

The increase in K- availability was attributed to direct addition of K through fertilizers to available pool of soil, in addition to K solubilized from K-bearing minerals by organic acids released from the organic manures

Table 3 : Soil fertility changes in response to different treatments of integrated nutrient management (pooled data: two seasons)

	Treatment	Soil pH	E C (dS m^{-1})	$CaCO_3$ (%)	Available nitrogen (kg/ ha)	Available phosphorus (kg/ ha)	Available potassium (kg/ ha)	Available sulphur (mg/ kg)
T_1	FYM (50 kg/ tree) + biofertilizers	7.61	0.27	3.72	291.7	18.49	339.7	14.30
T_2	100 % recommended dose of NPK	7.63	0.28	3.74	294.8	19.17	344.6	14.01
T_3	100 % recommended dose of N only + FYM (50 kg/ tree) + biofertilizers	7.60	0.25	3.70	298.5	18.81	341.7	14.42
T_4	100 % recommended dose of N and P + FYM (50 kg/ tree) + biofertilizers	7.60	0.25	3.69	304.4	20.22	343.8	14.57
T_5	T_2 + FYM (50 kg/ tree) + biofertilizers	7.58	0.26	3.69	307.5	20.75	361.0	14.84
T_6	T_2 + FYM (50 kg/ tree) + biofertilizers + S	7.57	0.26	3.70	311.7	20.80	363.9	15.49
T_7	T_2 + FYM (50 kg/ tree) + biofertilizers + S + Zn	7.57	0.25	3.66	312.2	20.69	363.8	15.50
T_8	T_2 + FYM (50 kg /tree) + biofertilizers + S + Zn + Fe	7.56	0.25	3.67	311.7	20.65	362.0	15.45
T_9	T_2 + FYM (50 kg/ tree) + biofertilizers + S + Zn + Fe + B	7.58	0.26	3.66	311.0	20.73	364.0	15.48
SE m (±)		0.03	0.02	0.05	2.53	0.55	4.16	0.29
CD at 5 %		NS	NS	NS	7.59	1.65	12.47	0.86

and reduction in K-fixation and release of K due to considerable improvement in soil interaction of organic available K under the influence of FYM + inorganic fertilizers. Considerable improvement in soil interaction of organic available K was also reported with clay with the incorporation of FYM + inorganic fertilizers (Zhang *et al.*, 2015).

The available sulphur in soil after harvesting of Nagpur mandarin was recorded displaying most significant gain (15.50 mg kg^{-1}) with the treatment involving 100 % NPK + FYM (50 kg/tree) + biofertilizers + S + Zn (T_7) , closely followed by treatment 100 % NPK + FYM (50 kg/tree) + biofertilizers + S (T_6) , 100 % NPK + FYM (50 kg/ tree) + biofertilizers + S + Zn + Fe + B (T_9) , 100 % NPK + FYM (50 kg/tree) + biofertilizers + S + Zn + Fe (T_8) and 100 % NPK + FYM (50 kg/tree) + biofertilizers (T_5) , but at par with each other and significantly superior over treatment 100 % recommended dose of N and P + FYM (50 kg/tree) + biofertilizers (T_4). However, significantly lowest S (14.01 mg/kg) was noted with treatment 100 % NPK (T_2). Increased sulphur availability in soil due to addition of organic manure was attributed to greater root proliferation and the increasing activity of sulphur-oxidizing bacteria .Our results are similar to those of Kumar *et al.* (2017) .

CONCLUSION

The best results obtained by adoption of integrated nutrient management involving combined use of FYM, biofertilizers and inorganic fertilizers, along with foliar micronutrient spray to realise holistic soil-plant response in terms of fruit yield , fruit quality and soil fertility status. Such concept is considered close to organic citrus.

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Response of wedge grafting and time on survival of mango *(Mangifera indica)* in polyhouse and open field condition

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Abstract

The experiment was conducted to find out optimum time for wedge grafting in mango *(Mangifera indica* L.*)* on different dates of grafting, viz. 15 November (D_1), 1 December (D_2), 15 December (D_3), 1 January (D_4), 15 January (D_5), 1 February (D_6),15 February (D_7) and 1 March (D_8) under polyhouse condition and open field condition. Wedge grafting gave more yield in March in polyhouse (69.31%) and open field condition (59.70%). There was better response to height of plants in polyhouse condition than open field condition The height of grafted plants was 46.09 cm, length of graft union 4.59cm and stem girth 1.55cm. The time taken for first sprout (9.33 cm), number of sprouts (9.08), length of sprout (21.45), number of leaves / plant (23.30), fresh weight of leaves (27.13), dry weight of leaves (18.15), fresh weight of grafted plant (75.30), dry weight of grafted plant (18.96).

Key words: Wedge grafting, Polyhouse condition, Open field condition, Grafting

Mango (*Mangifera indica* L.) of Anacardiaceae family, is most significant commercial fruit. For establishing permanent mango orchards, veneer grafting is considered the most efficient and cost-effective method (Tewari and Bajpai, 2002). Unlike wedge grafting, veneer grafting only requires one angled cut on the scion (Singh *et al.* 2005; Singh *et al.* 2007 and Ram and Pathak, 2005). However, timing is crucial for successful grafting in mangoes. Choosing the wrong time to graft, especially with fluctuating temperatures and humidity, can lead to failure. The wedge grafting is particularly useful for farmers who need reliable mango plants. In wedge grafting, they cut the scion (branch) at an angle on both sides, forming a wedge shape. Wedge grafting boasts a very high success rate, up to 90-100% (Yadav *et al.,* 2018) Hence, an experiment was conducted to find the effect of suitable time, survival percentage and performance of wedge grafting on mango to ensure availability of superior planting material for commercial cultivation.

MATERIALS AND METHODS

The experiment was conducted on mango in polyhouse and open field condition at Collage of Agriculture, Rewa, Madhya Pradesh, during November 2023 to March 2024.Wedge grafting of mango was carried in eight dates, season, *viz.* 15 November (D_1), 1 December (D_2), 15 December (D_3), 1 January (D_4), 15 January (D_5), 1 February (D_6),15 February (D_7) and 1 March (D_8) under polyhouse and open field conditions. The factorial randomized block design with two replications and eight treatment combinations was used. The grafting was done on 6–8-month old seedling. The seedlings were raised by using the local seeds. The seedlings at 3-4 leaf stage were transplanted to polythene bags (300 gauge thickness) with well-drained medium black soil and FYM (1:1) The healthy and quality seedlings were used for grafting.

The terminal one season old scion shoots which having 15 - 18 cm long of pencil thickness (0.5 - 1.0 cm) with 3 - 4 healthy buds and free from pest and disease were selected for grafting. Selected scions were defoliated on mother plant, one week prior to detachment of apical growing portion of selected shoots was beheaded, which helped in forcing the dormant grafts to swell. The grafts on scion were made ready to start sprouting at the time of grafting. For grafting lower end of selected and detached scion stick was prepared in the form of wedge of about 4-5 cm. The top portion of rootstock was decapitated at 15-20 cm height and then top portion of stem was split vertically about 4-5 cm in length forming "V" shape. The wedge shaped scion was inserted into "V" shaped slit and tied with polythene strip and then placed grafted plants in open field and polyhouse.

RESULTS AND DISCUSSION

The maximum plant height (46.09cm) was found on 15 February in polyhouse condition and in open field condition (43.63 cm) on 1 March. The minimum height (34.52 cm) of grafted plant in polyhouse condition was recorded on 1 December and 15 November (35.36 cm) in open field conditions. There was a significant interaction between polyhouse and open field conditions. Whereas, maximum height of grafted plant (43.54 cm) was on 15 February and minimum height (35.40 cm) of grafted plant on 1 December.

The maximum length (4.59 cm) was found on 15

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February in polyhouse condition and in open field condition (4.54 cm)the minimum length (3.44 cm) of graft union in polyhouse condition and open condition. (3.56 cm) on 15 January the same trend was in interaction between polyhouse condition and open field condition whereas. maximum length of graft union (4.56 cm) on 15 February and minimum length of graft union (3.50 cm) on 15 January.

The maximum graft survival (69.31 %) was found on 1 March in poly house condition and open field condition (59.70 %) on 15 February. The minimum (54.29 %) graft survival in polyhouse condition and open field condition (43.69 %) on 1 January. There was a significant interaction between polyhouse condition and open field condition whereas maximum graft survival (64.20 %) on 1 March and minimum graft survival (48.99 %) on 1 January. The similar findings were reported by Singh *et al.* (2005), Singh *et al.* (2007) and Ram and Pathak (2005).

The maximum steam girth (1.55 cm) was found in polyhouse condition on 15 November and on 1 December (1.30 cm) in open condition. The minimum stem girth (1.07 cm) of grafted plant in polyhouse condition and open field condition (0.90 cm) was recorded on 1 March. A similar trend was found in interaction between polyhouse condition and open field condition, The maximum stem girth (1.40 cm) was on 15 November and minimum (0.98 cm) on 1 March. There results are in close conformity with these of Tewari and Bajpai (2002).

The maximum time taken (11.55 days) for first sprout in polyhouse condition on in 1 January (19.06 days) with open field condition. The minimum time taken for 1 sprout in polyhouse condition (9.33 days) and open field condition (16.90 days) on 1 March. A similar trend was found in interaction between polyhouse and open field conditions, The maximum time taken for first sprout was (15.30 days) on 1 January and minimum time taken for first sprout was (13.11 days) on 1 March. This result in close conformity with those of Singh *et al.* (2005), Sivudu *et al.* (2014), and Visen *et al.* (2010).

The maximum number (9.08) of sprouts in polyhouse condition was recorded on 15 February and on 1 March 5.60 with open condition. The minimum number of sprouts in polyhouse condition (7.15) was recorded on 1 December and on 15 January (3.65) in open condition. A similar trend was found in interaction between polyhouse and open field conditions. The maximum number of sprouts was (7.28) on 15 February and minimum (5.55) on 15 January (Table 1).

The maximum length of sprout per plant in polyhouse condition (21.45cm) was recorded in 1 March and in open condition (17.25 cm) The maximum number of leaves per sprout per plant in polyhouse condition (23.30) and open condition (18.05) in 1 March. The minimum length of sprouts per plant in polyhouse condition (15.20) and in open condition (7.40) on 15 November.

The maximum fresh weight (27.13 g) of leaves in polyhouse condition was recorded on 15 November and (19.77g) in open condition. The minimum fresh weight (13.03g) of leaves in polyhouse condition was recorded on 1 March and open field condition (7.80 g). The maximum dry weight of leaves (18.15 g) in polyhouse condition was recorded on 15 November and (13.13 g) in open field condition. The minimum dry weight of leaves was in polyhouse condition (6.77 g) and open condition (3.21 g) on 1 march.

The maximum fresh weight (75.30g) of grafted plant in polyhouse condition and (55.94g) in open condition 15 November. The minimum fresh weight of grafted plant

Table 1. Effect of growth and phenotypic character of polyhouse and open condition

Treatment	Hight of of grafted plant (cm)	Length of graft union (cm)	Survival (%)	Time taken for first 1^{st} sprout (days)	Number of of sprouts	Leaf area index	Fresh weight of of leaves (g)	Dry weight of leaves (g)	Fresh weight of of grafted plants (g)	Dry weight of grafted plant (g)
D_1	35.35	3.72	59.85	13.18	5.74	6.01	23.45	15.64	65.62	17.00
D_2	35.40	4.41	54.39	13.38	5.61	4.98	21.92	13.77	62.30	15.73
D_3	37.60	4.01	50.34	13.95	6.00	3.63	20.10	11.41	56.63	14.30
D_4	38.64	4.15	48.99	15.30	6.45	2.87	18.32	9.93	53.57	13.76
D_5	41.65	3.50	50.35	14.69	5.94	2.18	16.20	7.90	51.28	12.44
D_6	39.89	4.51	61.29	13.88	6.38	1.72	14.35	6.54	50.25	11.76
D_7	43.54	4.56	62.83	13.32	7.28	1.28	11.84	6.18	46.62	10.54
D_8	41.58	3.89	64.20	13.11	7.24	0.89	10.41	4.99	43.11	9.99
S.Em±	0.68	0.07	1.95	0.23	0.14	0.08	0.55	0.82	0.37	0.17
C.D.(5%)	2.08	0.22	5.60	0.66	0.40	0.27	1.65	2.49	1.14	0.51

was in polyhouse condition (49.47 g) and open condition (36.75 g) on 1 March. The maximum dry weight (18.96 g) of grafted plants in polyhouse condition (15.04 g) and in open condition on 15 November. The minimum dry weight (12.33 g) of grafted plants in polyhouse condition and open condition (7.65 g) on 1 March. The similar results were reported by Sultan and Singh (1984) and Bhatnagar (1962).

CONCLUSION

Thus, it was concluded that wedge grafting in polyhouse condition gave better result than 15 November to 1 March. In open field condition, suitable period for wedge grafting was 1 January to 15 February. The Maximum percentage of graft survival obtained at polyhouse condition was (69.31%) on 1 March and in open field condition (59.70%) on 15 February.

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Evaluation of bottle gourd (*Lagenaria siceraria*) genotypes for growth, yield and quality under Bundelkhand agroclimatic conditions

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Abstract

The experiment was conducted to evaluate 30 bottle gourd (*Lagenaria siceraria* L.) genotypes for their morphological, yield, and quality character at RLBCAU, Jhansi, Uttar Pradesh, during *kharif* 2022. The genotypes were sown in a randomised block design with three replications, and observations were taken on 15 characters. There was a wide range of mean values among genotypes. Hybrid Green Gold (8.0, 6.60kg) and MAHY 8 (7.6, 6.40kg) were statistically superior for maximum number of fruits/plant as well as fruit yield/plant, respectively. The maximum vine length (800cm), total soluble solids (2.93 °Brix) and ascorbic acid content (16.97mg) were recorded in RBG 9 genotype. The minimum day to first flowering bud (26.0 days) and earliest node of first appearance of female flower (10.6) was observed on Pusa Samrudhi. Genotype MAHY 8 was earliest for days to first fruit set (37.3), followed by minimum days to fruit picking (46.0). However, maximum fruit length was recorded in Muskan (46.6 cm), followed by MAHY 8 (45.0cm). The highest fruit width was recorded in Narendra Shishir (12.6cm), and highest fruit weight in Pusa Santushti (0.867g).

Key words: Cucurbits, Evaluation, Germplasm, Productivity, Semi-arid, Bundelkhand

Bottle gourd (*Lagenaria siceraria*) fruits are valued for their culinary versatility, digestibility, and nutritional benefits, particularly in traditional diets. The yield potential of bottle gourd remains comparatively low in such climates, indicating considerable scope for genetic improvement and selection of climate-resilient genotypes (Samadia, 2011). Improving yield and fruit quality under stress-prone conditions is therefore a key breeding objective (Rathore *et al.,* 2025). Given the wide variability inherent in this cross-pollinated crop, systematic evaluation of genotypes is essential to identify promising lines for yield enhancement and quality improvement (Suma *et al.,* 2025). Therefore, study was undertaken to evaluate diverse genotypes for growth, yield, and fruit quality traits.

MATERIALS AND METHODS

The study was carried out from July to November during *kharif* 2022, at Rani Lakshmi Bai Central Agricultural University, Jhansi, Uttar Pradesh. The experimental site is situated at 25.30° N latitude and 78.32° E longitude at an altitude of 227 m above mean sea-level. The experimental material comprised 30 genotypes collected from different sources, CAUs, SAUs, ICAR-Institutions, local collections and private companies. The experiment was laid out in 3m × 3m plot size with a spacing of 0.6 m from plant to plant. The observations were recorded from five randomly selected plants from each genotype for 15 different characters. The statistical analysis of data was done using the analysis of variance approach (Fisher, 1950). The "f" value, or variance ratio, was used to evaluate the treatment effect's significance and non-significance to table value at 5% significance level. If the calculated value is more than the value, the effect is considered to be significant (Panse and Shukhatme, 1967).

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RESULTS AND DISCUSSION

Significant variation was observed among the genotypes for vine length and other associated traits, indicating the presence of considerable genetic variability. The maximum vine length (800 cm) was recorded in RBG 9, followed by IC-594545 (786.7 cm) and RBG 4 (786.7 cm). Whereas, minimum vine length was observed in Pusa Santushti (570 cm), followed by Sharada (593.3cm) and Surag (613.3cm). Vine length may have increased as a result of improved photosynthetic and other metabolic activities, local circumstances, genetic traits, increased nutrient utilization efficiency and an increase in numerous plant compounds that cause cell division and elongation. The genetic make-up of genotypes, as well as intrinsic qualities and vigour of crop may be the cause of variance in vine length. Similar results have been reported by Kritika and Bahadur (2024).

Days to first flowering bud ranged from 26.0 to 39.3 days with mean of 32.2 days. Out of 30 genotypes, the earlier flower bud appeared in Pusa Samrudhi (26.0 days), followed by Pusa Naveen (27.0 days), MAHY 8 (27.0 days) and Mahi (27.0 days). However, maximum days to first flower was taken by both Narendra Shishir (39.3 days) and RBG 10 (39.3 days), followed by RBG 2 (36.6 days) and Narendra Madhuri (36.0 days). Kumar (2021) and (Sharma *et al.,* 2025) also supported these results .

The node number to first male flower appearance ranged from 8.67 to 13.0 with a mean of 10.9. The lowest node number to first staminate flower appearance was observed in IC-594545 (8.67), followed by RBG 5 (9.0) and RBG 12 (9.0). While, highest was recorded in RBG 1 (13.0) and RBG 6 (13.0), followed by Narendra Madhuri (12.6) and Pusa Samrudhi (12.3). The node number in first pistillate flower appearance ranged from 10.6 to 15.3 with a mean of 12.9. The earliest node number to first female flower appearance was observed in Pusa Samrudhi (10.6), followed by IC-594545 (11.0), RBG 3 (11.0) and RBG 13 (11.0) (Tables 1 and 2). Whereas, maximum was recorded for RBG 6 (15.3) and Mahi (15.0),

Table 1: Performance of bottle gourd genotypes for plant traits

Genotype	Days to first flowering bud	First male flower at node	First female flower at node	Days to first fruit setting	Days to first fruit picking	Number of fruits plant	Vine length (cm)
Sharada	27.67	12.00	13.67	43.00	51.67	6.67	593.3
Pusa Santushti	31.00	9.33	10.67	46.33	55.00	6.00	570.0
Amrit F_1	34.00	10.33	13.33	48.00	56.67	7.67	696.7
Narendra Shishir	39.33	11.67	14.00	57.33	65.67	6.33	706.7
Surag	33.33	10.33	12.67	47.00	55.00	7.00	613.3
Madhu Sree	33.33	12.00	13.33	47.00	56.00	7.00	676.7
Pusa Samrudhi	26.00	12.33	13.67	42.00	51.00	7.33	620.0
IC-594545	31.67	8.67	11.00	45.33	54.00	6.67	786.7
RBG-1	31.33	13.00	14.33	45.67	53.67	5.00	680.0
RBG-2	36.67	9.67	11.67	54.33	65.33	5.33	733.3
RBG-3	30.67	9.33	11.00	46.67	55.33	5.33	730.0
RBG-4	30.00	9.33	11.33	46.00	54.00	6.67	786.7
RBG-5	32.67	9.00	11.33	45.33	50.67	4.67	623.3
RBG-6	32.33	13.00	15.33	43.67	52.00	7.00	706.7
RBG-7	27.33	10.67	13.00	45.67	52.33	6.67	650.0
Pusa Naveen	27.00	11.67	13.67	44.00	51.00	5.67	673.3
Hybrid Green Gold	27.33	11.67	13.67	45.67	53.00	8.00	723.3
MAHY 8	27.00	10.33	11.33	37.33	46.00	7.67	736.7
Muskan	30.00	10.00	11.33	44.67	52.33	6.33	776.7
Mahi	27.00	12.67	15.00	40.00	48.00	6.67	640.0
Narendra Madhuri	36.00	12.67	14.67	47.00	60.67	5.33	686.7
Arka Bahar	35.00	10.00	12.00	49.00	64.00	6.00	650.0
RBG-8	30.67	12.00	13.67	43.33	56.67	5.67	683.3
RBG-9	33.67	12.00	14.67	49.33	56.67	5.33	800.0
RBG-10	39.33	10.33	13.00	57.33	66.33	7.67	740.0
RBG-11	36.00	12.33	14.33	48.00	55.67	6.33	716.7
RBG-12	33.33	9.00	11.67	49.67	56.67	6.67	773.3
RBG-13	35.00	9.67	11.00	46.67	54.33	7.33	693.3
RBG-14	36.67	10.00	13.00	50.67	58.67	6.67	750.0
RBG-16	35.00	12.00	14.67	51.67	60.33	6.00	706.7
Mean	32.21	10.90	12.93	46.92	55.62	6.42	697.44
C.V.	3.73	8.58	8.93	3.56	3.59	8.18	5.00
S.E.(m)	0.69	0.54	0.66	0.96	1.14	0.31	20.70
C.D. at 5%	1.96	1.53	1.88	2.73	3.25	0.89	58.61
Maximum	39.33	13.00	15.33	57.33	66.33	8.00	800.00
Minimum	26.00	8.67	10.67	37.33	46.00	4.67	570.00

Table 2: Performance of bottle gourd genotypes for yield and fruits physico-chemical traits

Genotypes	Fruit length (cm)	Fruit width (cm)	Fruit weight (g)	Yield (kg. plant)	Yield (q/ha)	TSS (°Brix)	Protein content (mg)	Ascorbic acid (mg/100g)
Sharada	37.67	6.23	0.800	5.33	281.43	2.03	203.93	13.33
Pusa Santushti	19.33	9.90	0.867	5.17	293.70	2.17	155.13	13.83
Amrit F_1	38.33	6.20	0.767	5.87	325.13	2.20	170.50	15.63
Narendra Shishir	20.00	12.67	0.833	5.30	281.43	2.07	159.33	16.40
Surag	38.33	6.00	0.867	6.07	327.73	2.53	196.33	11.90
Madhu Sree	19.00	10.80	0.767	5.37	326.23	2.47	200.33	12.60
Pusa Samrudhi	37.00	6.87	0.867	6.30	333.67	2.73	218.67	15.17
IC-594545	10.00	12.07	0.867	5.83	287.00	2.37	165.13	16.67
RBG-1	33.33	6.13	0.800	3.93	207.03	2.53	195.33	11.23
RBG-2	42.33	5.33	0.700	3.70	201.07	1.83	182.67	16.40
RBG-3	32.33	5.77	0.700	3.73	199.20	2.03	145.93	16.60
RBG-4	12.33	11.70	0.667	4.57	241.43	2.83	182.33	12.37
RBG-5	19.00	6.50	0.733	3.43	193.67	2.33	173.67	14.23
RBG-6	20.00	11.10	0.633	4.33	244.03	2.13	178.40	13.87
RBG-7	31.67	5.20	0.767	5.10	285.13	1.80	181.30	13.50
Pusa Naveen	30.67	5.80	0.733	4.13	218.47	1.70	185.77	14.33
Hybrid Green Gold	38.33	6.43	0.833	6.60	344.80	2.33	181.65	14.03
MAHY 8	45.00	5.27	0.833	6.40	339.97	2.20	219.67	14.10
Muskan	46.67	4.37	0.733	4.63	246.97	2.40	138.13	16.00
Mahi	41.67	6.40	0.667	4.43	228.87	2.63	184.20	14.57
Narendra Madhuri	15.00	11.70	0.767	4.10	219.20	2.37	213.33	14.60
Arka Bahar	34.00	5.67	0.800	4.77	238.87	2.73	176.20	16.00
RBG-8	30.67	5.67	0.740	4.18	212.93	2.23	174.33	15.17
RBG-9	35.67	5.00	0.667	3.67	221.80	2.93	208.67	16.97
RBG-10	30.67	4.83	0.767	5.90	321.03	2.03	182.67	16.13
RBG-11	34.00	5.40	0.633	4.07	224.80	1.67	156.67	14.33
RBG-12	33.00	4.30	0.633	4.20	229.23	2.10	202.67	16.97
RBG-13	36.67	5.50	0.667	4.93	262.90	2.07	185.00	13.93
RBG-14	33.33	5.53	0.700	4.70	247.37	2.60	182.67	14.40
RBG-16	26.67	5.37	0.667	4.00	214.80	2.77	160.70	13.03
Mean	30.76	6.99	0.75	4.82	260.00	2.29	182.04	14.61
C.V.	7.35	12.24	11.75	14.32	14.32	3.80	4.35	2.5666
S.E.(m)	1.30	0.49	0.05	0.46	25.98	0.05	5.07	0.2132
C.D. at 5%	3.69	1.39	0.16	1.32	73.55	0.15	14.37	0.6034
Maximum	46.67	12.67	0.87	6.60	344.80	2.93	219.67	16.97
Minimum	10.00	4.30	0.63	3.43	193.67	1.67	138.13	11.23

followed by Narendra Madhuri (14.6), RBG 9 (14.6) and RBG 16 (14.6). The difference in flowering time in bottle gourd is also reported by Kappal *et al.* (2015).

Days to fruit set ranged from 37.3 to 57.3 days with a mean of 46.9 days. The minimum days to first fruit setting were recorded for MAHY 8 (37.3 days), followed by Mahi (40.0 days) and Pusa Samrudhi (42.0 days) (Table 1). However, the maximum number of days to first fruit setting was observed in Narendra Shishir (57.3 days) and RBG 10 (57.3 days), followed by RBG 2 (54.3 days). Days to first fruit picking ranged from 46.0 to 66.3 days with a mean of 55.6 days. The minimum number of days to fruit picking was recorded for MAHY 8 (46.0 days), followed by Mahi (48.0 days) and RBG 5 (50.6 days). However, maximum days to first fruit picking were observed in RBG 10 (66.3 days), followed by Narendra Shishir (65.6 days) and RBG 2 (65.3 days).

The number of fruits/plants ranged from 4.6 to 8.0, with a mean of 6.4. The maximum number of fruits/plant observed in Hybrid Green Gold (8.0), followed by MAHY 8 (7.6), and Amrit F_1 (7.6). Whereas, minimum number of fruits/plant recorded in RBG 5 (4.6), followed by RBG 1(5.0), RBG 2 (5.3) and RBG 3(5.3). The average fruit length per plant ranged from 10.0 cm to 46.6 cm, with a mean of 30.7 cm. The maximum fruit length was recorded in Muskan (46.6 cm), followed by MAHY 8 (45.0 cm) and RBG 2 (42.3 cm). Whereas, minimum fruit length was observed in IC-594545 (10.0 cm), followed by RBG 4 (12.3 cm) and Narendra Madhuri (15.0 cm). The average fruit width/plant ranged from 4.3 cm to 12.6 cm, with a mean of 6.9 cm.

The highest fruit width was recorded in Narendra Shishir (12.6 cm), followed by IC-594545 (12.0 cm), RBG 4 (11.7 cm) and Narendra Madhuri (11.7 cm). Whereas, minimum fruit width was recorded in Muskan (4.3 cm), followed by RBG 12 (4.3 cm) and RBG 10 (4.8 cm). The average fruit weight/plant ranged from 0.633 to 0.867 g with a mean of 0.750 g. The highest fruit weight was recorded in Pusa Santushti (0.867 g), IC-594545 (0.867 g) and Pusa Samrudhi (0.867 g). Whereas, lowest fruit weight was recorded in RBG 4 (0.667g), followed by RBG 9 (0.667 g) and RBG 13 (0.667 g).

Fruit yield ranged from 3.43 to 6.60 kg/plant with a mean of 4.82. The highest fruit yield was recorded in Hybrid Green Gold (6.60 kg), followed by MAHY 8 (6.40 kg) and Pusa Samrudhi (6.30 kg) (Table 2). Whereas, minimum fruit yield per plant was recorded in RBG 5 (3.43 kg), followed by RBG 9 (3.6 kg), RBG 2(3.7 kg) and RBG 3 (3.7 kg). Fruit yield ranged from 193.6 to 344.8 q/ha with a mean of 260.0 q. The maximum fruit yield was recorded in Hybrid Green Gold (344.8 q), followed by MAHY 8 (339.9 q) and Pusa Samrudhi (333.6 q). Whereas, minimum fruit yield was recorded in RBG 5 (193.6 q), followed by RBG 3 (199.2 q) and RBG 2 (201.0 q). Differences in fruit yield among genotypes might be due to their genetic makeup and their responses to soil macro- and microclimates. Similar findings were reported by Iqbal *et al.* (2019).

Protein content ranged from 138.1 mg to 219.6 mg with a mean of 182.0 mg. The high protein content was observed in MAHY 8 (219.6 mg), followed by Pusa Samrudhi (218.6 mg) and Narendra Madhuri (213.3 mg). The lowest protein content was observed in Muskan (138.1 mg), followed by RBG 3 (145.9 mg) and Pusa Santushti (155.1 mg). The TSS ranged from 1.67 °Brix to 2.93 °Brix with a general mean of 2.29 °Brix. The highest TSS was in RBG 9 (2.93 °Brix), followed by RBG 4 (2.8 °Brix) and RBG 16 (2.7 °Brix). The lowest value was observed in RBG 11 (1.67 °Brix), followed by Pusa Naveen (1.70 °Brix) and RBG 2 (1.83 °Brix). The ascorbic acid content (mg/100 g) ranged from 11.23 mg to 16.97 mg with a mean of 14.61 mg. The highest value for ascorbic acid content was observed in RBG 9 (16.97 mg) and RBG 12 (16.97 mg), followed by IC-594545 (16.67 mg). However, the lowest value was recorded in RBG 1(11.23 mg), followed by Surag (11.90 mg) and RBG 4 (12.37 mg). The similar findings were recorded by Bhatt *et al.,* (2022), Dubey *et al.,* (2022), Kumar *et al.* (2021), Venkatraman, (2021), Ahmad *et al.,* (2019), Chikkeri *et al. (2018)* and Shree *et al.* (2018) to most of the characters .

CONCLUSION

Thus, Hybrid Green Gold, MAHY 8 and Pusa Samrudhi are recommended for cultivation in the Bundelkhand region and may serve as valuable genetic resources for improvement programmes.

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Conflict of interest

The authors declare no conflict of interest

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Effect of spacing and foliar spray of novel (banana pseudostem -based organic liquid nutrients) on cowpea (*Vigna unguiculata*) production under mango-based agroforestry system

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Abstract

The study was undertaken to check economic possibility of cowpea [Vigna unguiculata (L.) Walp.] under mango-based agroforestry system with various spacings and foliar sprays of novel (banana pseudostem - based organic liquid nutrients) during 2021-22 at ASPEE College of Horticulture, Navsari Agricultural University, Navsari, Gujarat, India. The trial was framed with eight treatment combinations of two factors which contain two levels of spacings and four foliar sprays of fertilizers (after initiation of flowering and 15 days after first spray) under mango orchard and open growing condition in a randomized block design with factorial concept consisting of three replications. Under mango-based agroforestry system and in open condition, highest net realization (₹ 1,57,437/ha and ₹ 1,89,259/ha, respectively) and BCR (2.58 and 1.98, respectively) were obtained in S2F4 [45 cm × 45 cm spacing and novel plus (banana pseudostem - based organic liquid nutrients + botanical pesticides) sprayed at 1 %].

Key words: Economics, Mango, Cowpea, Agroforestry system, BCR

A number of fruit-based agroforestry systems have been developed in almost all the regions of India (Kumar and Chaturvedi, 2017). Integration of annual crops with fruit trees yields multiple outputs that ensures production and income generation in a sustainable manner (Randhawa, 1990). Fruit-based agroforestry system integrates cultivation of vegetable and fruit crops and silvi component. Nowadays, use of organic fertilizers in vegetable production is also increasing by researchers and farmers. Thus, there is a need to generate the information regarding tree-crop interaction with use of organic fertilizers on cowpea under commercial fruit tree canopies. Therefore, study was intended to investigate the effect of various spacing and foliar sprays of novel (banana pseudostem- based organic liquid nutrients) on economics of cowpea under mango-based agroforestry system.

MATERIALS AND METHODS

The field trail was conducted during 2021-22 at ASPEE College of Horticulture, Navsari Agricultural University, Navsari, Gujarat (for mango-based agroforestry system) and College Farm, N. M. College of Agriculture, NAU, Navsari (for open condition). Navsari is geographically situated at latitude of 20° 57' N and longitude of 72° 54' E and at an altitude of about 10 m above the mean sea-level. The soil of south Gujarat is locally known as 'black cotton soil'. The experimental soil was deep moderately drained clayey soils classified as deep black soil predominated with montmorillonite clay mineral by its origin. It is medium in fertility.

The experiment comprised eight treatment combinations of two factors which contains two levels of spacing, *viz.* S_1: 30 cm x 30 cm and S_2: 45 cm x 45 cm and four levels of foliar spray of novel organic liquid fertilizers, *viz.* F_1: control, F_2: novel @ 1 % (banana pseudostem based organic liquid nutrients), F_3: novel prime @ 1 % (banana pseudostem-based organic liquid nutrients + botanical fungicide) and F_4: novel plus @ 1 % (banana pseudostem-based organic liquid nutrients + botanical pesticides) with three replications and analyzed as per randomized block design with factorial concept (Panse and Sukhatme, 1985). The same treatments were applied in both growing conditions, *i.e.* under mango-based agroforestry system and open condition.

Cowpea variety Anand was grown under 10 years old mango orchard planted at 9 m x 9 m spacing. For applying recommended dose of N, P, K (20:40:00 kg/ha) to cowpea, commercial grade of neem coated urea and di-ammonium phosphate (DAP) were used. Novel plus and novel prime (banana pseudostem-based organic liquid nutrients, *i.e.* patented product of NAU) were procured from banana pseudostem unit, soil and water management research unit, NAU, Navsari. Different novel solutions collected,

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were considered as 100% in concentration and used to prepare the required solution according to treatments. Various cultural operations, *viz.* tillage, application of FYM, rotavator, layout, bed preparation, fertilizer application (½ N + full P), application of pre-emergence weedicide, sowing of seeds, irrigation, weeding, hoeing, remaining (½ N) fertilizer application was carried out in both the growing conditions.

For the purpose of economic analysis, yield parameters of cowpea were recorded at each picking and then total of all pickings were averaged out. Various yield and economic parameters were calculated as per the procedure given below: Total weight of pods harvested from the randomly selected and tagged five plants in each net plot was recorded in each picking and summed up. The average weight of pods per plant was multiplied by number of plants in each net plot of each replication. Then it was converted according to the size and number of plants in gross plot. Then, yield per plot was converted into the yield in tonne per hectare for each plot in each replication.

To obtain marketable yield, unmarketable yield (affected by insect and pest) was subtracted from total yield. The cost of cultivation for each treatment was worked out by considering the cost of all operations right from the preparation of land to the harvesting of crop. The price of cowpea vegetable was accounted on the basis of prevailing market rate. The cost of fertilizers is taken as form the current market price. The gross realization in terms of rupees per hectare was worked out on the basis of marketable crop yield. The prevailing market price of inputs and outputs was accounted to calculate the gross returns.

The net returns per hectare was calculated by deducting the cost of cultivation from gross returns per hectare. The benefit cost ratio (BCR) was calculated by an incremental cost of different treatment and benefit obtain through an increase in production due to the respective treatment.The benefit cost ratio was calculated as per following formula:

$$Benefit{:}cost\ ratio = \frac{Net\ realization}{Cost\ of\ cultivation}$$

RESULTS AND DISCUSSION

The maximum total yield (3.73, 4.61 and 4.17 tonnes/ha) respectively in 2021, 2022 and pooled analysis and marketable yield (3.50, 4.36 and 3.93 tonnes/ha, respectively) was obtained in treatment combination of S_2F_4 (45 cm x 45 cm; novel plus @ 1 % (banana pseudostem based organic liquid nutrients + botanical pesticides) under mango-based agroforestry system (Table 1). In open condition, maximum total yield (9.21, 10.75 and 9.98 tonnes/ha) respectively in 2021, 2022 and pooled analysis and marketable yield (8.76, 10.24 and 9.50 tonnes/ha, respectively) were also obtained in the same treatment combination S_2F_4 (45 cm x 45 cm; novel plus @ 1 % (banana pseudostem based organic liquid nutrients + botanical pesticides). Among different treatment combinations, maximum gross realization was obtained from treatment combination S_2F_4 (₹ 2,82,850/ha), followed by S_2F_3(₹ 2,77,100/ha) under mango-based agroforestry system (Table 2). However, minimum gross realization was received from treatment combination of S_1F_1 (₹ 2,41,800/ha).

In open condition, treatment combination S_2F_4 gave highest gross realization (₹ 2,85,000/ ha), followed by treatment combination of S_1F_4 (₹ 2,75,750/ha). The lowest gross realization (₹ 2,03,450/ha) was received from treatment combination of S_1F_1 (30 cm x 30 cm; control) (Table 2). Under mango-based agroforestry system, highest net realization of ₹ 1,57,437/ha was registered in treatment combination of S_2F_4, followed by S_2F_3 (₹ 1,51,687/ha). The lowest net return was obtained with S_1F_1 (₹ 1,14,223/ha).

In open condition, maximum net realization (₹ 1,89,259/ha) on account of cowpea pod production was obtained with treatment combination S_2F_4, followed by S_2F_3 (₹ 1,76,309/ha). However, minimum net return of ₹ 1,03,381/ha was earned from S_1F_1.

Under mango-based agroforestry system, treatment combination of S_2F_4 registered maximum BCR (2.58), followed by S_2F_3 (2.46). However, minimum benefit: cost ratio (BCR) (1.66) was obtained in S_1F_1 treatment combination.

In open condition, highest BCR (1.98) was obtained in treatment combination of S_2F_4 (45 cm x 45 cm spacing and novel plus @ 1 %), followed by S_2F_3 (45 cm x 45 cm spacing and novel prime @ 1 %) with BCR (1.84). The lowest BCR (1.03) was found in S_1F_1 (30 cm x 30 cm spacing and without foliar spray of novel fertilizers). The higher yield, gross realization, net realization and benefit: cost ratio were observed with wider spacing in comparison to closer spacing under mango-based agroforestry system as well as in open condition might be due to less competition for light, moisture and nutrients associated with wider spacing and thereby having an edge in producing more reproductive parts when compared to higher plant population density (Karukonda *et al*., 2020). Similar trend of result is also reported by Shukla and Singh (2021) in cluster bean (*Cyamopsis tetragonoloba*).

The plants which received foliar spray of banana pseudostem-based organic liquid nutrients had produced more yield than those without foliar application. The increase in yield might be due to supply

Table 1: Yield of cowpea as affected by spacing and foliar spray of fertilizers under mango-based agroforestry system and in open condition

Treatment	2021		2022		Pooled	
	Total yield (tonnes/ha)	Marketable yield (tonnes/ha)	Total yield (tonnes/ha)	Marketable yield (tonnes/ha)	Total yield (tonnes/ha)	Marketable yield (tonnes/ha)
Cowpea cultivated under mango						
S_1F_1	2.35	2.17	3.17	2.96	2.76	2.56
S_1F_2	2.56	2.37	3.16	2.94	2.86	2.65
S_1F_3	2.80	2.60	3.44	3.21	3.12	2.91
S_1F_4	3.25	3.04	4.02	3.76	3.64	3.40
S_2F_1	2.59	2.41	3.32	3.11	2.96	2.76
S_2F_2	3.18	2.99	3.83	3.61	3.51	3.30
S_2F_3	3.53	3.32	4.39	4.15	3.96	3.74
S_2F_4	3.73	3.50	4.61	4.36	4.17	3.93
Cowpea cultivated in open condition						
S_1F_1	6.35	6.00	7.97	7.56	7.17	6.78
S_1F_2	7.09	6.71	8.93	8.49	8.01	7.60
S_1F_3	7.95	7.55	9.90	9.44	8.93	8.50
S_1F_4	9.08	8.65	10.25	9.73	9.67	9.19
S_2F_1	6.95	6.58	8.95	8.52	7.95	7.55
S_2F_2	7.77	7.37	9.43	8.97	8.60	8.17
S_2F_3	8.74	8.32	10.29	9.82	9.52	9.07
S_2F_4	9.21	8.76	10.75	10.24	9.98	9.50

Table 2: Economics of cowpea as affected by spacing and foliar spray of fertilizers under mango-based agroforestry system and open condition

Treatment	Marketable yield (tonnes/ha)	Fixed cost (₹/ha)	Variable cost (₹/ha)	Total cost of cultivation (₹/ha)	Gross realization (₹/ha)	Net realization (₹/ha)	BCR	BCR (including mango)
Mango								
Mango	5.50			76825	165000	88175	1.15	
Cowpea under mango-based system								
S_1F_1	2.56	43952	6800	50752	76800	26048	0.51	1.66
S_1F_2	2.65	43952	7986	51938	79550	27612	0.53	1.68
S_1F_3	2.91	43952	8236	52188	87200	35012	0.67	1.82
S_1F_4	3.40	43952	8236	52188	102000	49812	0.95	2.10
S_2F_1	2.76	43952	3200	47152	82800	35648	0.76	1.91
S_2F_2	3.30	43952	4386	48338	98900	50562	1.05	2.20
S_2F_3	3.74	43952	4636	48588	112100	63512	1.31	2.46
S_2F_4	3.93	43952	4636	48588	117850	69262	1.43	2.58
Cowpea in open condition								
S_1F_1	6.78	86469	13600	100069	203450	103381	1.03	-
S_1F_2	7.60	86469	15972	102441	228100	125659	1.23	-
S_1F_3	8.50	86469	16472	102941	254850	151909	1.48	-
S_1F_4	9.19	86469	16472	102941	275750	172809	1.68	-
S_2F_1	7.55	86469	6400	92869	226450	133581	1.44	-
S_2F_2	8.17	86469	8772	95241	245200	149959	1.57	-
S_2F_3	9.07	86469	9272	95741	272050	176309	1.84	-
S_2F_4	9.50	86469	9272	95741	285000	189259	1.98	-

Selling rate of cowpea @ ₹ 30.0/kg   Selling rate of mango @ ₹ 30.0/kg

Table 3: Cost of cultivation of cowpea under mango orchard and open condition (₹/ha)

Description			Rate	Cost (₹)	
	Under Mango	Open condition		Under mango [cultivated area of cowpea is 6,000 m²]	Open condition [cultivated area of cowpea is 10,000 m²]
Preparatory tillage					
Ploughing by tractor with cultivator	2 x 4 = 8 hrs	2 x 8 = 16 hrs	@ ₹ 300/hr	2400	4800
Ploughing by tractor with Rotavator	2.5 hrs	5 hrs	@ ₹ 650/hr	1625	3250
			Total	**4025**	**8050**
Layout and sowing					
Layout, Preparation of beds and channel	5 labours for 2 days	10 labours for 2 days	@ ₹ 268/ labour/day	2680	5360
Seed sowing	10 labours for 1 day	20 labours for 1 day	@ ₹ 268/ labour/day	2680	5360
Gap filling	3 labours for 1 day	5 labours for 1 day	@ ₹ 268/ labour/day	804	1340
			Total	**6164**	**12060**
Manures and fertilizers					
FYM	7.5 tonnes/ha	15 tonnes/ha	@ ₹ 800/ tonne	6000	12000
DAP	43.5 kg	87 kg	@ ₹ 27/ kg	1175	2349
Urea	6.61 kg	13.21 kg	@ ₹ 5.9/ kg	39	78
Expenditure on manures application	5 labours for 1 day	10 labours for 1 day	@ ₹ 268/ labour/day	1340	2680
Expenditure on fertilizer application	2 labours for 1 day	3 labours for 1 day	@ ₹ 268/ labour/day	536	804
			Total	**9090**	**17911**
Intercultural operations					
Weeding	5 labours 3 times	10 labours 3 times	@ ₹ 268/ labour/day	4020	8040
			Total	**4020**	**8040**
Irrigation application					
Irrigations	10 (@ 10 hr for 1 ha)	10 (@ 20 hr for 1 ha)	@ ₹ 40/hour	4000	8000
Irrigation application charges	13 labours	25 labours	@ ₹ 268/ labour/day	3484	6700
			Total	**7484**	**14700**
Plant protection					
Labour for spraying	(2 labours spray^{-1}) 2 days	(2 labours spray^{-1}) 3 days	@ ₹ 268/ labour/day	1072	1608
Pendimethalin 30.0 % EC	1 spray 500 ml ha^{-1}	1 spray 1 litre ha^{-1}	@ ₹ 230/ litre	115	230
Imidacloprid 17.8 % SL	2 spray of 75 ml ha^{-1}	2 spray of 150 ml ha^{-1}	@ ₹ 1,750/ litre	263	525
			Total	**1450**	**2363**
Harvesting and marketing					
Harvesting	(4 labours for 1 day) 7 times	(8 labours for 1 day) 7 times	@ ₹ 268/ labour/day	7504	15008
Uprooting the plants	5 labours for 1 day	10 labours for 1 day	@ ₹ 268/ labour/day	1340	2680
			Total	**8844**	**17688**
			Total fixed cost	**41077**	**80812**
			Interest on working capital @ 7 %	**2875**	**5657**
			Gross total	**43952**	**86469**

of more nutrients at critical stage (flowering and fruit setting). Which ultimately resulted in higher yield, net realization and BCR. The probable reason behind higher yield under novel plus foliar spay might be due to higher content of major and micro nutrients as compared to others (Desai *et al.*, 2016 and Champaneri *et al.*, 2021). Present findings are similar with the results of Kavitha *et al.* (2019), Mandaliya (2021) and Rabade *et al.* (2022) in cowpea; Selvarani *et al.* (2021) and Akshika Bhawariya *et al.* (2022) in cluster bean (*C. tetragonoloba*), Meena *et al.* (2017) in urd bean (*Vigna mungo*), Parikh *et al.* (2020) in turmeric, Sowmya *et al.* (2024) in garlic, Dongre and Choudhary (2024) in guava and Bhatt *et al.* (2025) in potato.

Table 4: Photosynthetically active radiation (µ mol/m²/s) under mango-based agroforestry system and open condition

Treatment	Mango-based agroforestry system			Open condition		
	S_1: 30 cm x 30 cm	**S_2:** 45 cm x 45 cm	**Mean**	**S_1:** 30 cm x 30 cm	**S_2:** 45 cm x 45 cm	**Mean**
F_1: Control	358.24	358.09	**358.16**	1352.58	1307.53	**1330.05**
F_2: Novel (1 %)	344.55	329.81	**337.18**	1313.24	1331.87	**1322.55**
F_3: Novel prime (1 %)	307.45	323.46	**315.45**	1333.33	1286.60	**1309.97**
F_4: Novel plus (1 %)	335.78	353.00	**344.39**	1322.41	1277.06	**1299.73**
Mean	**336.50**	**341.09**		**1330.39**	**1300.76**	
	S	**F**	**SF**	**S**	**F**	**SF**
S.Em. ±	7.77	10.99	15.54	19.61	27.74	39.23
C.D. at 5 %	NS	NS	NS	NS	NS	NS
C.V. %		11.24			7.30	

However, net realization and benefit: cost ratio of cowpea was recorded higher when they were grown in open condition as compared to under mango-based agroforestry system (Table 2). The probable reason for highest net realization and BCR under open growing condition might be due to higher availability of photosynthetically active radiation in open growing condition as compared to mango-based agroforestry system (Table 4). The lower yield under mango-based agroforestry system was probably due to poor photosynthetic capacity of plants as they are not receiving proper sunlight under tree canopies.

CONCLUSION

Under mango-based agroforestry system and open growing condition, highest net realization and BCR were obtained in treatment combination S_2F_4 [45 cm × 45 cm spacing and novel plus (banana pseudostem based organic liquid nutrients + botanical pesticides) @ 1 %], while S_1F_1 [30 cm x 30 cm spacing and without foliar spray of novel fertilizers] recorded least net realization and BCR. Thus, growing of cowpea under mango orchard can provide additional income to mango growers until canopy intermingled.

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Effect of foliar application of organics on growth and yield of coriander (*Coriander sativum*)

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Abstract

A field experiment was conducted to find out the effect of foliar application of organics on growth and yield of coriander (*Coriander sativum* L.) var. GDLC-1 at Regional Horticultural Research Station, ASPEE College of Horticulture, NAU, Navsari, Gujarat during *rabi* season of 2022-2023. The experiment was laid out in randomized block design with three replications and nine treatments, *viz. Panchagavya* @ 1.5 % (T_1), *Panchagavya* @ 3 % (T_2), novel organic liquid nutrients @ 1.5 % (T_3), novel organic liquid nutrients @ 3 % (T_4), vermiwash @ 1.5 % (T_5), vermiwash @ 3 % (T_6), cow urine @ 1.5 % (T_7), cow urine @ 3 % (T_8) and the control (T_9). The foliar spray was given 20 and 45 days after sowing. The foliar application of novel organic liquid nutrients @ 1.5 % (T_3) showed maximum total chlorophyll content (1.64 mg/g and 1.71 mg/g), photosynthetic rate (12.24 μmol m^{-2} s^{-1} and 13.10 μmol m^{-2} s^{-1}), transpiration rate (3.48 m mole m^{-2} s^{-1} and 3.69 m mole m^{-2} s^{-1}) and stomatal conductance (0.21 mol m^{-2} s^{-1} and 0.22 mol m^{-2} s^{-1}) 30 and 50 DAS, respectively. The maximum plant height (39.14 cm and 27.87 cm), number of branches/plant (11.73 and 20.20), petiole length (18.57 cm and 10.07 cm), leaf length (3.93 cm and 3.77 cm) and leaf width (4.05 cm and 3.84 cm) were observed under the same treatment. Among treatments, foliar application of novel organic liquid nutrients @ 1.5 % (T_3) recorded significantly minimum days taken for first cutting (30.00) with maximum fresh weight of herbage/plant (18.47 g and 24.40 g), herbage yield at first and second cutting (9.13 t/ha and 11.70 t/ha) 30 and 50 DAS as well as total fresh weight of herbage (20.83 t/ha) at both the cuttings. The maximum net income of ₹632592.00/ha with a benefit:cost ratio of 6.56 was also calculated with same treatment.

Key words: Foliar, Organics, Growth, Economic return, Yield, Liquid nutrients

Coriander (*Coriander sativum* L.) having chromosome number 2n=2x=22, belongs to family Apiaceae (Umbelliferae). In India, it is grown in Madhya Pradesh, Gujarat, Rajasthan, Assam, West Bengal, Orissa, Uttar Pradesh and Andhra Pradesh (MA FW, 2021). Foliar fertilization or foliar feeding entails the supply of nutrients, plant hormones, stimulants and other beneficial substances in liquid form to plant through leaves and stems. *Panchagavya* an organic input, can act as a growth stimulant and immunity booster. Vermiwash is liquid extract of organic waste material of earthworm culture and it is used as a major nutritive for promoting growth of all green plants (Nath *et al*., 2009). Cow urine contains 95 % water, 2.5 % urea and remaining 2.5 %, a mixture of salts, hormones, enzymes and minerals (Bhadauria, 2002). Novel organic liquid nutrients is a growth booster (Jadhav *et al*., 2014). Since, organic liquid fertilizers prove beneficial in addressing transient nutrient deficiencies and promoting plant growth throughout the season, an experiment was conducted to find out the effect of foliar application of organics on growth and yield of coriander.

MATERIALS AND METHODS

The experiment was conducted on coriander var. GDLC-1 during *rabi* season of 2022-2023 at Regional Horticultural Research Station, ASPEE College of Horticulture, NAU, Navsari, Gujarat, in randomized block design with three replications and eleven treatments, *viz. panchagavya* @ 1.5 % (T_1), *panchagavya* @ 3 % (T_2), novel organic liquid nutrients @ 1.5 % (T_3), novel organic liquid nutrients @ 3 % (T_4), vermiwash @ 1.5 % (T_5), vermiwash @ 3 % (T_6), cow urine @ 1.5 % (T_7), cow urine @ 3 % (T_8) and the control (T_9). The foliar spray was given 20 and 45 days after sowing as per the treatments. The soil of experimental site was dark greyish black type having medium to poor drainage and high water-holding capacity. The experimental plots were prepared by one deep ploughing followed by one harrowing. Flat beds of 2.4 m x 2.4 m size were prepared and seeds were sown at 20 cm x 10 cm spacing during last week of November 2022. The cultural practices and nutrient management (FYM @ 10 t/ha with RDF @ 20:10:00 NPK kg/ha) were carried out as par the recommendations. Observations on different physiological attributes and growth parameters were recorded 30 and 50 days after sowing from five randomly selected plants, whereas yield attributing characters were recorded after harvesting. Statistical analysis of data was done as per Panse and Sukatme (1985).

RESULTS AND DISCUSSION

The foliar application of novel organic liquid nutrients 1.5 % (T_3) recorded maximum total chlorophyll

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content (1.64 mg/g and 1.71 mg/g), photosynthetic rate (12.24 µmol m^{-2} s^{-1} and 13.10 µmol m^{-2} s^{-1}), transpiration rate (3.48 m mole m^{-2} s^{-1} and 3.69 m mole m^{-2} s^{-1}) and stomatal conductance (0.21 mol m^{-2} s^{-1} and 0.22 mol m^{-2} s^{-1}) 30 and 50 days after sowing (Table 1). It might be due to presence of nitrogen, gibberellic acid and cytokinin in novel organic liquid nutrients. Chlorophyll component is made up from nitrogen and it is functioning in promoting vegetative growth and green colouration of plant foliage. Nitrogen involve in the formation of chlorophyll which lead to an effective photosynthesis rate of plant (Yadav *et al.*, 2013). The transpiration and photosynthetic rate might be due to opening of stomata to escape the water at the time of transpiration. Simultaneously, exchange of gases crucial for occurrence of photosynthesis. Increase in photosynthetic rate might be due to iron (Fe) which is highly associated with chlorophyll synthesis which later on boosted up to the photosynthesis (Pinal *et al.*, 2017). Value of stomatal conductance showed the passage rate of carbon dioxide (CO_2) entering the leaf stomata and value of water vapour exiting through stomata.

The foliar application of novel organic liquid nutrients at 1.5 % (T_3) gave maximum plant height (39.14 cm and 27.87 cm), number of branches/plant (11.73 and 20.20), petiole length (18.57 cm and 10.07 cm), leaf length (3.93 cm and 3.77cm) and leaf width (4.05 cm and 3.84 cm) 30 and 50 days after sowing. This might be due to nitrogen which present in novel organic liquid fertilizer increased the rate of vegetative growth, which resulted in maximum leaves and leaf area. Moreover, nitrogen increased the cation exchange capacity of plant roots and that make them efficient in absorbing other nutrients ions like phosphorus, potassium *etc.* It also accelerates the synthesis of chlorophyll, proteins and amino acids, which are essential components of the major photosynthesis process in plants. Spraying of novel organic liquid nutrients, which contain plant growth regulators such as NAA, gibberllic acid, cytokinin, along with a balanced composition of macronutrients and micronutrients. This formulation effectively stimulates cell elongation, contributing to a notable enhancement in plant growth. The same effects in vegetative growth have also been reported by Parikh *et al.* (2023).

The foliar application of novel organic liquid nutrients at 1.5 % (T_3) recorded maximum yield 30 and 50 days after sowing. The minimum days taken for first cutting was 30.00 days and with maximum fresh weight of herbage/plant (18.47 g and 24.40 g), herbage yield first and second cutting (9.13 t/ha and 11.70 t/ha) 30 and 50 days after sowing with herbage yield at both cuttings (20.83 t/ha) (Table 2). The higher yield per plant and total yield might be due to higher production of dry matter, height of plant and branches produced per plant. All these factors are very closely related to crop yield. The other reasons may be effect of novel organic liquid nutrients which contain macro and micronutrients. The nutrients N and K at higher rate exerted a significant positive influence on yield. The enhancement of yield could have been further facilitated

Table 1: Effect of foliar application of organics on physiological parameters and growth attributes of coriander

Treatment	Total chlorophyll content (mg/g)		Photosynthetic rate (µmol $m^{-2}s^{-1}$)		Transpiration rate (m mole $m^{-2}s^{-1}$)		Stomatal conductance (mol m^{-2} s^{-1})		Plant height (cm)		Number of branches/plant		Petiole length (cm)		Leaf length (cm)		Leaf width (cm)	
	30 DAS	50 DAS	30 DAS	50 DAS	30 DAS	50 DAS	30 DAS	50 DAS	30 DAS	50 DAS	30 DAS	50 DAS	30 DAS	50 DAS	30 DAS	50 DAS	30 DAS	50 DAS
T_1	1.39	1.45	9.39	9.47	2.70	2.88	0.16	0.17	35.40	24.28	9.73	17.53	15.74	8.19	3.19	3.24	3.49	3.41
T_2	1.41	1.59	10.08	10.28	2.81	2.93	0.16	0.18	34.21	23.23	10.13	18.07	16.84	8.37	3.32	3.45	3.52	3.45
T_3	1.64	1.71	12.24	13.10	3.48	3.69	0.21	0.22	39.14	27.87	11.73	20.20	18.57	10.07	3.93	3.77	4.05	3.84
T_4	1.47	1.61	10.46	11.06	2.92	2.98	0.17	0.18	34.95	24.05	9.47	17.40	18.06	8.59	3.39	3.53	3.69	3.48
T_5	1.59	1.66	10.88	11.24	3.17	3.21	0.17	0.19	35.74	25.15	9.80	17.93	16.99	8.82	3.42	3.35	3.78	3.59
T_6	1.62	1.66	11.71	12.22	3.34	3.56	0.20	0.21	37.00	25.69	10.73	18.60	18.19	9.47	3.62	3.65	3.93	3.72
T_7	1.21	1.44	8.66	8.80	2.45	2.67	0.15	0.16	32.86	22.40	8.80	16.80	15.47	7.69	3.13	3.24	3.39	3.36
T_8	1.35	1.52	9.81	9.74	2.69	2.84	0.16	0.17	33.83	23.11	9.27	17.27	16.87	8.33	3.24	3.37	3.58	3.45
T_9	1.10	1.21	8.29	8.31	2.33	2.51	0.13	0.16	30.61	21.74	8.67	14.53	15.03	7.33	3.07	3.16	3.23	3.11
S.Em. (±)	0.05	0.06	0.40	0.39	0.14	0.13	0.01	0.01	1.23	1.13	0.51	0.74	0.49	0.42	0.17	0.12	0.15	0.13
CD (5 %)	0.16	0.18	1.19	1.17	0.42	0.39	0.02	0.03	3.68	3.40	1.54	2.21	1.47	1.26	0.50	0.36	0.45	0.38
CV (%)	6.59	6.76	6.73	6.43	8.43	7.48	8.05	8.00	6.10	8.12	9.04	7.27	5.03	8.50	8.53	6.16	7.19	6.29

Table 2: Effect of foliar application of organics on yield and economics of coriander var. GDLC-1

Treatmen	Days taken for first cutting	Days taken for second cutting	Fresh weight of herbage/plant (g)		Herbage yield (t/ha)			Cost of production (₹/ha)	Cost of treatments (₹/ha)	Fixed cost (₹/ha)	Total cost (₹/ha)	Gross income (₹/ha)	Net income (₹/ha)	BCR
			30 DAS	50 DAS	30 DAS	50 DAS	Both cuttings							
T_1	31.33	51.67	13.47	20.13	7.07	9.77	16.84	40910	11800	38311	91021	589400	498379	5.48
T_2	31.33	51.00	15.20	21.60	7.43	10.80	18.23	40910	22200	41473	104583	638050	533467	5.10
T_3	30.00	50.33	18.47	24.40	9.13	11.70	20.83	40910	8160	47388	96458	729050	632592	6.56
T_4	31.00	52.00	15.67	21.33	7.97	10.50	18.47	40910	14920	42019	97849	646450	548601	5.61
T_5	30.67	51.33	16.47	21.40	8.03	10.67	18.70	40910	4520	42543	87973	654500	566528	6.44
T_6	30.67	51.00	17.33	22.20	8.80	11.23	20.03	40910	7640	45568	94118	701050	606932	6.45
T_7	32.33	52.33	13.60	17.20	6.80	8.47	15.27	40910	2440	34739	78089	534450	456361	5.84
T_8	32.33	52.67	14.07	19.27	7.03	9.77	16.80	40910	3480	38220	82610	588000	505390	6.12
T_9	33.67	52.33	12.33	16.73	6.50	8.37	14.87	40910	0	33829	74739	520450	445711	5.96
S.Em.±	0.68	1.10	0.70	0.88	0.44	0.62	0.72							
C.D. at 5 %	2.04	NS	2.09	2.64	1.33	1.87	2.15							
C.V. %	3.74	3.70	7.97	7.45	10.08	10.64	6.99							

Selling price of coriander: 35 ₹/ kg

by optimizing the synthesis of carbohydrates and their efficient translocation to the potential storage organs through better growth and more number of branches per plant. This process also facilitates the ready availability of essential nutrients, leading to enhanced photosynthetic activity and increased the yield. Similar findings were also reported by Patil *et al.* (2017) and Vashi *et al.* (2022).

The higher net realization and maximum benefit:cost ratio, ₹ 632592.00/ha and 6.56, respectively were recorded under 1.5% novel organic liquid nutrients (T_3). This might be due investment cost was less and yield was higher in this treatment which gives higher benefit cost ratio. This finding is in agreement with Parikh *et al.* (2023).

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Assessment of genetic diversity among tomato (*Solanum lycopersicum*) genotypes under salt-affected conditions

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Abstract

The experiment was conducted in a randomized block design with 38 genotypes (including three checks) of tomato (*Solanum lycopersicum* L.), in three replications for thirteen quantitative traits. The analysis of variance showed that mean squares due to genotypes were significant for all the characters. Based on the Mahalanobis D2 statistics, 38 genotypes were grouped into 15 clusters. The maximum number of genotypes (9) were grouped into Cluster I. The highest inter-cluster distance was observed between Cluster XI and Cluster VI, indicating that crossing genotypes from these two clusters is likely to produce highly heterotic and diverse segregants. Highest per cent contribution towards clustering of genotypes were observed in total soluble solids, followed by unmarketable fruit yield/plant, average fruit weight and equatorial diameter of fruits.

Key words: Cluster, Divergence, Distance, Self-affected conditions, Genotypes, Variability

Tomato (*Solanum lycopersicum* L.), a member of family Solanaceae, is warm season, self-pollinated vegetable crop. It is grown all over the world with wider consumption both in raw or in processed forms (Jethava *et al*., 2024; Kumar *et al.* 2024).The information on genetic diversity, its nature and degree are useful for selecting desirable parents from a germplasm for the successful breeding programme (Sarawg *et al*., 2007; Prakash *et al*. (2019); Thakur *et al*. (2025). In general, genetically diverse parents are expected to produce hybrids with greater vigor and yield potential. Hence, it necessitates study of genetic divergence among existing varieties and germplasm collection for identification of parents for hybridization programme (Singh *et al*.,2023; Neha *et al.,* 2025). D^2 statistics developed by (Mahalanobis 1936) provides a measure of magnitude for divergence between two genotypes. Keeping in view, present study was undertaken to assess their utility in developing heterotic combinations for commercial use.

MATERIALS AND METHODS

The experiment was conducted at Department of Vegetable Science, Acharya Narendra Deva University of Agriculture and Technology, Narendra Nagar (Kumarganj), Ayodhya, Uttar Pradesh, India, during *rabi* 2019.Thirty-eight genotypes including three checks (ArkaVikas, KashiAman and DVRT-2) were used in arandomized block design with three replications. Each genotype consisted of tworow spaced 60 cm apart with plant-to-plant spacing of 50 cm. The soil has high pH (8.5-10), concentrations of exchangeable sodium ion (>15%) and electrical conductivity (EC) less than 4 ds/cm. Observations were recorded for days to 50% flowering, plant height (cm), number of locules per fruit, pericarp thickness (mm), polar diameter of fruit (cm), equatorial diameter of fruit (cm), number of fruits per cluster, average fruit weight (g), number of fruits per plant, marketable fruit yield per plant (kg), unmarketable fruit yield per plant (kg), total fruit yield per plant (kg), and total soluble solids (°Brix). Mahalanobis D^2 statistics and genotypes were grouped into different clusters following Tochers method as described by (Rao 1952) and (Mahalanobis 1936).

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RESULTS AND DISCUSSION

The 38 tomato genotypes were grouped into 15 distinct non-overlapping clusters (Table 1). Cluster I contained the highest number of genotypes (9), followed by Cluster III (5) and Cluster IV (4). Clusters II, V, VI, VII, VIII, IX, and X each included two genotypes, while Clusters XI to XV comprised a single genotype each. This indicated presence of considerable diversity in genotypes. The major clusters reveal genetic divergence analysis. Although the genotypes of same origin or geographic region were also found to be grouped simultaneously in the same cluster. The instance of grouping of genotypes of different origin or geographic region in the same cluster were frequently observed. Thus, there is no parallelism between genetic and geographic diversity.

The minimum intra-cluster distance (0.00) was found for cluster XI, XII, XIII, XIV, XV and maximum for cluster X (167.17), followed by cluster IX (148.68), cluster I (148.29), cluster III (138.85), cluster VIII (132.41) and cluster IV (124.24) (Table 2). The maximum inter-cluster distance was found between cluster XI to VI (1745.89), followed by cluster VI to V (1602.37), cluster XIII to V (1532.42), cluster XIII to XI (1452.04), cluster XIV to XIII (1284.15), cluster X to XI (1253.19), cluster VIII to V (1215.37), cluster XIV to XI (1192.74), cluster XV to II (1143.93), cluster XIV to II (1031.96), cluster XV to VI (1018.13), cluster XV to XIII (1007.25) and cluster IX to VI (957.58) were very high.

The minimum inter-cluster D^2 value was found in cluster XIV to V (191.73), followed by cluster XI to V (195.90), cluster XIII to IX (211.19), cluster XIV to IV (220.65), cluster XII to VII (221.65), cluster VIII to VI (237.18), cluster X to IV (243.89), cluster VII to II (244.77), cluster VII to IV (250.84), cluster V to IV (254.30), cluster XII to I (255.56), cluster VIII to I (258.31) and cluster IX to III (261.97).

Lower inter-cluster values between clusters suggested that genotypes of the clusters were not much genetically diverse from each other, while higher inter-cluster distance indicated more genetic divergence between genotypes of those clusters. These results are in close conformation with those of Naveen *et al.* (2018),Kiran*et al.*(2020), Limbani*et al.* (2020),Yadav*et al.*(2020).

The cluster means for different traits indicated considerable differences between clusters. The entire cluster from cluster I to cluster XV had average mean performance for most of the characters, exhibiting extreme cluster mean values for none of the characters (Table 3). Cluster V to XV had in general medium mean performance for most of the characters, showing extreme cluster means for none of the characters.

Cluster V showed maximum mean value (5.22) for pericarp thickness, cluster VI maximum mean value (7.08) for total soluble solids. Cluster X showed maximum mean values(23.95) for number of fruits/plant and (124.23) for unmarketable fruit yield/plant. Cluster XI showed maximum mean values (89.10) for plant height and (6.16) for locules/fruit. Cluster XII showed maximum mean value (4.72)for fruits/clusters. Cluster XIV showed maximum mean value (50.56)for days to 50% flowering. Cluster XV showed maximum mean values (7.65)for polar diameter of fruit, equatorial diameter of fruit (8.68), average fruit weight (90.63), marketable fruit yield/plant (1478.14) and total fruit yield per plant (1550.13). Cluster VII had not showed maximum mean value for any character but it had showed minimum mean values (4.16) for polar diameter of fruit and total soluble solids (5.48).

Cluster XII showed minimum mean values (4.99)for equatorial diameter of fruit, average fruit weight (32.83), number of fruits per plant (11.61), marketable fruit yield/plant (423.39), unmarketable fruit yield/plant (32.12) and total fruit yield/plant (455.51). Cluster XIII showed minimum mean values for days to 50% flowering (38.65),

Table 1: Clustering pattern of 30 genotypes of tomato on the basis of Mahalanobis' D^2 statistics

Cluster number	No. of genotypes	Genotypes
I.	9	NDT-3, DVRT-2, NDT-5-2, ArkaVikas, 12345, 2013/TODVAR-1, WT, 2013/TODVAR-5, S5Xndt-3-2-1-1-1
II.	2	NDT-8, 2015/TODINDVAR-1
III.	5	NDT-5-3-1-1, WT-1-1, 2012/TLCVRes.-7-1, WT-1-2, KashiAman
IV.	4	NDT-3-1-2, S5XNDT-3-2-1-1, 2013/TODVAR-2-2-1-1, NDTH-11W-17-1-3
V.	2	NDT-2-1-1, NDTH-11W-22-1-1-2
VI.	2	NDT-5-3-1-2, NDTH-11W-22-1-2-1
VII.	2	NDT-5-1-2-1, S5XNDT-3-2-1-1-2
VIII.	2	NDT-2-3, NDT-5-1-2-2
IX.	2	UtkalKumari, NDT-2-1
X.	2	NDT-3-1-1, S5XNDT-3-2-2-1
XI.	1	NDTH-11W-22-1-2-2
XII.	1	2013-TODVAR/-2-2-2
XIII.	1	Local collection-1
XIV.	1	NDTH-11W-8-2-1
XV.	1	3535

Table 2: Average on intra and inter- clusters D^2 values for 15 clusters in tomato

Cluster number	I	II	III	IV	V	VI	VII	VIII	IX	X	XI	XII	XIII	XIV	XV
I.	**148.29**	364.37	274.17	352.61	711.77	371.36	279.63	258.31	352.92	552.34	743.04	255.56	326.30	571.53	546.71
II.		**59.22**	748.49	539.14	893.98	745.92	244.77	569.62	795.34	334.00	696.17	483.16	556.27	1031.96	1143.93
III.			**138.85**	292.78	610.64	524.35	408.98	307.25	261.97	719.50	789.75	278.39	567.19	342.27	484.55
IV.				**124.24**	254.30	933.14	250.84	598.06	243.89	473.75	286.76	324.59	925.75	220.65	528.46
V.					**86.08**	1602.37	461.69	1215.37	364.55	655.81	195.90	567.72	1532.42	191.73	694.81
VI.						**106.52**	827.70	237.18	957.58	1253.19	1745.89	566.71	302.48	1192.74	1018.13
VII.							**107.35**	511.26	388.53	319.64	450.98	221.65	623.92	456.84	830.34
VIII.								**132.41**	601.31	768.08	1249.29	409.56	211.19	876.80	751.57
IX.									**148.68**	606.63	455.73	431.32	792.53	298.62	286.86
X.										**167.17**	366.07	718.18	783.38	904.60	993.88
XI.											**0.00**	781.13	1452.04	487.63	833.98
XII.												**0.00**	621.39	321.04	870.36
XIII.													**0.00**	1284.15	1007.25
XIV.														**0.00**	664.58
XV.															**0.00**

Table 3: Intra-cluster group means for 13 characters in tomato

Character	Days to 50% flowering	Plant height	Locules per fruit	Pericarp thickness	Polar diameter	Equatorial diameter	Fruits Per cluster	Average Fruit weight	Numbers of fruits Per plant	Marketable yield Per plant	Unmarketable yield Per plant	Total fruit yield per plant	TSS
	44.02	64.04	4.51	4.27	5.39	6.06	3.68	56.40	16.34	886.22	67.10	953.32	6.45
	39.19	71.62	5.25	4.85	4.64	5.60	4.57	43.71	22.89	934.68	117.94	1052.61	6.39
	45.59	70.31	3.80	3.75	6.33	6.14	3.55	57.38	13.85	748.16	45.96	794.12	6.02
	46.04	86.85	5.43	4.12	6.20	6.93	3.81	64.42	14.60	889.33	79.51	968.84	5.83
	47.38	84.44	5.09	5.22**	6.55	7.22	4.67	79.88	18.75	1385.57	77.18	1462.76	5.64
	47.20	70.75	4.31	5.01	5.45	5.50	3.02	45.87	16.80	734.38	44.40	778.78	7.08**
	41.48	66.82	4.85	3.77	4.16*	5.24	3.67	35.97	21.74	724.58	70.70	795.27	5.48*
	45.49	58.43	4.94	2.62	5.96	6.32	4.19	44.25	17.23	708.32	62.99	771.30	6.59
	43.95	60.28	3.39	3.64	6.60	7.28	4.02	78.10	14.20	1022.97	70.11	1093.08	6.05
	41.41	61.93	5.75	2.28	5.44	5.56	4.20	56.28	23.95**	1233.37	124.23**	1357.60	5.81
	42.66	89.10**	6.16**	3.39	6.40	7.16	4.36	87.19	15.42	1315.29	120.59	1435.88	5.81
	41.50	63.60	5.64	5.05	4.74	4.99*	4.72**	32.83*	11.61*	423.39*	32.12*	455.51*	5.65
	38.65*	45.53*	2.79*	1.95*	4.91	5.29	3.83	46.27	17.66	586.70	66.39	1053.58	6.88
	50.56**	86.90	5.57	4.10	6.34	7.01	4.49	68.95	14.88	990.25	35.22	1025.47	5.60
	40.69	57.17	4.86	4.91	7.65**	8.68**	2.85*	90.63**	17.27	1478.14**	71.99	1550.13**	6.72

plant height (45.53), locules per fruit (2.79) and pericarp thickness (1.95). Cluster XV showed minimum mean value for fruits/cluster (2.85).These results are in close conformation with those of Rojalin *et al.*(2015), Yadav*et al.*(2020) and Srinivasulu*et al.* (2020).

The highest contribution to genetic divergence among genotypes was observed for total soluble solids (25.30%), followed by unmarketable fruit yield per plant (16.63%), average fruit weight (9.88%), equatorial fruit diameter (9.44%), and polar fruit diameter (8.25%). The remaining traits like plant height (7.12%), total fruit yield per plant (6.36%), marketable fruit yield per plant (5.40%), pericarp thickness (5.14%), number of fruits per cluster (3.97%), locules per fruit (1.69%), and days to 50% flowering (0.83%) showed comparatively low contribution to total divergence (Table 4). These findings are in close agreement with the results of Jogi *et al.* (2018), Dhyani *et al.* (2019), Prabakaran *et al.* (2019), and Kiran *et al.* (2020). The crosses between entries separated by large inter-cluster distance and having high cluster mean values for one or other character will be helpful in improvement of tomato.

Table 4. Per cent contribution of 13 characters towards total genetic divergence in tomato

Character	Contribution (%)
Days to 50% flowering	0.831
Plant height	7.115
Locules per fruit	1.687
Pericarp thickness	5.140
Polar diameter	8.251
Equatorial diameter	9.444
Fruit per cluster	3.969
Average fruit weight	9.876
Fruit per plant	0.000
Marketable fruit yield per plant	5.399
Unmarketable fruit yield per plant	16.628
Total fruit yield per plant	6.361
TSS	25.298

CONCLUSION

The formation of distinct clusters with considerable inter-cluster distances indicates the presence of substantial genetic variability, offering greater opportunities for heterosis breeding and selection. The maximum inter-cluster distance was observed between Cluster XI and Cluster VI, suggesting that crossing genotypes from these clusters is likely to generate highly heterotic hybrids and diverse segregating populations.

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Assessment of liquid bio-botanicals on growth and yield of okra (*Abelmoschus esculentus*) under south- eastern region of Rajasthan

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Abstract

A field experiment was conducted during *kharif* seasons of 2022 and 2023, at Agricultural Research Station, Ummedganj, Kota, Rajasthan, to evaluate the effects of various liquid bio-botanicals on growth and yield of okra (*Abelmoschus esculentus* L.) under the agroclimatic conditions of south- eastern Rajasthan. The randomized block design with three replications and ten treatments: control (T_0), vermiwash spray (10%) at 30, 45, and 60 days after sowing (DAS) (T_1), jeevamrut (500 L/ha) at sowing and at 30, 45, and 60 DAS (T_2), cow urine spray (10%) (T_3), panchagavya spray (5%) (T_4), amritpaani (500 L/ha) (T_5), and combinations was used. The integrated application of jeevamrut and amritpaani (500 l/ha each at sowing), along with foliar sprays of panchagavya (5%) and vermiwash (10%) at 30, 45, and 60 DAS, significantly enhanced plant height, number of leaves, and number of branches compared to the control. This treatment also reduced days to first picking and improved fruit length, fruit weight and number of pickings. Further, it recorded higher gross returns, net returns, and benefit-cost ratio over other treatments. Thus, the integrated use of liquid bio-botanicals is a promising strategy for improving okra productivity and farm profitability under organic production systems in southeastern Rajasthan.

Key words: Bio-botanicals, Jeevamrut, Panchagavya, Vermiwash, Amritpaani, Organic farming

Okra (*Abelmoschus esculentus* L.), belonging to the family Malvaceae, is an important vegetable crop widely cultivated in tropical and subtropical regions. The crop possesses a chromosome number of 2n = 130 and is believed to have originated in Ethiopia. In India, okra is one of the most economically significant vegetable crops due to its wide adaptability, high nutritional value, and continuous market demand. Organic farming systems generally depend on bulky organic manures. The liquid organic formulations have recently gained considerable attention in organic farming. They promote growth and yield, and contribute to improving soil quality and microbial activity, thereby strengthening the sustainability of organic production systems (Mahanta and Dhar, 2021).The FYM plays a crucial role in maintaining soil fertility, improving soil physical properties, and enhancing microbial activity in alternative agricultural systems (Jarvan *et al*., 2017). Panchagavya, has positive influence on crop growth, yield, and overall plant health (Tharmaraj *et a*l., 2011; Vallimayil and Sekar, 2012).

Similarly, jeevamrut enhances soil microbial activity and improves nutrient availability to crops, thereby supporting plant growth and productivity. Vermiwash acts as a bio-stimulant that promotes crop growth, enhances plant vigour, and improves resistance to environmental stresses (Shivasubarmanian and Ganeshkumar, 2004). Likewise, amritpani has been reported to exhibit synergistic effects with beneficial microorganisms such as mycorrhiza and organic formulations like panchagavya, improving crop performance and overall productivity (Sakubai *et al.,* 2014).Therefore, an experiment was conducted to evaluate the effect of different liquid bio-botanicals on yield of okra under organic production conditions.

MATERIALS AND METHODS

The field experiment was conducted during the *kharif* seasons of 2022 and 2023 at the Agricultural Research Station, Ummedganj, Kota, Rajasthan, India. The experimental site is located at 25°11′ N latitude and 75°50′ E longitude and falls under Agro-Climatic Zone V (Humid South-Eastern Plain) of Rajasthan and Zone VIII (Central Plateau and Hills) of India. The okra variety 'Pusa Bhindi-5' was used as the test crop in both years. The soil of the experimental field was medium clay loam, deep, well-drained, and slightly alkaline in reaction with a pH of 7.70. Farmyard manure (FYM) containing approximately 0.5% N, 0.2% P, and 1.0% K was applied and incorporated into the soil 15 days before sowing of okra seeds. Jeevamrut was applied at a rate of 500 L/ha at the time of sowing along with the first irrigation. Liquid organic formulations were applied to the crop at 30, 45, and 60 days after sowing (DAS) as per the treatment schedule.

The experiment was conducted in a randomized block design (RBD) consisting of ten treatments with three replications, with each experimental plot comprising

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five rows. The data recorded for different growth and yield parameters were subjected to statistical analysis to determine the significance of treatment effects.

Panchagavya solution (5%) was prepared by diluting 500 ml of well-fermented and filtered Panchagavya in 10 L of water. The prepared solution was applied as a foliar spray at 30, 45, and 60 days after sowing (DAS) according to the respective treatments. The spray solution was applied uniformly using a knapsack sprayer at a rate equivalent to 500 L/ha. Jeevamrut was applied to the soil at the time of sowing at a rate of 500 L/ha. The application was carried out when the soil was adequately moist to facilitate better microbial activity and nutrient availability. Jeevamrut was also applied along with irrigation water in the okra crop as per the treatment schedule. Amritpani was applied as a soil drench at 30, 45, and 60 DAS along with irrigation water to ensure proper infiltration and distribution in the root zone. Vermiwash solution (10%) was prepared by diluting 1000 ml of filtered vermiwash in 10 L of water. The solution was applied as a foliar spray at 30, 45, and 60 DAS using a knapsack sprayer, according to the treatment combinations in the okra crop. All foliar applications were carried out during the morning hours to ensure maximum absorption and to minimize evaporation losses.

To determine the initial fertility status of the experimental soil, composite soil samples were collected prior to sowing from random locations across the field (at least five spots) at a depth of 0–15 cm using a screw-type soil auger. The collected samples were analyzed to determine the physico-chemical properties of the soil. The data were recorded on plant height (cm), number of leaves per plant, and number of branches per plant at 20, 40, and 60 DAS, as well as at the final harvest. Yield attributes such as fruit length (cm), fruit weight (g), days to first picking, and number of pickings were also recorded.

The pooled data were analyzed using the standard analysis of variance (ANOVA) procedure applicable to a randomized block design. The significance of variation among treatments was tested using the F-test at the appropriate probability level. Wherever the treatment effects were found to be significant, the critical difference (CD) was calculated to compare the treatment means. The statistical analysis was performed following the methods described by Panse and Sukhatme (1985).

RESULTS AND DISCUSSION

A analysis of data on plant height and the number of leaves per plant, influenced by various liquid organic manures, revealed significant enhancements in these parameters at different growth stages (20, 40, and 60 days after sowing) (Table 1). This improvement was particularly notable with the application of T_9 (jeevamrut @500 litres/ha at sowing + amritpaani @500 litres/ha at sowing + foliar spray of panchagavya 5% + foliar spray of vermiwash 10% at 30, 45, and 60 DAS). However, at the last harvest stage, there was no significant difference among all treatment groups, although there was a marked increase compared to the control. In case of the number of branches per plant, significant changes in among the treatments and as well as from control treatment was recorded at 20, 40, and 60 DAS and at the final harvest.

A critical analysis of the data on fruit length, fruit weight (g), days taken to picking and number of pickings as influenced by various liquid organic manures, revealed significant enhancements in these parameters in 2022, 2023 and pooled analysis (Fig. 1). This improvement was particularly notable with the application of T_9 (jeevamrut @500 litres/ha at sowing + amritpaani @500 litres/ha at sowing + foliar spray of panchagavya 5% + foliar spray of vermiwash 10% at 30, 45, and 60 DAS).

The influence of liquid organic manures on economic parameters was found to be significant across both years of study (2022 and 2023) as well as in the pooled analysis. The highest pooled values for gross return, net return,

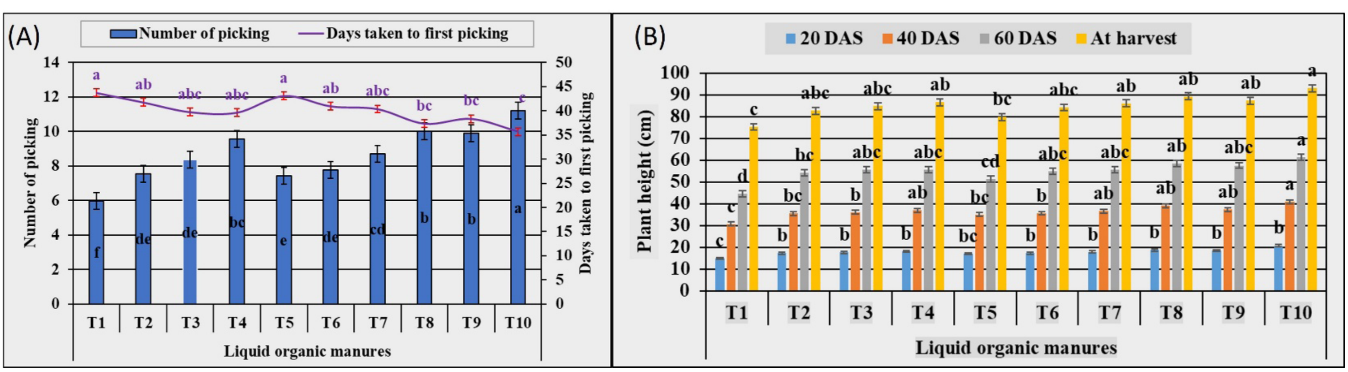

Fig. 1: Effect of different liquid organic manure treatments on (A) days taken to first picking and number of pickings in okra, and (B) height of okra at different stages

Tabel 1. Effect of liquid organic manures on the economics of various treatment combinations used in okra

Treatment	Cost of cultivation (₹/ha) (A)			Estimated yield (q/ha) (B)			Gross return (₹/ha) C= (B×price*)			Net return (₹/ha) D= (C-A)			Net return due to treatment combinations (₹/ha) (E)			B:C ratio F= (D/A)		
	2022	2023	Average	2022	2023	Pooled	2022	2023	Pooled	2022	2023	Pooled	2022	2023	Pooled	2022	2023	Pooled
T_0	60715	64315	62515	77.41	79.53	78.47	154813	174972	164892	94098	110657	102377	0	0	0	1.55	1.72	1.64
T_1	63415	67315	65365	82.20	87.12	84.66	164406	191667	178036	100991	124352	112671	6892	13695	10294	1.59	1.85	1.72
T_2	61865	65565	63715	90.88	94.10	92.49	181769	207014	194391	119904	141449	130676	25805	30792	28299	1.94	2.16	2.05
T_3	64165	68065	66115	98.46	101.98	100.22	196924	224353	210638	132759	156288	144523	38661	45631	42146	2.07	2.30	2.18
T_4	62815	66715	64765	80.92	83.35	82.13	161832	183367	172599	99017	116652	107834	4918	5996	5457	1.58	1.75	1.66
T_5	67915	71815	69865	89.10	90.71	89.90	178195	199557	188876	110280	127742	119011	16181	17085	16633	1.62	1.78	1.70
T_6	64315	68215	66265	93.78	98.91	96.35	187553	217610	202582	123238	149395	136317	29139	38739	33939	1.92	2.19	2.05
T_7	64565	68565	66565	104.66	110.08	107.37	209319	242177	225748	144754	173612	159183	50655	62955	56805	2.24	2.53	2.39
T_8	63965	67965	65965	101.81	104.26	103.03	203613	229364	216488	139648	161399	150523	45550	50742	48146	2.18	2.37	2.28
T_9	73765	77965	75865	119.59	128.33	123.96	239175	282331	260753	165410	204366	184888	71312	93709	82511	2.24	2.62	2.43

*Selling price of okra, ₹20/kg in 2022 and ₹22/kg in 2023.

and benefit-cost (B:C) ratio were recorded under treatment T_9, which included the application of jeevamrut (500 L/ha) and amritpaani (500 L/ha) at sowing, along with foliar sprays of panchagavya (5%) and vermiwash (10%) at 30, 45, and 60 days after sowing (DAS). In contrast, the lowest values for all economic parameters were observed in the control (T_0). The superior economic performance of T_9 may be attributed to higher okra yields, especially in 2023, which also saw better market prices. Moreover, application of liquid organic manures appeared to reduce disease incidence, thereby protecting crop yield despite higher input costs. These higher returns ultimately compensated for increased expenditure. The findings align with previous research by Akhtar *et al.* (2018), Patel *et al.* (2018), Parewa *et al.* (2021), Radwan *et al.* (2021), and Kumawat *et al.* (2022), which reported improved economic outcomes due to increased productivity and the cost-effective nature of organic inputs.

The Influence of liquid organic manures on plant height, number of leaves and number of branches was recorded as significant during both the years of investigation, over the control. The increase in plant height was due to improved soil chemical and physical characteristics under manure application. Plants responded to the improved conditions under manure. The significant increase in total yields in manured plots might also be attributed to the increasing branching.

The maximum pooled value of fruit length, fruit weight and number of pickings while, shortened days taken to pickings were observed under T_9 treatment (*i.e.* jeevamrut @500 litres/ha at sowing + amritpaani @500 litres/ ha at sowing + foliar spray of panchagavya 5% + foliar spray of vermiwash 10% at 30, 45 and 50 das). in, okra more branching accounts for increasing number of pickings as pod developed in the axil of every branch once flowering has begun. similarly, the significant changes in fruit length, fruit weight is due to difference in soil structure and fertility. another possible reason for this might be due to that cow dung in jeevamrut acts as a media for the growth of beneficial microorganisms and cow urine provides nitrogen which is essential for crop growth upon fermentation with other ingredients in jeevamrut. These results are in consonance with findings of Siddappa (2015) in field bean, Basavaraj Kumbar (2016).

The analysis of data revealed that application of different liquid organic manures significantly influenced plant growth parameters of okra during both the years of study and in the pooled analysis (Table 1). Growth attributes such as plant height, number of leaves per plant, and number of branches per plant were significantly affected by the treatments at different growth stages (20, 40, and 60 days after sowing).

Among various treatments, combined application of T_9 (jeevamrut @ 500 L/ha at sowing + amritpaani @ 500 L/ha at sowing + foliar spray of panchagavya 5% + foliar spray of vermiwash 10% at 30, 45, and 60 DAS) recorded the highest values for plant height and number of leaves per plant at all observed growth stages. However, at the final harvest stage, the differences among the treatments were statistically non-significant, although all treated plots showed comparatively higher values than the control. This suggests that the application of liquid organic manures had a more pronounced effect during the early and mid-growth stages of the crop.

Similarly, number of branches per plant was significantly influenced by the different treatments at 20, 40, and 60 DAS as well as at the final harvest stage. The maximum number of branches per plant was recorded under treatment T_9, whereas the lowest values were observed in the control treatment (T_0). Increased branching in okra is particularly important because it contributes to the development of more flowering sites, which ultimately enhances yield potential.

The improvement in growth parameters under liquid organic manure treatments may be attributed to enhanced nutrient availability, improved soil microbial activity, and better soil physical conditions. Organic formulations such as jeevamrut and panchagavya are known to contain beneficial microorganisms, enzymes, and plant growth-promoting substances, which stimulate plant metabolism and vegetative growth. Additionally, the foliar application of liquid formulations such as panchagavya and vermiwash may have facilitated rapid nutrient absorption through leaves, thereby improving physiological efficiency and vegetative growth of the crop.

The data indicate that different liquid organic manure treatments significantly influenced yield attributes of okra, including fruit length, fruit weight, days to first picking, and number of pickings during both years (2022 and 2023) as well as in the pooled analysis (Fig.1).The treatment T9 (jeevamrut @ 500 L/ha at sowing + amritpaani @ 500 L/ha at sowing + foliar spray of panchagavya 5% + foliar spray of vermiwash 10% at 30, 45, and 60 DAS) consistently recorded the highest fruit length, fruit weight, and number of pickings, along with comparatively fewer days required for first picking. In contrast, lowest values for these yield parameters were observed under the control treatment (T_0).

The higher fruit length and fruit weight under treatment T_9 may be attributed to improved nutrient availability and enhanced plant vigour resulting from the combined application of soil-applied and foliar organic formulations. Jeevamrut, which contains fermented cow dung, cow urine, pulse flour, and jaggery, serves as a rich source of beneficial microorganisms and nutrients that enhance soil biological activity and nutrient mineralization. Cow dung present in jeevamrut acts as a medium for microbial proliferation, while cow urine provides nitrogen and other essential nutrients that support plant growth. These processes may have contributed to improved nutrient uptake and better development of reproductive structures.

Furthermore, increase in number of pickings under treatment T_9 could be associated with the higher number of branches observed in the same treatment. In okra, fruits develop in the axils of leaves and branches once flowering begins; therefore, increased branching directly contributes to higher fruit production and extended harvesting duration. Improved soil fertility and soil structure under organic manure treatments may also have supported better root development and nutrient uptake, thereby enhancing fruit development and overall productivity.

The results are consistent with the findings reported by Siddappa (2015) in field bean and Basavaraj Kumbar (2016), who also observed improved growth and yield attributes with the application of organic liquid formulations.

The application of liquid organic manures significantly influenced the economic returns from okra cultivation during both the experimental years and in the pooled analysis. Among the treatments, T_9 recorded the highest gross return, net return, and benefit–cost (B:C) ratio, while the control treatment (T_0) recorded the lowest economic returns. The superior economic performance of treatment T9 can be attributed primarily to the higher fruit yield obtained under this treatment. The improved growth and yield attributes resulting from the combined application of jeevamrut, amritpaani, panchagavya, and vermiwash ultimately translated into greater marketable yield and higher returns to the growers.

Although the application of multiple organic inputs slightly increased the cost of cultivation, the additional yield obtained under this treatment compensated for higher input costs and resulted in improved profitability. Another possible factor contributing to the higher economic returns under organic treatments could be the improved plant health and reduced disease incidence often observed in crops receiving organic amendments. Enhanced microbial activity and improved soil health under organic management practices may have strengthened plant resilience against environmental stresses, thereby protecting yield potential.These results are consistent with earlier findings reported by Akhtar *et al.* (2018), Patel *et al.* (2018), Parewa *et al.* (2021), Radwan *et al.* (2021), and Kumawat *et al.* (2022), who also observed improved economic returns in vegetable crops with the use of organic inputs due to enhanced productivity and relatively lower input costs compared to conventional practices.

CONCLUSION

The application of liquid organic manures significantly improved the growth, yield attributes, and economic returns of okra. Among the treatments, the combined application of jeevamrut (500 L/ha) and amritpani (500 L/ha) at sowing along with foliar sprays of panchagavya (5%) and vermiwash (10%) at 30, 45, and 60 DAS (T_9) produced the best results in terms of plant growth, fruit and yield parameters.

Conflict of Interest Statement

The authors declare no conflict of interest

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Validation of ToLCNDV resistant germplasm in melon (*Cucumis melo*) and cucumber (*Cucumis sativus*) through challenged inoculation

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Abstract

The study was carried out to validate the Tomato Leaf Curl New Delhi Virus (ToLCNDV) resistance in four promising accessions of melon (*Cucumis melo* L.) and cucumber (*Cucumis sativus* L.) accessions, *viz*. IC629818-A, IC629818-B, IC629821, and IC410617 along with checks PI 124111, Pusa Uday and Punjab Naveen through artificial inoculation at glasshouse facility of ICAR-IARI, New Delhi, during 2024-25. These lines were identified through field screening against ToLCNDV during the *kharif* seasons of 2021–2024 at the ICAR-NBPGR, New Delhi. Under artificial inoculation in the glasshouse, accessions IC629818-A and IC629818-B exhibited no visible symptoms of ToLCNDV and recorded a vulnerability index of 0. PCR amplification had further confirmed the absence of viral infection in these lines. This is perhaps the first report wherein any melon germplasm showed immune/highly resistant reaction to ToLCNDV. The cucumber genotype IC410617 had little symptom (VI=4.5) with no amplification of viral coat protein and thus categorized as resistant. These lines show potential for their utilization as novel sources of resistance for breeding of ToLCNDV-resistant cucumber and melon cultivars.

Key words: ToLCNDV, Resistant germplasm, Cucumber, Melon, Vulnerability index, Artificial screening, Challenged inoculation

Cucumber (*Cucumis sativus* L.) and melons (*Cucumis melo* L.) constitute an important group of Cucurbitaceous vegetables cultivated for their nutritional benefits. Cucumber cultivation during the rainy season in Northern India is becoming a challenge due to the spread of Tomato Leaf Curl New Delhi Virus (ToLCNDV), which compels the farmers to increase insecticide applications per harvest, thereby posing serious threats to human health and the environment. Moreover, in many countries, this virus has presented a serious threat to melon crops (Juárez *et al*., 2014). Huge crop yield loss has been reported worldwide due to infection of plant viruses that are predicted to cost over $30 billion a year (Rabadán *et al*., 2023). These consequences highlight how crucial it is to create and carry out efficient virus-resistance breeding programmes for melons and other crops. The primary method of controlling this virus is by using pesticides to regulate its vectors, which poses a serious risk to both human health and the environment. The best strategy to combat with diseases and pests is genetic resistance, which necessitates identification of resistance sources from the gene pool to use in development of resistant varieties. Keeping this in view, the present investigation was planned to validate and identify ToLCNDV resistant genotypes in cucumber and melon through artificial screening.

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Based on the disease response of 48 cucumber and melon genotypes to ToLCNDV under field conditions during 2023–24 (data not presented), seven genotypes, including the most resistant ones along with three checks, were selected for validation of resistance through challenge inoculation using viruliferous whiteflies. A virus-free stock of whiteflies (*Bamacia tabaci*) was reared on healthy cucumber plants in controlled environment (28 ± 2°C, 60 ± 10% RH, 16 hr light-8 hr dark photoperiod) in whitefly rearing chamber at the Advance Centre for Plant Virology, ICAR-Indian Agricultural Research Institute, New Delhi during March 2024. A culture of ToLCNDV was maintained on tomato plant (accessions no: MW399221) grown under controlled conditions in an insect-proof greenhouse.

Thirty seedlings of each genotype were sown in plastic pots, and 15 days-old seedlings were used for inoculation. Whiteflies were allowed an acquisition access to ToLCNDV-infected tomato plants maintained under greenhouse condition for 24 hr. Ten adult virulent whiteflies were then allowed for access to each accession at the second true-leaf stage for 48 hrs within mylar cages. Symptoms on the upper leaves of each genotypes were recorded by visual evaluation using the following scale (Islam *et al*., 2010), where 0= no symptoms; 1= mild symptoms on young leaves covering >10% of leaf area; 2= mosaic on young leaves covering >25% of leaf area symptoms; 3= mosaic on young leaves covering >50% of leaf area symptoms and 4=mosaic on young leaves covering >75% of leaf area symptoms. For better comparison of resistance among the genotypes, vulnerability index (V) values were calculated using the formula as given below.

Vulnerability Index (VI)= $(0n_o+1n_1+2n_2+3n_3+4n_4+5n_5)/nt(nc-1) \times 100$

where, $n_0, n_1, n_2...n_5$ = number of plants in score 0, 1, 2...5 respectively; nt = total number of plants; nc = total number of categories. On the basis of the vulnerability indices, the genotypes were categorized into five categories, VI=0 immune; VI= 1-25, resistant; VI= 26-50, moderately resistant; VI= 51-75, moderately susceptible; VI= 76-100, susceptible (Islam *et al*., 2010).

Total DNA was isolated at 4th week after inoculation (Doyle and Doyle, 1990; Murray and Thompson, 1980) from all the tested genotypes, and PCR amplification was conducted using ToLCNDV-specific primer pair AG155F-AG158R (Roy *et al*., 2021) for ToLCNDV. The symptoms in glasshouse-grown plants started 1st week after inoculation with minute yellow spots; later, they progressed to yellow mosaic and curling and covered the entire leaf very quickly within 3 weeks in susceptible genotypes.

Arya melon, IC629818-A and IC629818-B, showed no symptom of yellowing and curling of leaves (VI=0) while the check, Pusa Uday and Arya IC629821, had mild symptoms and thus assessed as moderately resistant (VI=25.75-49.81) (Table 1). Cucumber accession IC410617, exhibiting a vulnerability index (VI) of only 4.5%, was identified as resistant to ToLCNDV, whereas the check variety Punjab Naveen was classified as moderately susceptible (VI = 64.75). These findings are in agreement with the field study of Pandey *et al.* (2022), which reported that most Arya melon accessions were resistant to ToLCNDV.

Table 1. Response of cucumber and melon genotypes screened for resistance against ToLCNDV through whitefly mediated challenged inoculation

Genotype	Species	Vulnerability Index %	Disease reaction
IC410617	*Cucumis sativus* L.	4.50	R
Arya IC629821	*Cucumis melo* ssp *melo* var. *alwarensis*	31.14	MR
PI 124111	*Cucumis melo* ssp *melo*	25.75	MR
Punjab Naveen	*Cucumis sativus* L.	64.75	MS
Pusa Uday	*Cucumis sativus* L.	49.81	MR
Arya IC629818-B	*Cucumis melo* ssp *melo* var. *alwarensis*	0.00	I
Arya IC629818-A	*Cucumis melo* ssp *melo* var. *alwarensis*	0.00	I
	CD@ P < 0.05	5.33	
	SE(m)	1.81	
	C.V. %	5.41	

The artificially inoculated plants, when subjected to PCR amplification with coat protein primers, did not show any amplification in Arya IC629818-A, IC629818-B, and cucumber IC410617, confirming the absence of viral genomes (Fig. 1). Though Arya IC629821 showed some symptoms in glasshouse-grown plants which placed it in moderately resistant category but in PCR study, no band was observed, revealing that very low virus load is present (Fig. 1). Check variety Punjab Naveen showed a clear-cut band of higher intensity showing presence of viral coat protein in good concentration.

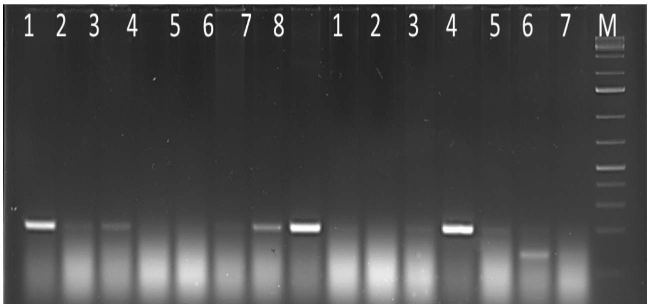

Fig. 1 PCR profiles of susceptible and resistant accessions grown under artificial epiphytotic conditions, sampled and checked for the presence of ToLCNDV using ToLCNDV coat protein-specific primer pairs. Primers used – AG155/ AG158, Expected amplicon- ~200 bp, Lane 1: IC629818-A; Lane 2: IC629818-B; Lane 3: PI 124111; Lane 4:Punjab Naveen; Lane 5:Pusa Uday; Lane 6: IC629821; Lane 7: IC410617; Lane M: 1000 bp Marker

The PI 124111 showed moderately resistant reaction in greenhouse with faint/low intensity of band (Fig 1.) Lower band intensity in a PCR reaction indicates a lower concentration of DNA. This might be possible that viral coat protein was able to multiply but the concentration was not enough to produce any prominent band in the PCR profile or express more symptoms in plants. That is why plants were categorised as moderately resistant on the basis visual symptoms. Previously PI 124111 has been reported to show partial resistance to ToLCNDV (Romay *et al.*, 2019). Five Indian melon genotypes, belonging to subsp. *agrestis* (Naudin) Pangalo, were reported to exhibit resistance to the Spanish isolate of ToLCNDV (López et al., 2015). The wild gene pool has also been utilized for introgression of ToLCNDV resistance in tomato (Singh and Rai, 2023).

Candidate genes for high level of resistance against ToLCNDV were identified from wild melon germplasm (Saez *et al.*, 2017) and recently new source of resistance from (*C. melo* var. *callosus)* and *C. melo* var. *momordica* have been reported from Indian genepool (Padmanabha *et al.*, 2023). This shows the potential of diversity present in cucumber and melons coming from Indian gene centre. However, this is perhaps the first report where a germplasm is showing immune reaction against ToLCNDV disease in melons. Arya melon collected from Alwar district of Rajasthan is a unique type of melon used for salad purpose (Pandey *et al*., 2020). This can serve as a potential source of resistance to other melon types which are crossable with each other. In cucumber, only a few sources of resistance are reported. In our previous study, IC410617 showed field resistance to ToLCNDV (Ranjan *et al*., 2015; Pandey *et al*., 2022). Identification of IC410617 as a novel source of resistance to ToLCNDV in cucumber is very important for successful cucumber cultivation during *kharif* season in northern India.

CONCLUSION

Thus, we report new sources of resistance to ToLCNDV in cucumber and melon. The field resistance of cucumber accession IC410617 and melon accessions Arya IC629818-A and IC629818-B against ToLCNDV has been validated through artificial screening under controlled conditions.The artificially inoculated plants of IC629818-A and IC629818-B genotypes had no symptoms as well as no viral DNA in their PCR profiles. Cucumber IC410617 had very little symptom but no viral DNA in the PCR profile. This is perhaps the first report documenting a melon landrace exhibiting immunity to ToLCNDV. Since melons are inter-crossable within the species, the genes conferring ToLCNDV resistance can be readily introgressed into commercial melon cultivars, facilitating the development of virus-resistant varieties.

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ANNOUNCEMENT

Call for Applications: Editorial Board Members and Early Career Editorial Members (2026–2028)

Greetings from SHRD

The editorial team of *Current Horticulture* invites applications and nominations for appointment to the **Editorial Board** and **Early Career Editorial Member (ECEM)** positions for the term **January 2026 to December 2028 (three years)**.

Current Horticulture is an international, peer-reviewed journal published by the Society for Horticultural Research and Development (SHRD). The journal is dedicated to disseminating high-quality research and review articles in horticultural science, encompassing fruit, vegetable, floriculture, plantation crops, postharvest technology, biotechnology, and allied disciplines. It aims to promote impactful scientific advancements and sustainable production systems in horticulture.

Applications for **Editorial Board Membership** are invited from accomplished researchers, academicians, and professionals with proven expertise and a strong publication record in relevant areas. Members of the Editorial Board are expected to contribute to the peer review process, provide guidance on journal policies and scope, support the maintenance of high academic standards, and actively promote the journal within the scientific community.

The journal also seeks to engage motivated young researchers as **Early Career Editorial Members**. This initiative is intended to build editorial capacity and involve emerging scientists in scholarly publishing. Early Career Editorial Members will assist in manuscript screening, support the peer review process, and contribute to outreach and promotion activities under the guidance of senior editors.

Interested candidates are requested to submit a brief curriculum vitae along with a statement of interest outlining their area of expertise and willingness to contribute to the journal. Applications and nominations may be sent **on or before 30 April 2026**. The editorial team looks forward to the participation of dedicated and dynamic researchers to further strengthen the scientific quality, visibility, and global impact of *Current Horticulture*.

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