Review Article

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Aonla rust, its causes, epidemiology and management- A review

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ABSTRACT

Aonla or Indian Gooseberry is an important fruit crop grown in the Indian sub-continent, celebrated for its exceptional nutritional and rejuvenating properties and highly valued in Unani and traditional Indian medicine for its medicinal benefits. However, Aonla rust caused by the fungus *Ravenelia (Phakopsora) emblicae*, poses significant threat to productivity of this fruit crop as it can severely impact crop yield and quality, leading to economic losses for farmers. This fungal disease thrives in warm, humid conditions and is exacerbated by factors such as improper planting density and poor water management. The effects of Aonla rust include reduced fruit production, lower fruit quality, and overall tree health decline, ultimately affecting the economic viability of Aonla farming. Effective management of Aonla rust involves a combination of cultural practices, chemical treatments, use of bioagents and the use of resistant varieties. The common chemical treatment interventions involve foliar application of wettable sulphur / Mancozeb 75 WP/ Chlorothalinil @ 0.2% and Copper oxychloride 50 WP @ 0.3% at 10-12 days' intervals starting in August. Bio-agents such as *Trichoderma harzianum* and *Pseudomonas fluorescens* and organic pesticides like neem oil, NSKE, and neem leaf extract have also been proven effective in disease control. Strategies such as proper spacing, pruning, and the application of fungicides are essential in disease management. Additionally, the development and use of rust-resistant Aonla varieties is crucial for sustainable production. Ongoing research and integrated disease management approaches will be key to mitigating the impact of Aonla rust and ensuring the continued success of Aonla cultivation.

Introduction

Emblica officinalis Gaertn. or *Phyllanthus emblica* L. is commonly known as Indian Gooseberry or Aonla which thrives in the tropical and subtropical regions of India, China, Indonesia, Myanmar, Sri Lanka, and the Malay Peninsula (Benthal, 1946; Liu *et al.,* 2008; Macmillan, 1943; Perianayagam *et al.,* 2005; Thakur *et al.,* 1988). Additionally, it has been reported to grow naturally in other parts of the world, including Cuba, Puerto Rico, Iran, Iraq (Hooper and Field, 1937), Hawaii, Florida (Barrett, 1956; Sturrock, 1959), Java, the West Indies, and Trinidad (Webster, 1956). India

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holds the top position globally in terms of both the area dedicated to Aonla cultivation and its production. It is grown at an estimated area of 100,000 ha with an annual production of 12.06 lakh tonnes (National Horticulture Board, 2022). In India, the plant is found abundantly in northeastern states such as Mizoram, Tripura, and Assam, particularly in the Khasi and Garo hills of Meghalaya (Pandey *et al.,* 1993). Intensive cultivation, however, is primarily concentrated in Uttar Pradesh, especially in the districts of Pratapgarh, Azamgarh, Varanasi, and Jaunpur (Bajpai and Shukla, 2002). Other states, including Maharashtra, Gujarat, Rajasthan, Madhya Pradesh, Jharkhand, Chhattisgarh, Bihar, West Bengal, Orissa, Andhra Pradesh, Karnataka, Haryana, and Himachal Pradesh, also cultivate Indian gooseberry (Chandra and Singh, 2015; Rawat and Uniyal, 2003).

Aonla is a significant minor crop in India and belongs to the family Euphorbiaceae (Kumar and Sagar, 2009). Although, it is classified as an underutilized crop (Scartezzini *et al.,* 2006), such crops have the potential to enhance national food security (Mayes *et al*., 2012). This plant is adaptable to various soil types, ranging from sandy loam to clay, and thrives in arid and semi-arid regions. Its cultivation is beneficial for farmers, as it grows well even on marginal lands. It is known for its numerous health benefits and is frequently used in Ayurveda and Unani medicine (Pathak, 2003). It is particularly noted for its high vitamin C content, which is about 20 times greater than that of orange (Tarwadi and Agte, 2007). Aonla juice, being highly acidic, helps protect the vitamin C from degradation during heating or drying (Hassan *et al.,* 2014). Ascorbic acid in Indian gooseberry acts as an antioxidant due to its ability to scavenge free radicals (Cort, 1982). However, besides ascorbic acid, Indian gooseberry is rich in polyphenols such as ellagic acid, gallic acid, and hydrolysable tannins (including Emblicanin A, Emblicanin B, punigluconin, and pedunculagin), which also contribute significantly to its antioxidant properties (Bhattacharya *et al.,* 1999; Ghosal *et al.,* 1996). Research by Ihantola-Vormisto *et al.* (1997) has shown that Indian gooseberry possesses anti-inflammatory and antipyretic properties. Tasduq *et al.* (2005) and Reddy *et al.* (2010) have demonstrated hepatoprotective activity of Aonla fruit by using its 50% hydroalcoholic extract and 5% aqueous extracts respectively. The fruit's anti-tumor properties are largely attributed to its polyphenolic compounds, particularly tannins and flavonoids. For instance, pyrogallol extracted from Indian gooseberry has been shown to have an anti-proliferative effect on human lung cancer cell lines (Yang *et al.,* 2009), while gallic acid from its leaves induces apoptosis in human hepatocellular carcinoma cells (Huang and Zhong, 2011). Additionally, extracts from *E. officinalis* fruits inhibit the transcription factor AP1 and disrupt the expression of viral oncogenes, which may prevent the development and progression of cervical cancer, making the fruit a potential source for drug development against Human Papilloma Virus (HPV)-induced cervical cancer (Mahata *et al.,* 2013). Beyond these properties, Indian gooseberry also exhibits hypolipidemic (Thakur and Mandal, 1984; Yokozawa *et al.,* 2007), hypoglycemic (Jamwal *et al.,* 1959; Liu *et al.,* 2012), and analgesic effects (Perinayagam *et al.,* 2004). In India, popular commercial cultivars include 'Banarasi,' 'Francis,' 'Chakaiya,' 'Kanchan (NA-4),' 'NA-6,' and 'NA-7' (Pathak, 2003; Scartezzini *et al.,* 2006). The cultivated area in India under Aonla variety "NA-7" spans about 85% of total cultivated area. Pakistan cultivates the 'Desi,' 'Shisha,' and 'Banarasi' varieties, while in China, cultivars such as 'Langen,' 'Fen'gan,' 'Liuyuebai,' 'Bian'gan,' 'Quibai,' and 'Shan'gan' are prevalent. Indian gooseberry is commonly processed into products like pickles, candy, Murabba (whole fruit preserve), juices, mouth fresheners, and fruit leathers, which have substantial economic value (Bhattacherjee *et al.,* 2011; Daniel and Dudhade, 2010; Nath and Sharma, 1998). It serves as the main ingredient in Chyavanprash, a popular Ayurvedic preparation, with the Chyavanprash industry in India valued at Rs 200 crore, involving both large companies and small-scale producers. Due to the fruit's sour and astringent taste, it is typically consumed in processed forms rather than raw. However, the cultivation of Indian gooseberry faces challenges, notably from Aonla rust, a significant disease caused by the fungus *Ravenelia emblicae*, which affects the leaves, stems, and fruits of the plant. This disease can lead to severe yield losses, with reports indicating up to a 50% reduction in fruit yield under severe infection. It also impacts fruit quality, reducing its market value and overall profitability for farmers.

Aonla can be grown in light as well as heavy soils except purely sandy soil. Calcareous soil with rocky substratum also promotes Aonla growth. The Aonla plant thrives in well-drained, fertile loamy soil, which is ideal for achieving higher yields. It is adaptable to dry regions and can also grow in moderately alkaline soils. It can be grown in acidic to saline/ sodic (pH up to 9.5, ESP-35 and ECe-6-9 ds/m) soils (Pathak and Pathak, 2001). Although, it is considered a subtropical fruit, it can be cultivated successfully in tropical climates as well. Annual rainfall of 630-800 mm has given good yields. In India, Aonla is cultivated in a wide range of altitudes, including areas near the sea coast up to an altitude of 1800 meters (Pathak, 2003). Mature Aonla tree can tolerate freezing as well as high temperature of 48° C, but the plants are susceptible to frost in winter and sometimes heavy damage occurs owing to frost in hot arid ecosystem of western part of Rajasthan (Pathak *et al.,* 2006). After fruit set in spring, the fruits remain dormant throughout summer without any growth. This quality makes it highly suitable fruit crop for dry arid region.

This review synthesizes the current knowledge on Aonla rust and highlights the importance of continued research and extension efforts to support Aonla growers in managing this devastating disease.

Major diseases of Aonla

Aonla is considered a hardy crop but there are few important diseases, which create quantitative as well as qualitative losses in fruit yield of Aonla. Among them, rust (*Ravenalia emblica*e Styd.), anthracnose (*Colletotrichum* state of *Glomerella cingulata*) (Mishra and Shivpuri, 1983), dieback (*Botrydiplodia theobromae* Pat.) (Arya *et al.*, 1987), blue mould (*Penicillium citrinum*). Losses of Aonla during storage are considerable mainly due to sprouting and contamination by microorganisms. Rajam (1992) reported that among the post-harvest disease of Aonla in India, sooty mould (*Capnodium sp*.), fruit rot (*Penicillium indicum, P. oxalicum, Aspergillus niger*), soft rot (*Phomopsis phyllanthi* Punith), black soft rot (*Syncephalastrum racemosum*) are important diseases which cause heavy loss to the growers (Singh *et al.,* 2010). Rust disease, anthracnose and post-harvest diseases are major constraints in its crop production in some parts of Uttar Pradesh and Rajasthan states. In other countries like China, the brown spot, false anthracnose and powdery mildew have been reported widely.

Etiology, occurrence and distribution of Aonla rust

Aonla rust is a significant and economically detrimental disease affecting Indian gooseberry (*Phyllanthus emblica*). The primary pathogen responsible for this disease is *Ravenelia emblicae*, a biotrophic, obligately parasitic, fungus that depends on living host tissue for its growth and reproduction. This pathogen poses a serious threat to Aonla cultivation not only in Uttar Pradesh but also in other major Aonla-producing states such as Rajasthan, Andhra Pradesh, Tamil Nadu, and Haryana. The disease was first identified and reported in Rajasthan, India in 1967 (Tyagi, 1967) and has since been observed in various regions across India, causing substantial losses to growers (Tyagi, 1967; Rawal, 1993). Aonla rust requires living host for causing infection and establishment under favourable conditions. It causes fruit and leaf infection leading to significant fruit losses.

Disease symptoms and development

The initial symptoms of Aonla rust include the appearance of small, circular, orange-brown pustules on the upper surface of the leaves. As the disease progresses, these pustules enlarge, reaching sizes of 3-4mm, and may turn the leaves yellow or red. In some cases, the pustules also develop on green leaves. On fruits, the disease manifests as brown to black pustules that often arrange themselves in a ring. Over time, these pustules coalesce, covering larger areas of the fruit surface, which can significantly reduce the marketability of the produce (Tyagi and Pathak, 1988; Jat and Goyal, 2004; Jarial *et al.,* 2011; Prakash and Misra, 1993).

Interestingly, it has been noted that severe infections on fruits do not always coincide with symptoms on leaves, and vice versa, indicating a complex disease dynamic (Tyagi, 1967). This variability in symptom expression can complicate early detection and management efforts.

Pathogen biology and disease cycle

The genus *Ravenelia* is one of the largest (third) genera of the order Pucciniales. The genus is global in distribution and species are reported to infect different plants. With the discovery of type species, more than 250 described species are widely distributed in subtropical and tropical regions (Cummins 2003; Ebinghaus, 2020). It parasitizes the trees and bushes of families Fabaceae and Euphorbiaceae with high host specificity towards Mimosoideae, Faboideae and Caesalpinioideae (Fabaceae). The genus was introduced in the year 1853 by Berkeley; over time, the genus has undergone several taxonomic and systematic transformations. However, all species of *Ravenelia* shared the most prominent morphological features including the production of multicellular teliospores on compound pedicels composed of two to several hyphae with autoecious and macro- and demi- to hemi-, and, more rarely, to microcyclic modes of their life cycle. With the addition of many species in recent years, this genus has become one of the largest genera of rust fungi (Avasthi *et al.* 2024).

Ravenelia emblicae primarily infects the leaves of the Aonla plant, where it forms characteristic rust pustules. These pustules contain uredospores, which are dispersed by wind, facilitating the spread of the disease (Fig. 1). The infection process begins when the uredospores land on the leaf surface and germinate, leading to the formation of specialized structures called appressoria. These appressoria penetrate the epidermal cells of the leaf, establishing the infection and leading to the development of new rust pustules (Fig. 1). As the cycle continues, new uredospores are released from the pustules, causing secondary infections throughout the growing season. The disease thrives in warm and humid conditions, particularly during the onset of the rainy season. Such environmental conditions are ideal for the pathogen's sporulation and dissemination, leading to widespread outbreaks (Raj and Arya, 2005).

Two distinct types of rust have been identified on Aonla plants (Jansen and Cardon, 2005). The first type is leaf rust, caused by *Phakopsora phyllanthi*, and the second is ring rust, caused by *Ravenelia emblicae*. The rust caused by *Ravenelia emblicae* was first documented on Aonla in Saharanpur, Uttar Pradesh, by Nirwan *et al*., 1969-1971. This disease has since been recognized as a significant issue in traditional Aonla varieties in Rajasthan, particularly in the Udaipur district, and has also been reported in other regions,

including Lucknow and Pratapgarh in Uttar Pradesh (Pathak *et al.,* 2003; Singh and Mishra, 2007). *Ravenelia emblicae* has been associated with substantial losses in key Aonla-growing areas of Uttar Pradesh (Rawal, 1993). While both *Phakopsora phyllanthi* and *Ravenelia emblicae* have been reported in various major Aonla cultivation regions across India (Pathak *et al.,* 2003; Singh and Mishra, 2007; Rawal, 1993), there has yet to be a report of *Phakopsora phyllanthi* affecting Aonla in Himachal Pradesh.

Ravenelia emblicae is a biotrophic fungus, meaning it depends on living host tissue for its growth and reproduction (Smith *et al.,* 2019). The fungus primarily targets the leaves of its host, where it forms characteristic rust pustules (Jones and Brown, 2021). These pustules house uredospores, which are dispersed by wind, thereby spreading the disease (Doe and Lee, 2020). The disease cycle begins when uredospores germinate on the leaf surface, leading to the formation of appressoria, which penetrate the leaf's epidermal cells (Miller *et al.,* 2018). This process establishes the infection, resulting in the development of rust pustules (Taylor and Green, 2022). The cycle continues as new uredospores are produced and released, leading to secondary infections throughout the growing season (Nguyen and Patel, 2023).

Epidemiology and environmental factors

Aonla rust, caused by *Ravenaliia emblicae*, is significantly influenced by various environmental factors, which play a critical role in the disease's development and spread. Understanding these factors is crucial for effective disease

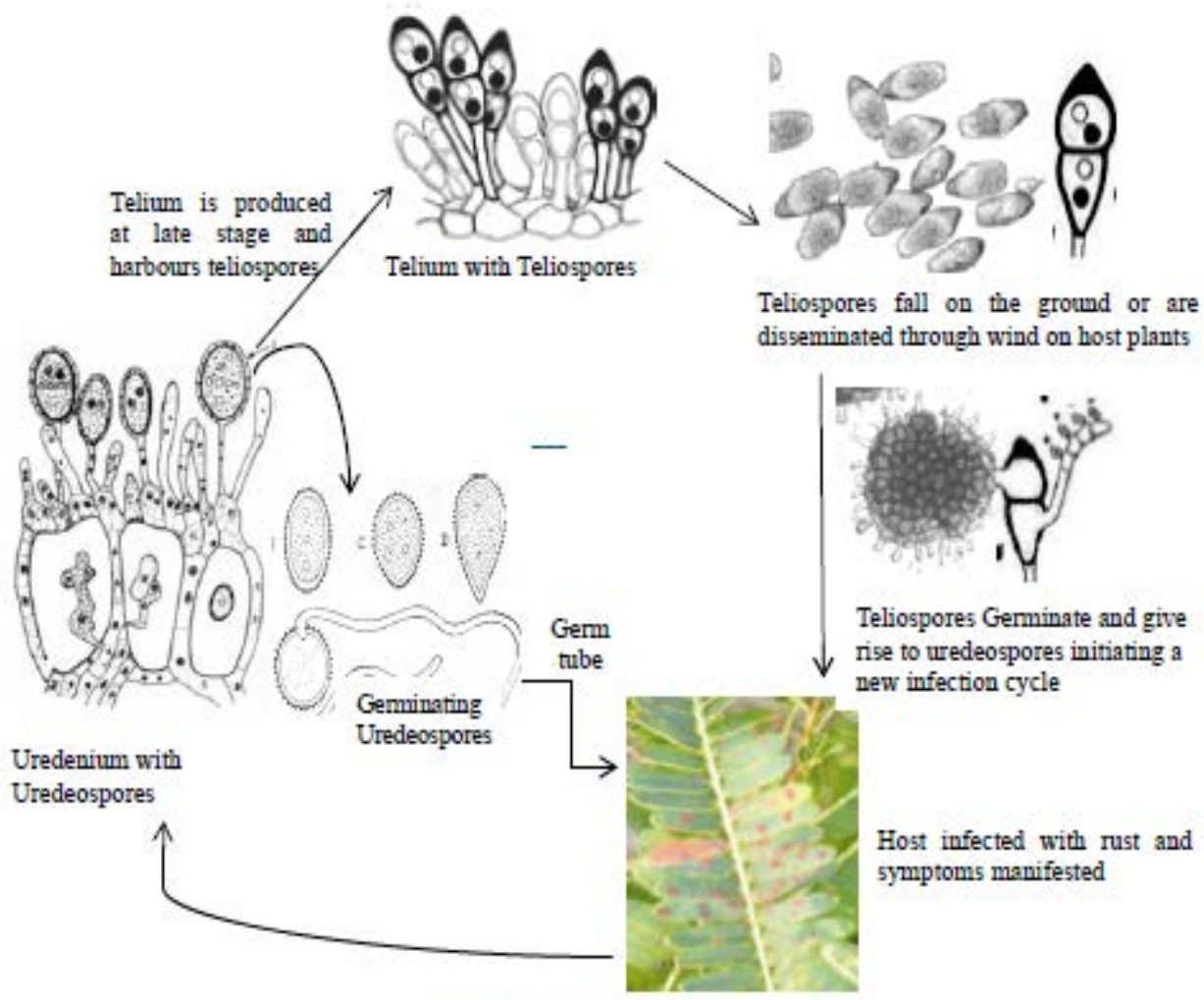

Fig. 1. Disease cycle of Aonla Rust

management. The onset of the rainy season typically marks the beginning of Aonla rust outbreaks. This period is characterized by warm and humid conditions, which are ideal for the pathogen's development. Specifically, high humidity levels combined with moderate temperatures ranging between 20-25°C provide an optimal environment for the pathogen to thrive (Raj and Arya, 2005; Saharan and Meena, 2001). During this period, the fungus sporulates,

producing uredospores that are capable of infecting the host plant. The high humidity helps maintain the moisture on leaf surfaces, which is necessary for the germination of these spores and subsequent infection.

Rainfall is another critical factor in the epidemiology of Aonla rust. It aids in the dispersal of uredospores, which are carried by raindrops and wind to new infection sites. Additionally, rainfall creates a favorable microclimate by increasing humidity and reducing evaporation, which are essential conditions for the pathogen's life cycle. However, while rainfall promotes the spread of the disease, it also presents a challenge for disease management, as it can wash away protective fungicide sprays applied to the crop, necessitating more frequent applications.

Correlation studies have provided further insights into how different environmental factors correlate with the severity of Aonla rust. For instance, temperature (both maximum and minimum) and evaporation have been found to have a highly significant negative correlation with the percent disease index (PDI) (Pandey *et al.,* 2007; Anonymous, 2002-03). This means that higher temperatures and greater evaporation rates are associated with lower disease severity. This could be due to the fact that higher temperatures and increased evaporation lead to drier conditions, which are less favorable for the pathogen's survival and infection processes. On the other hand, relative humidity, particularly in the evening, and rainfall have shown a significant positive correlation with PDI. High relative humidity in the evening can prolong the leaf wetness period, providing the necessary conditions for spore germination and infection. Similarly, increased rainfall not only aids in spore dispersal but also sustains the high humidity levels needed for the disease to proliferate. These findings highlight the importance of monitoring environmental conditions to predict disease outbreaks and implement timely control measures.

The significance of these correlations is underscored by studies conducted by Kumar *et al.,* 2023; Kumar *et al.,* 2024; Devi *et al.,* 2022; Sharma, 2020; Singh *et al,*. 2015; Mitra *et al.,* 2011. Singh *et al.* (2023) spray prediction model for Aonla rust disease using machine learning techniques can be used to predict weather the weather conditions of a particular day-minimum temperature, maximum temperature, morning relative humidity, evening relative humidity, rainfall and sunshine hours, are conductive or non-conductive for growth of rust disease in Aonla plants which further confirm the strong relationship between environmental factors and Aonla rust incidence. These studies emphasize the need for an integrated disease management approach that takes into account the prevailing weather conditions. For instance, in regions where high humidity and moderate temperatures are expected during the rainy season, preventive measures such as timely fungicide applications and the use of resistant cultivars should be prioritized.

Management strategies

Aonla rust, caused by *Ravenalia emblicae*, is a significant disease that can result in substantial yield losses for growers (Singh *et al.,* 2010). Several management practices have been studied and found effective in controlling this disease.

Chemical control

The application of fungicides is a common approach to managing Aonla rust. Sprays of wettable sulphur (0.2%) or Mancozeb 75 WP (0.2%) have been shown to effectively manage rust disease when applied at intervals of 10-12 days, starting in early August (Singh *et al.,* 2009; Singh *et al.,* 2014). Additionally, Copper oxychloride 50 WP at a 0.3% concentration, combined with deep ploughing and healthy cultivation practices, has been found beneficial in controlling the disease (Singh *et al.,* 2009, Singh *et al.,* 2014). Chlorothalonil, applied at 0.2% concentration, resulted in the minimum disease severity (5.80%) and the maximum percentage of disease control (71.80%). A combination of 1% *Trichoderma viride* and 0.1% chlorothalonil also showed significant effectiveness, with a disease severity of 8.82% and disease control of 57.12%, along with a significant increase in fruit yield compared to other treatments (Jat *et al.,* 2013; Kumar *et al.,* 2017; Maheshwari and Haldhar 2018).

Integrated management approaches

Singh *et al.* (2023) explored the efficacy of different fungicides, bioagents, and organic pesticides in managing Aonla rust. The study evaluated mancozeb (0.2%), copper oxychloride (0.3%), wettable sulphur (0.2%), *Trichoderma harzianum* (1%), *Trichoderma viride* (1%), *Pseudomonas fluorescens* (1%), neem oil (0.5%), neem seed kernel extract (NSKE) (5%), and neem leaf extract (5%) on Aonla cultivars NA-7 and Chakaiya. Amongthem, mancozeb (0.2%) was the most effective, showing the highest reduction in disease intensity over control, followed by copper oxychloride (0.3%) and wettable sulphur (0.2%).

Bio-agents and organic pesticides

Bio-agents such as *Trichoderma harzianum* and *Pseudomonas fluorescens* also showed promising results in reducing disease intensity, although they were slightly less effective than chemical fungicides. Organic pesticides like neem oil, NSKE, and neem leaf extract had a moderate impact on disease control, with neem oil being the most effective among them. However, water treatment was found to be the least effective Singh *et al.,* 2009; Singh *et al.,* 2010; Singh *et al.,* 2014; Jat *et*

al., 2013; Singh *et al.,* 2023; Kumar and Singh, 2000; Devi *et al.,* 2022; Singh *et al.,* 2015).

Future aspects of Aonla rust management

Development of disease-resistant varieties

- **Ongoing research:** Significant research efforts are focused on breeding Aonla varieties with genetic resistance to Aonla rust. Advances in molecular biology and genetic engineering offer the potential to develop varieties that are inherently resistant to rust, thereby reducing the need for chemical treatments and increasing sustainability in Aonla cultivation (Mawalagedera *et al*., 2016; Gantait *et al*., 2021).
- **Biotechnological approaches:** Techniques such as marker-assisted selection (MAS) and CRISPR/Cas9 gene editing are being explored to identify and manipulate genes responsible for disease resistance. These tools can accelerate the development of resistant varieties and ensure long-term protection against Aonla rust (Thilaga *et al.*, 2017).

Climate change and disease dynamics

- **Impact of climate change:** Climate change is expected to alter the epidemiology of Aonla rust by affecting the environmental conditions that favor its spread. Warmer temperatures and changes in precipitation patterns could lead to more frequent and severe outbreaks. Understanding these dynamics will be crucial for developing adaptive management strategies (Prajapati *et al.*, 2020; Kumar *et al*., 2024)
- **Predictive modeling:** Future research may focus on predictive modeling to anticipate Aonla rust outbreaks based on climatic data. Such models could help farmers take preemptive actions, such as adjusting planting schedules or applying preventive treatments at optimal times (Singh *et al*., 2023; Kumar *et al.*, 2024; Agarwal *et al*., 2023; Prema *et al*., 2023.)

Extension and farmer education

- **Capacity building:** The future of Aonla rust management also depends on effective extension services that educate farmers on the latest research and best practices. Building capacity among farmers through training programs and access to timely information will be essential for the widespread adoption of new technologies and approaches.
- **Digital tools:** The use of digital tools, such as mobile apps and online platforms, to disseminate information about disease identification, management practices, and weather forecasts could become more prevalent. These tools can empower farmers to make informed decisions and respond quickly to disease threats (Bhattacharyya *et al*., 2021).

Conclusion

Aonla rust is significantly influenced by various environmental factors including high humidity levels combined with moderate temperatures ranging between 20-25°C and reduction in evaporation at the onset of rainy season. Fungal uredospores are carried by raindrops and wind to new infection sites. Different environmental factors correlate with the severity of Aonla rust. Temperature and evaporation have been found to have a highly significant negative correlation with the percent disease index. Higher temperatures and greater evaporation rates are associated with lower disease severity. Aonla rust remains a significant challenge to its cultivation, but advancements in research and management practices offer hope for better control. Integrated approaches that combine cultural, chemical, and biological methods, along with the development of resistant cultivars, are key to sustainably managing this disease. The common disease control treatment interventions involve foliar application of Chemicals such as wettable sulphur, Mancozeb, Chlorothalinil and Copper oxychloride; Bio-agents such as *Trichoderma harzianum* and *Pseudomonas fluorescens* and organic pesticides like neem oil, NSKE, and neem leaf extract. Strategies such as proper spacing, pruning, and the application of fungicides have been proven beneficial in disease management. Additionally, the development and use of rust-resistant Aonla varieties is crucial for sustainable production.

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Conflict of Interest

The authors have no conflict of interest.

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Review Article

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Role of metabolome in enhancing crop resilience to abiotic stress in horticultural crops-A review

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ABSTRACT

Abiotic stresses, including drought, salinity, extreme temperatures, and nutrient deficiencies, pose significant challenges to global agriculture, threatening food security and crop productivity. The metabolome, encompassing the complete set of metabolites within an organism, plays a pivotal role in plant responses to such stresses. Metabolomics, the large-scale study of metabolites, provides insights into the biochemical pathways and mechanisms underlying stress tolerance. Plants deploy complex metabolic reprogramming to mitigate stress effects, involving osmoprotectants, antioxidants, phytohormones, and secondary metabolites. For instance, compounds like proline, glycine betaine, and sugars act as osmolytes to maintain cellular homeostasis, while antioxidants such as ascorbate and glutathione mitigate oxidative damage. Stress-responsive phytohormones like abscisic acid (ABA) regulate stomatal closure and activate downstream signaling pathways. Furthermore, secondary metabolites, including flavonoids and alkaloids, contribute to stress resistance by modulating growth and defense. Advancements in metabolomics technologies, such as mass spectrometry and nuclear magnetic resonance, enable comprehensive profiling of stress-induced metabolites, offering opportunities to identify key biomarkers and pathways. These insights facilitate targeted breeding and biotechnological interventions to develop stress-resilient crops. Integrating metabolomics with genomics, transcriptomics, and proteomics can provide a holistic understanding of stress tolerance mechanisms, paving the way for sustainable agricultural practices. This study underscores the critical role of the metabolome in enhancing crop resilience to abiotic stress and highlights the potential of metabolomics in addressing global agricultural challenges through precision breeding and metabolic engineering.

Introduction

The most advanced "omics" study to describe the metabolic profile of living things is metabolomics (Dubery *et al.*, 2013). The metabolome, is a last downstream output of the genome, that is made up of many tiny molecules with

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molecular masses under 2000 Da that vary greatly in terms of their structure and chemical makeup. Detection followed by identification, and measurement of every molecule (or subset) in biological samples is known as metabolomics (Kumar *et al*., 2016). According to Kosmides *et al*. (2013) metabolomics investigations can identify metabolites from exogenous sources including medications and diets in addition to endogenous metabolites. Accordingly, metabolomics includes a wide range of small molecules or metabolic intermediates, including organic acids, ketones, aldehydes, amines, steroids, amino acids, peptides, lipids, nucleic acids, carbohydrate, vitamins, hormones, signaling molecules, and secondary metabolites like flavonoids and polyphenols (Collino *et al*., 2013).

Metabolomics is a broad word that refers to the comprehensive qualitative and quantitative investigation of the metabolites found in living things under specific environmental conditions (Freund and Hegeman, 2017). Metabolomics reproduces more detailed information on the biological regulatory mechanisms than transcriptomics and proteomics (Dos Santos *et al*., 2017). The study of metabolic profiling of higher plants for the clarification of their stress tolerance mechanisms has benefited greatly from the development of several metabolomics techniques, such as nuclear magnetic resonance (NMR) spectroscopy and mass spectrometry (MS). Over the past fifteen years, various metabolomics techniques have been applied to the study of plants and how they interact with their surroundings. In order to reduce agricultural losses brought on by imposed stresses, metabolic reactions of plants to different stresses are becoming more and more significant. Numerous reports about focused studies looking at metabolites at the level of a specific metabolic pathway have been published. These investigations laid the groundwork for more extensive study conducted over the past 20 years. In plant science, metabolomics is becoming more and more significant for improving plant quality, finding essential natural compounds, and above all evaluating how different abiotic stresses affect a plant's physiology and growth (Genga *et al*., 2011). The rapidly developing field of metabolomics aids in plant phenotyping, which may help to improve the crop plants' nutritional value (Alla *et al*., 2012).

Plant metabolomics is a powerful tool that can be applied in a variety of fields, including fingerprinting of genotypes and ecotypes, comparing mutants with their wild types, determining the activity of bioactive compounds of medicinal plants, and the effects of the environment on plant physiology. This is because plants contain a remarkably diverse range of metabolites when compared to animals (Wolfender *et al*., 2013). A variety of metabolites are produced by plants, some of which are necessary for normal metabolic functions including respiration and photosynthesis (primary metabolites) or for environmental survival (secondary metabolites). Although secondary metabolites are not necessary for plant growth, development, or reproduction, they are critical for plant survival and fecundity, particularly for defense and signaling (Wolfender *et al*., 2013). The number of metabolites in plants is so high that it is estimated that there are 2,00,000 metabolites in total, of which 7000–15,000 are found in each species and approximately 3000–5000 are found in leaves (Kim *et al*., 2010). Over the past 20 years, plant metabolomics has advanced to a promising concept and been cleverly applied in the fields of plant biotechnology and physiology (Cusido *et al*., 2014).

Plant science has effectively used metabolomics techniques like NMR, capillary electrophoresis-mass spectrometry (CE-MS), direct injection mass spectrometry (DIMS), high-performance liquid chromatography (HPLC) with photodiode array detection, thin layer chromatography with ultraviolet (UV) detection, and mass spectrometry (MS) in conjunction with gas chromatography (GC) or liquid chromatography (LC) (Wolfender *et al*., 2013). Plant metabolomics is commonly studied using GC-MS, LC-MS, DIMS, and NMR, among other techniques. The great sensitivity and resolution of MS make it superior to NMR. Prior to being passed via the mass analyzers, the analytes in MS are first ionized. Numerous ionization methods exist, including matrix-assisted laser desorption/ionization (MALDI), rapid atom bombardment, electrospray ionization (ESI), atmospheric pressure chemical ionization, and electron impact ionization (EI). Targeted and nontargeted metabolomics are two types of metabolic techniques that can be used to research the many metabolites found in plants.

According to Li *et al*. (2014), targeted metabolomics is thought to be the most promising method for measuring a subset of metabolites that have been chemically described and interpreted. Because various isomeric forms of the same metabolite exist and cannot be distinguished based alone on their m/z ratio, it is challenging to identify the hundreds of metabolites that are found in untargeted metabolomics. Stress encounters greatly increase the quantity and concentration of metabolites that are typically generated in plants. Understanding the up and down regulation of metabolites is crucial for understanding how plants react to stress. Nonetheless, researches has put forward how the plants' metabolism responded to particular abiotic stressors, such as salinity (Chandna *et al*., 2013), heat (Bokszczanin and Fragkostefanakis, 2013), drought (Seki *et al*., 2007), or heavy metal stress (Sytar *et al*., 2013).

Metabolomics profiling of drought stress

One of the most harmful stresses to plants is drought, especially in areas where plants are irrigated by rain, which causes significant alterations in metabolism. When there is a shortage of water, physiological changes tend to increase water intake and decrease water loss, which has an impact

on metabolism. As a result, among biological reactions, osmoregulators which include sugars, polyamines, alcohols, and amino acids, particularly proline accumulate to maintain cell turgor (Chaves *et al*., 2003). Therefore, a number of metabolomics investigations into drought stress and proline buildup in dehydrated leaves have been conducted in leaf tissues, frequent proline buildup has been documented in a wide range of plants under conditions such as high salinity, heavy metals, and cold temperatures that might result in poor water availability (Hochberg *et al*., 2013). Most of the metabolomics investigations were conducted on aerial components or primarily leaves. The metabolomics level of dehydration in *Arabidopsis thaliana* L. has been thoroughly investigated. ABA is formed during dehydration, and the species' aerial portion collected polyamines and amino acids in an ABA-dependent way. Raffinose was also produced without the help of this hormone (Urano *et al*., 2009). Reactive oxygen species (ROS) are induced by drought stress and comprise free radicals (superoxide radicals, alkoxy radicals, and hydroxyl radicals) as well as non-radicals (singlet oxygen and peroxide of hydrogen) (Anjum *et al*., 2017). These are extremely reactive and poisonous substances that harm proteins, carbohydrates, lipids, and DNA, disrupting cellular equilibrium. According to Davis *et al*. (2014) it has an impact on the plant's height, canopy, root development, and leaf area index. Terpenes, other cyanogenic glucosides, and other volatile components are essential for preventing adverse environmental effects in certain crop plants. Seasonal variations and drought specifically cause these chemicals to release (Griesser *et al*., 2015). In salvia plants, diterpene provides drought tolerance by inducing the ROS scavenging system (Munn´e-Bosch *et al*., 2001).

A. *thaliana* aerial portions also acquired flavonols and anthocyanins during water shortage, which may suggest that these chemicals help mitigate drought stress in addition to those biomarkers of drought stress. Drought stress has a greater effect on leaves than on other plant parts (Kang *et al*., 2019). Analysis of the effects of relative humidity in apple plants revealed that terpenes including α-pinene, camphene, β-pinene, limonene, β-caryophyllene, and (E, E)-α-farnesene are released when relative humidity is low (Vallat *et al*., 2005). Two MYB transcription factors, MdMYB88 and MdMYB124, control lignin deposition in apple xylem during drought by influencing MdMYB46, another regulator (Geng *et al*., 2018).

It was discovered that the phytoalexin content of drought-tolerant grapes rose during drought, which further promotes resistance to biotic stress (Hatmi *et al*., 2015). The impact of water stress on the metabolome depends on the genotype, while several metabolite contents in the leaves of the two wine cultivars (Shiraz and Cabernet Sauvignon) showed similar changes, the cv. Shiraz had more metabolites and less altered stomatal regulation than the other cultivar. In both cultivars, glycerate and galactonate dropped whereas nicotinate was the only organic acid to increase. Additionally, the phenylpropanoid pathway was altered in both genotypes, and specific amino acids (the drought stress-associated proline, as well as threonine, tryptophan, valine, leucine, and phenylalanine) were markedly elevated. Glutamate, however, rose in Shiraz and fell in Cabernet Sauvignon (Hochberg *et al*., 2013).

Particularly drought-susceptible (DS) and drought-tolerant (DT) cultivars' responses to stress have been characterized thanks to metabolomics methods, which have also made it possible to find possible biomarkers associated with this kind of stress (Guo *et al*., 2020). Asparagine, methionine, and γ-aminobutyric acid (GABA) all markedly elevated in DT but not in DS. Glycolic, malonic, glucoheptonic, and galactonic acids were among the organic acids that dramatically rose in DT. In DT, there was a notable accumulation of unsaturated fatty acids, such as linolenic and linoleic acids. Furthermore, there was a notable accumulation of secondary antioxidant metabolites in DT, such as fluorine and 5-methoxytryptamine. Additionally, DT showed a considerable rise in phenolic chemicals (ferulic acid, salicylic acid, and 4-hydroxycinnamic acid) and aromatic amino acids (phenylalanine). Both DT and DS cultivars showed a considerable rise in other metabolites, such as glucose-1-phosphate, D-fructose 1, 6-bisphosphate, pyruvic acid, D-glyceric acid, oxalic acid, and 2-methylfumarate. Similarly, amino acids, including proline, glycine, serine, valine, beta-alanine, threonine and isoleucine, accumulated considerably in both DT and DS.

Metabolomics methods have also been used to study the temporal dynamics of metabolite reprograming of DT and DS cultivars under drought stress. The degree of osmotic adjustment and the type of organic solutes that accumulate may also be impacted by the type of drought (acute vs. cyclic), as well as its frequency and intensity. Furthermore, it was discovered that the phytoalexin deficient4 (PAD4) gene, which is associated to phytoalexin production, plays a role in both drought tolerance and biotic interactions (Szechynska-Hebda *et al*., 2016). While the levels of TCA-cycle metabolites (malate, succinate, alpha-ketoglutarate, and citrate) were greatly reduced, those of glycolysis intermediates (pyruvate, glucose, and dihydroxyacetone phosphate) were drastically lowered as well.

The production of phenylalanine, tryptophan, tyrosine, isoleucine, and alanine may be impacted by a decrease in pyruvate. However, the TCA cycle metabolite oxaloacetate and its amino-acidic byproducts lysine and methionine also dropped. Likewise, temperature stress led to a decrease in sugar alcohols including galactitol and mannitol. Drought reduces the output of monoterpenes in spearmint and rosemary (Delfine *et al*., 2005).

Zhang *et al*. (2014) found that drought in potatoes caused transcriptional alterations linked to the manufacture of terpene and flavonoids. By changing their metabolic

pathways, arbuscular mycorrhization (AM) gives plants the ability to withstand abiotic stress and produce protective SMs. When tomato plants experience drought stress brought on by AM, terpenes and other non-volatile isoprenoids including ABA, chlorophylls, and carotenoids are essential (Asensio *et al*., 2012; Shrivastava *et al*., 2015). Drought causes biochemical alterations in terpinene in cumin. Additionally, phenyl-1,2-ethanediol is converted to cumin aldehyde by water constraint, which may have defensive effects (Rebey *et al*., 2012). Monoterpenes, glucosides, terpenoids, carotenoids, and other volatile organic compounds (VOCs) released during drought in grapes and rosemary were linked to the prevention of drought damage (Liu *et al*., 2014).

Genes such as flavonone-3-hydroxylase, flavonol synthase, and β-carotene hydroxylase-1, which are crucial for the manufacture of flavonoids, carotenoids, and other phenolic compounds, were stimulated at the transcript level in potatoes under drought stress. Drought resistance in potato cultivars is influenced by the expression level of these genes (Fan *et al*., 2008; Andr´e *et al*., 2009). Remarkably, it was discovered that tea cultivars that could withstand drought had higher levels of the metabolites glycine, asparagine, valine, isoleucine, proline, and leucine than cultivars that could not withstand drought. Additionally, it is known that a number of amino acids function as osmolytes in cultivars that can withstand drought (Shi and Chan, 2014).

Metabolomics profiling of heavy metals stress

Heavy metals are defined as those with densities more than 5 gcm^{-3}. Fifty-three of the elements found in nature are classified as heavy metals. Because they are soluble under physiological conditions, 17 of the discovered heavy metals are accessible to living cells and are vital to both the environment and living things. Because they are toxic to living organisms at higher concentrations, several metals, such as Cu, Ni, Zn, Co, Cr, V, and W, are classified as trace elements. Other metals, such as Hg, Ag, Sb, As, Cd, and Pb, seem to be fatal to plants even though they have no direct role in plant physiology or metabolism (Nazar *et al*., 2012). The form in which heavy metals are found in soil is one of the important variables that contributes to their toxicity.

The growth and physiology of various plant species are more adversely affected by metals that are present in soluble and bio exchangeable forms, which also have higher bioavailability. Because they play a major role in vital biological processes, metals like iron, manganese, and copper are necessary for plants in one way or another. In both plants and animals, Fe, Mn, Cu, Mn, and Ni serve as cofactors for the enzymes. Heavy metals are created by the pedogenic and decaying processes of rocks and are classified based on their content in soil. In addition to natural processes, industrialization, fast population growth, the generation of anthropogenic biosolids, and agrochemical wastes raise the risk of heavy metal contamination of soil (Kashem *et al*., 2009). Because heavy metals interfere with the soil's ability to absorb vital mineral nutrients, they negatively impact a plant's metabolism, development, and reproduction.

Cadmium is the most significant heavy metal in terms of its catastrophic effects since it is more soluble and mobile in soil than other heavy metals. Plants require complex physiological and biochemical coordination, protein structure alterations, and metabolite profile variations for proper signaling and stress tolerance in order to withstand the stress caused by heavy metals. Changes in the accumulation of carbohydrates in leaf tissues can be used to measure the impacts of heavy metals (Manivasagaperumal *et al*., 2011).

Additionally, Pb poisoning reduces the mobilization of stored meals, which results in suppression of germination and seedling development due to a decrease in radical production, deterioration of proteolytic activities, and disturbance of cellular osmoregulation (Cokkizgin and Cokkizgin, 2015). Furthermore Because of its detrimental effects on chlorophyll production, transpiration, root growth, and cell division, lead not only slows down seed germination but is also associated with poor seedling growth (Jiang and Liu, 2010). In a study by Nouri *et al.* (2013), tomato plants were exposed to different concentrations of lead in contaminated soil. The results showed several negative impacts on plant growth and development such as reduced germination rate, stunted growth, chlorosis and leaf damage, altered nutrient uptake.

Proline builds up significantly under heavy metal stress and may play a significant part in osmotic adjustment, enzyme structure and function stability, organelle and cellular biomolecule stabilization (Ahmad *et al*., 2011). Nonetheless, a reduction in proline concentration has been noted in hydrophytes such as Ceratophyllum, Wolffia, and Hydrilla in response to Cd exposures (Dhir *et al*., 2004). Because they may neutralize ROS by giving electrons to hydrogen atoms, phenolic substances found in plants, such as flavonoids and lignin, are crucial in the fight against metal stress tolerance.

The phytophenolics have the ability to detoxify H_2O_2 produced in reaction to heavy metal stress by acting as antioxidants. Furthermore, under specific stressors, phytophenolics also function as pro-oxidants. According to reports, many plants treated with high amounts of Cd have higher phenolic acid contents (Sytar *et al*., 2013). Phytochelatins are cysteine-rich polypeptides that aid in metal chelation and are both heavy metal-inducible and heavy metal-binding. Heavy metal accumulation and phytochelatin synthesis have been found to positively correlate in a number of investigations. The phytochelatins are thiolic peptides that are produced primarily from glutamate, cysteine, and glycine by the heavy metal-activated enzyme phytochelatin synthase. These peptides then form complexes with the harmful metal ions

and are then carried to the vacuole (Komal *et al*., 2014). phytohormones such as auxin, cytokinin, and gibberellin can reduce the symptoms of stress By lowering heavy metal absorption and reestablishing growth and primary metabolite levels (Piotrowska-Niczyporuk *et al*., 2012).

The antioxidant capacity of plants developing under heavy metal stress is increased by phytohormones, which also increase the levels of nonenzymatic antioxidants like glutathione and ascorbate and the activities of antioxidative enzymes. An essential component of the signal transduction cascade that starts the plant stress response are phytohormones.

Metabolomics profiling of salinity stress

Worldwide, salinity is a common abiotic stressor that has a negative impact on crop plant yield (Negrao *et al*., 2017). Excessive salinity results in ion toxicity and reduced nutrient uptake, osmotic imbalance, and metabolic disorders that disrupt a variety of physiological processes and slow down the plants' overall growth (Meng *et al*., 2016). Plants react to these negative impacts by altering the metabolite pool, hormone balance, and gene expression at the cellular level.

A plant that is under stress uses less energy than it would otherwise. Alongside the shifts in the concentrations of Alteration of metabolites, including transcriptome and proteome, is also thought to be an adaptation mechanism used by plants. The rise in the cellular accumulation of suitable solutes, which comprises a variety of soluble and neutral organic molecules, is the metabolic reaction that can be best explained. In order to balance the buildup of Na+ in the vacuoles and extracellular spaces, the accumulation of suitable solutes can help lower the cytoplasm's water potential. Three interconnected factors are crucial to establishing tolerance: (1) prevention of damage, (2) restoration of homeostatic conditions under stress, and (3) potential slower growth (Sobhanian *et al*., 2010).

Osmoprotectants, ROS scavengers, and/or metabolites involved in energy metabolism are among the metabolites whose levels are changed by salinity. Salinity exposure affects the synthesis, storage, and transportation of a variety of primary and secondary metabolites (Fraire-Velazquez and Emmanuel, 2013). While the alterations in the metabolome as a whole in reaction to abiotic stressors have not yet been fully understood, the metabolic changes in response to salt are well characterized (Widodo *et al*., 2009). Because these metabolic and regulatory pathways are more negatively impacted by salinity, metabolites that serve as a foundation for plants to develop stress tolerance can be categorized as the metabolites of various metabolic and regulatory pathways, such as photosynthesis, amino acid biosynthesis, ROS scavengers, and the tricarboxylic acid cycle (TCA cycle). Upon exposure to stress conditions like salinity, photosynthesis is the most affected physiological process.

The plants' capacity to photosynthesize is reduced in high-salinity environments because there are fewer raw materials available as a result of the roots' diminished ability to absorb minerals and water (Grewal, 2010). Furthermore, the closing of stomata may be the cause of the decrease in photosynthesis since it lowers stomatal conductance, which in turn lowers the rate at which CO_2 diffuses. A decrease in transpiration rate may also be interpreted as a plant adaptation to lessen the amount of salt that is mobilized to the leaf tissues (Wu *et al*., 2013). Significant accumulation of 3-PGA signifies enhanced Calvin cycle under high salinity (Wu *et al*., 2013) The two most crucial Calvin cycle enzymes, phosphoribulokinase and sedoheptulose-1, 7-bisphosphatase, are in charge of controlling the cycle throughout the dark/light transition. The metabolites produced by the activities of these enzymes accumulate when they are upregulated in saline environments.

Salinity stress in sugar beet led to a rise in the amount of sugars and sugar derivatives, including trehalose, xylose, mannose, arabinose, inositol, and sucrose (Hossain *et al*., 2017).Trehalose, which may play a significant role in shielding photosynthetic proteins from this stressor combination, was particularly accumulated by tomato plants exposed to a combination of heat and salinity (Rivero *et al*., 2014). High salinity also negatively impacts energy metabolism processes like respiration and carbohydrate metabolism in addition to photosynthesis. In order to combat osmotic stress brought on by salt, plants have been shown to accumulate a variety of soluble carbohydrates, including sucrose, hexoses, raffinose, trehalose, mannobiose, and sugar alcohols. As sources of carbon and energy for the cells, soluble carbohydrates are essential to plant metabolism. Salt stress changes the amounts of soluble carbohydrates since the carbohydrate content is linked to several vital physiological functions, including respiration and photosynthesis. In addition to helping with turgor preservation, cell membrane stabilization against ROS effects, and protein breakdown prevention under stress, soluble sugars also function as osmoprotectants under osmotic stress (Lu *et al*., 2013).

The most crucial metabolites needed by plants exposed to saline environments are amino acids, which balance their cellular osmotic concentration and scavenge reactive oxygen species. According to reports, high salinity conditions raise the amounts of certain amino acids (Zhao *et al*., 2014). Plants' ability to withstand salt is measured by amino acids such valine, leucine, and threonine (Sanchez *et al*., 2008). Many plants exposed to high salinity exhibit a decrease in the quantity of arginine, methionine, and cysteine the main components of total free amino acids. Following the commencement of salinity stress, plants accumulate proline as an adaptation mechanism (Kumari *et al*., 2015). As an osmolyte and a scavenger of ROS, proline is an essential amino acid that builds up during salinity stress and shields

cells from harm caused by the salt. Additionally, proline serves as a molecular chaperone to preserve the stability and integrity of enzymes. In addition to altering the GABA shunt and causing the accumulation of different osmolytes like proline, prolonged salinity with high salt dosage also caused shikimate-mediated secondary metabolisms with elevated levels of aromatic amino acids like tyrosine, tryptophan, and phenylalanine (Zhang *et al*., 2011).

When plants are under stress, ABA serves as a vital transmitter to keep their water status stable. ABA is carried throughout the plants and accumulates in the tissues of the roots and leaves as a result of salinity stress. However, the pH of the xylem/apoplast affects ABA compartmentation, which in turn controls how much ABA the stomata receive. Thus, in stressful situations, ABA affects stomatal opening and shutting, regulating transpirational water loss. The generation of H_2O_2, an intermediary signal for stomatal closure, and the amount of Ca^{2+} in the guard cell's cytoplasm are linked to ABA-induced stomatal closure under salinity (Kim and Wang, 2010).

Metabolomics profiling of cold stress

Cold stress is one of the main elements that determines how plants are organized and how they turn out. It includes both high and low temperature shocks and is thought to be the main abiotic stressor for seedlings (Awasthi *et al*., 2015). Plant scientists are interested in temperature stress because of climate change, which has a negative impact on agricultural output globally. (Hasanuzzaman *et al*., 2013). Important physiological processes, such as the equilibrium between primary and secondary metabolites and hormones, or the link between water and membrane consistency, respiration, and photosynthesis, can be harmed by temperature increases Hemantaranjan, 2014). There is ultimately minimal economic gain as a result of the created disruptions, which hinder metabolic development and reduce plant growth and development.

For instance, amino acids are crucial for the production of various proteins, polyamines, phenylpropanoids, glucosinolates, auxins, and indole alkaloids during cold stress, as well as for N fixation into glutamine (Hildebrandt, 2018). Polyamines play a key role in protecting plants under cold stress. They are involved in stabilizing membrane structures and mitigating oxidative damage, especially in the case of freezing temperatures (Székvári, 2011). This amino acid accumulates in plants during stress, including cold stress. It acts as an osmoprotectant by stabilizing proteins and cellular structures, preventing cellular dehydration and oxidative damage. Proline accumulation is commonly observed in many horticultural crops like tomatoes and peppers (Naylor and Morgan, 1974).

Raffinose was found to be a potential biomarker of cold tolerance in Arabidopsis (Korn *et al*., 2010). Even though ecotypes influence the overall reaction. The majority of this species' heat shock metabolite reactions, including increases in amino acids derived from pyruvate and the TCA-cycle, were similar to those brought on by cold (Kaplan *et al*., 2004). With global temperatures rising heat is a significant abiotic stressor that consistently affects crops in many nations. It appears that low-temperature-activated signaling triggers modifications in cold-regulated gene expression, which contribute to the active reconfiguration of the metabolome.

Metabolomics mediated crop improvement

The ongoing development of cultivars that can withstand environmental disturbances and generate higher-quality and more abundant product is required because of the growing need for food and fodder. Breeding techniques that are quicker, more accurate, and less expensive are necessary in modern agriculture to increase crop quality and yields (Khakimov *et al*., 2014). This calls for the creation of biomarkers to assess the quality of finished goods, genetic modification, and high-throughput analytical methods such as metabolomics for crop breeding to increase crop yields under stress.There are numerous benefits of using metabolites as a primary plant phenotype. A valuable technique for examining relationships between phenotypes and QTLs, mQTLs, and Whole Genome Association Studies (GWAS) that use mQTL (mGWAS) to identify and assess changes in metabolic adaption under different stressors in metabolomics (Templer *et al*., 2017).

Metabolite profiling's ability to identify mQTLs and mGWAS has improved metabolomics' standing in metabolic marker-assisted plant breeding. Numerous mQTLs and mGWAS have discovered a correlation between metabolic content and genomic area in a number of crop species. Phytate in mustard, secondary metabolite composition in rice, vitamin A content, starch content, lignocellulosic biomass quality, and carotenoids in tomatoes, kernel composition, and multi-traits in sugar beets are a few examples. Utilizing mQTL/mGWAS to identify tomato fruit size, flavor, color, and nutrient content has demonstrated that the metabolomics approach is a practical method or instrument for understanding how plants respond to different environmental stressors. As a result, the findings helped breeders to increase crop resistance for several environmental stressors. Therefore, the accuracy and effectiveness of plant breeding are significantly improved by metabolically assisted varietal development (Christ *et al*., 2018; Hill *et al*., 2015).

Finding metabolic biomarkers for particular environmental conditions or plant growth stages may result from a thorough analysis of metabolites associated with plant growth and development as well as responsiveness to various stress situations. Nonetheless, the identification of metabolic

biomarkers requires the extensive application of univariate in addition to multivariate data analysis of metabolic data. To identify a group of biomarkers to distinct developmental stages and their function during pest interaction, GC-MS-based non-targeted metabolic profiling has been used in rice (Agarrwal *et al*., 2014). Throughout all of their developmental stages, tomato flesh and seeds showed altered metabolic makeup, which was useful for breeding actions that targeted particular stages. sugars, Fatty acids, organic acids and amino acids, were the main biomarker sets identified by the salinity stress adaptive metabolites of both wild and farmed soybeans (Li *et al*., 2019).

Finding metabolite indicators that are unique to stages and stress will aid in crop breeding efforts by providing a focused approach. Additionally, new insights on gene annotation are produced by combining data from genomics and metabolomics. Finding the genetic regions controlling the amount and quality of metabolites has been made easier with the use of the integrated omics method (Abdelrahman *et al*., 2019).

Conclusion

The metabolome plays a pivotal role in enhancing crop resilience to abiotic stress by providing insights into the biochemical and physiological responses plants deploy to survive adverse conditions. Key metabolites, such as osmoprotectants, antioxidants, and secondary metabolites, act as defense agents, helping plants mitigate damage caused by stressors like drought, salinity, heat, and nutrient imbalances. These molecules contribute to maintaining cellular homeostasis, protecting macromolecules, and sustaining metabolic flux under stress conditions. By integrating metabolomics with advanced breeding and biotechnological approaches, researchers can identify stress-resilient crop varieties. Tools such as genome-wide association studies (GWAS) and gene editing technologies like CRISPR-Cas9 can be leveraged to enhance or engineer metabolic pathways linked to resilience. Furthermore, metabolomics aids in understanding the dynamic interactions between plants and their environment, enabling the development of more targeted and sustainable agricultural practices. In summary, the metabolome serves as both a diagnostic tool and a functional component in building crop resilience. Its study not only deepens our understanding of plant stress physiology but also opens avenues for developing climate-resilient crops, which are essential for global food security in the face of environmental challenges.

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Conflict of Interest

The authors have no conflict of interest.

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Research Article

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Consequences of front line demonstration of Tomato cv. Arka Rakshak in Pali district of western Rajasthan

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ABSTRACT

Tomato plays an important role in supplementing the income of small and marginal farmers of major area of the India. The main constraints of low quality and productivity of this vegetable in arid region of Rajasthan (India) may be due to partial adoption of improved production technology practices by the tomato growers. The present study was undertaken by ICAR-CAZRI, Krishi Vigyan Kendra, Pali-Marwar (Rajasthan) to address the yield gap through Front Line Demonstrations (FLDs) on tomato crop during three different years (2019-20, 2022-23 and 2023-24) at three selected villages (total 45 demonstrations) of the Pali district. Prevailing farmer's practices were treated as control for comparison with recommended practices. In the three years data it was observed that improved practices helped in managed the incidence of pest and diseases, increased productivity and quality of the produced. Due to this an average yield of 429.17 q/ha was obtained in demonstrated plot over control (364.23 q/ha) with an additional yield of 64.94 q/ha and the increasing the average tomato productivity by 17.75 per cent. The average extension gap and technology gap ware 64.94 q/ha and 320.83 q/ha, respectively, with the average technology index of 42.77 per cent during the demonstration years. Besides this, the demonstrated plots gave higher gross return (Rs. 5,31,871), net return (Rs. 3,60,704) with higher benefit cost ratio (3.08) when compared to farmer's practice. The performance of demonstrated package of practices even though after FLD programme, which shows positive impact of FLD on adoption of demonstrated production technology.

Introduction

Tomato (*Solanum lycopersicum*) is grown almost throughout the world including tropical and temperate regions. It is cultivated both in the green houses on protective structures as well as under natural conditions. It ranks first among processed vegetables. It is consumed fresh in salad, fried in culinary preparations and processed in various forms *viz*. Ketchup, sauces, puree, paste, powder, juice, soup and chutney etc. The fast foods such as pizza, burger, noodles, *etc*. will not taste the same without addition of tomato sauces. Tomato is a rich source of vitamins A and C and is referred to as "poor man's orange". It adds variety of colours to the food. Tomato is a very good appetizer and its soup is said to be a good remedy for patients suffering from constipation. Lycopene that imparts red colour to ripe tomatoes is reported to possess anti-cancerous properties. It also serve as a natural anti-oxidant as the Beta-carotene functions to help prevent

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and neutralize free radical chain reaction and ascorbic acid is an effective scavenger of superoxide, hydrogen peroxide, singlet oxygen and other free radicals. It is one of the most sensitive vegetable crops and fails miserably if growing conditions are too harsh. It is highly sensitive to frost and dry and hot weather results in flower drops and poor fruit set. In India during 2021-22 it was cultivated in 8.41 million hectare area with a production of 203.36 Lakh Tonne (Anonymous, 2021-22). In Rajasthan its area and production were 18,120 hectare and 88,730 tonnes respectively. In Rajasthan the productivity of tomato was recorded 4.90 t/ha, which was almost three time lower than the India's productivity *i.e.* 25.04 t/ha (Anonymous, 2017-18). Average productivity of tomato crop is quite low and there exists a good scope to improve its average productivity in Rajasthan as well as in India to fulfil both domestic and national needs. The growth, yield and fruit quality of tomato are largely dependent on number of interacting factors. On the other hand tomato is a long duration crop with high yield which removes large quantities of nutrients from the soil. Like macronutrients, micronutrients are equally significant in plant nutrition. There is a need to go for balanced fertilization of both macro and micronutrients since micronutrients play a profound role in various metabolic functions of plant. The other reason of low productivity it might be due to the unavailability of disease resistant varieties, the farmers of Pali district were facing a reduction in tomato yield as well as quality.

The main objective of front line demonstration (FLD) is to introduce suitable agriculture practices like high yielding varieties, disease resistant variety, seed treatment, spacing, timely sowing, nutrient management including micronutrients, growth hormones, pest and disease management etc. among the farmers accompanied with organizing extension programmes (field day) for horizontal dissemination of the technologies. FLD is playing a very important role for transfer of technologies and changing scientific treatment of the farmers by seeing and believing principle. In order to have better impact of the demonstrated technologies for farmers and field level extension functionaries, Front Line Demonstrations was conducted at farmer's field, in a systemic manner, to show case the high yielding new varieties, to convince them to about the potential of improved production technologies to enhance yield of tomato. Generally, the agricultural technology is not accepted by the farmers as such in all respects. There is always gap between the recommended technology by the scientist and its modified form at the farmer's level which is major absentee in the efforts of increasing agricultural production in the country. It is need of the hour to reduce this technological gap between the agricultural technology recommended by the scientists or researchers and its acceptance by the farmers on their field. In view of the above facts, front-line demonstrations were undertaken in a systematic manner on farmer's field to show the worth of improved practices and convince the farmers to adopt in their farming system.

Material and Methods

The Krishi Vigyan Kendra (Farm Science Centre), a cutting-edge scientific organisation, is crucial in connecting research experts and farmers. The primary goal of KVK, Pali is to shorten the time gap between technology production at the research facility and its distribution to the farmers in order to steadily increase productivity and income from the agricultural and related sectors. Front Line Demonstration is one of these powerful tools for technology transfer since it demonstrates in real life the power of new technologies to increase yield and profit. The purpose of the current study was to determine the effects of improved variety on Arka Rakshak (First F_1 hybrid with triple disease resistance to tomato leaf curl virus, bacterial wilt and early blight) with improved production technology on farmer households' socioeconomic development. The soil of the farmer's field was sandy clay loam in texture while depth of soil is moderate too deep about 50 to 75 cm. It is suitable for cultivation but for low rainfall and high evaporation causes saline (pH 7.93 to 8.20) nature. Organic carbon at the farm field soil ranges from 0.22 to 0.33% and Nitrogen in surface layer is low (231.75 to 277.00 kg/ha) whereas P_2O_5 (14.33 to 15.00 kg/ha) and K_2O (210.33 to 214.33 kg/ha) is medium. The mean minimum and maximum annual temperature was 4.1°C and 41.2°C, respectively and total mean rainfall was 323.21, 450.30 and 552.60 mm during the demonstration year 2019-20, 2022-23 and 2023-24, respectively.

In total, 45 demonstrations were conducted at 45 farmer fields in five chosen villages (Dayalpura, Kurki, Kanawas, Sinla and Ras) of the Jaitaran block of Pali (Rajasthan) where farming situation was irrigated medium soil. Every frontline demonstration was set up on 0.1 ha of land, with the nearby 0.2 ha serving as the comparison control (farmer's practise). By providing them quality training in various aspects of tomato production, technical guidance for agricultural inputs (seed of Arka Rakshak, fertilizers and plant protection) and marketing (harvesting, grading and packing) of tomato. To illustrate the results of the front line demonstration to the farmers of the same village and neighbouring villages, field days were also conducted in each cluster.

KVK scientists, collected information (data) from the demonstration and farmers' practice on production costs and returns through repeated field visits from front-line demonstration plots and farmers' execution plots and analysed with using simple statistical techniques. After that, average yield, extension gap, technology gap, technology index, cost of cultivation, net returns, and benefit–cost ratio were computed. An average of cost of cultivation, yield and net returns of different farmers was analysed by the formula as given below.

Table 1. Use of improved production technology of tomato in the study area and adoption gaps

S. No.	Practices (Technology intervention)	Demonstrated Improved practices (IP)	Farmer's practices
1.	Variety	Arka Rakshak (Triple disease resistant hybrid variety)	Private hybrids
2.	Soil testing	Have done in all locations	Not in practice
3.	Seed rate and sowing method	200 g/ha and line sowing in nursery bed	400-500 g/ha and broadcasting
4.	Seed treatment	Seed was treated with Carbendazim	Not in practice
5.	Neem cake application	Apply @ 250 kg/ha before final bed preparation	Not in practice
6.	Transplanting method	Transplanting in raised bed distance at 90 cm x 60 cm	Flat bed transplanting at 75 cm x 45 cm spacing
7.	Mulching	Follow the silver black plastic mulching	Not in practice
8.	Sowing & transplanting time	1st week of September & 2nd week of October	1st week of September & 2nd week of October
9.	Irrigation method	Micro/Drip system	Channel/ furrow method (surface irrigation)
10.	Fertilizer dose	On the basis of soil test recommendation	Application without recommendation
11.	Weedicide dose	Pendimethalin @ 1.0 kg/ha was applied immediately after transplanting	Hand weeding/ rarely used
12.	Training of plant	Stake the plant after 30 days of transplanting and remove the branches up to 30 cm height	Stake the plant at flowering stage and no removal of branches
13.	Multiplex nutrient spry	@ 2.5 g/ litter water and three (03) spray. First spray just before flowering, second spray during flowering or 25 days after first spray and third spray when fruits are bean size	No application of any type of supplement/ micronutrient
14.	Plant protection measures	Need based three spray of Imidacloprid 17.8 % SL or Thiamethoxam 25 WP (0.3 g/l) and other systematic fungicide	Irregular use of chemicals
15.	Other plant protection measures	Install of yellow sticky traps at appropriate period for indication of white flies and regular spray of neem oil	Not followed

Average = $[F_1 + F_2 + F_3 .....Fn] / N$ F_1 = Farmer, N = No. of farmers (45)

Gross return was calculated by multiplying yield into prevailing local market price of the fruit obtained by the farmers. The technology gap and technological index along with the benefit cost ratio were calculated by using following formula as given below.

The data thus collected were tabulated and statistically analysed (Kumar and Singh, 2023) to interpret the FLD's results.

Results and Discussion

The data were analysed, and the technology gap, extension gap, and technology index were calculated according to the formula and an economic analysis was performed according to procedure, with the results presented in Table 2 and 3.

Yield analysis

The perusal of data (Table 2) indicate that due to initiation of front line demonstrations the tomato yield ranged from 402.67 q/ ha to 454.34 q/ha in demonstration practice plots and from 351.78 q/ ha to 376.4 q/ha in farmer's practice plot in three years of demonstrations conducted. An average yield of 429.17 q/ha was obtained under demonstration practice plots as compared to farmer's practice plots 364.23 q/ha in

consecutively. The per cent increase in yield over farmer's practice was highest (20.70) during 2023-24. However variations in the yield of tomato in different years might be due to the variations in soil moisture availability, improved variety (Arka Rakshak), improved production techniques and change in the location of demonstrations every year. The average yield of tomato is increased by 17.75 per cent over the yield obtained under farmer's practices of tomato cultivation. The result revealed the positive effects of FLD over the farmer's practices as it enhanced the yield of tomato in Jaitaran block of Pali (Rajasthan). The improved tomato yield in the demonstration practices was attributed primarily to the use of improved hybrids Arka Rakshak with improved technologies such as seed treatment, mulching, transplanting methods, spacing, balanced nutrient application and spray including secondary and micronutrients, integrated pest and disease management, weed management and irrigation methods. The results confirm the findings in different crops by Misra *et al*. (2019), Chaitanya *et al*. (2020), Parmar *et al*. (2020), Rathod *et al*. (2022) and Singh *et al*. (2022).

Extension gap

Extension gap of 50.89, 66 and 77.94 q/ha was observed (Table 2) during 2019-20, 2022-23 and 2023-24 respectively. On an average extension gap in three years FLD programme was 64.94 q/ha. This emphasized the need to educate the farmers through various techniques for the adoption of improved agricultural production technologies to reverse this trend of wide extension gap. More and more use of latest production technologies like trellising in tomato with high yielding variety/hybrid will subsequently change this alarming trend of galloping extension gap. Similarly, extension gap in different location in front line demonstrations were documented by Misra *et al*. (2019), Chaitanya *et al*. (2020), Parmar *et al*. (2020), Rathod *et al*. (2022) and Singh *et al*. (2022) in tomato and other crops.

Technology gap

The technology gap, the differences between potential yield and yield of demonstration practice plots was 347.33, 319.50 and 295.66 q/ha (Table 2) during 2019-20, 2022-23 and 2023-24, respectively. On an average technology gap under three year FLD programme was 320.83 q/ha. This may be attributed to dissimilarities in soil fertility, salinity and to erratic rainfall and other vagaries of weather in the demonstration area. Hence, location specific recommendations may become necessary to narrow down the gap. These findings are similar to the finding of Singh *et al.* (2018), Rai *et al*. (2019), Misra *et al*. (2019), Yadav and Tripathi (2019) in other crops.

Technology index

The technology index shows the feasibility of the demonstrated technology at the farmer's field. The technology index varied from 39.42 to 46.31 (Table 2). On an average technology index of 42.77 per cent was observed during the three years of FLD programme, which shows the effectiveness of technical interventions. This accelerates the adoption of demonstrated technical interventions to increase the yield performance of tomato. The results of the present study are in consonance with the finding Singh *et al.* (2018), Rai *et al*. (2019), Misra *et al*. (2019), Yadav and Tripathi (2019), Rathod *et al*. (2022) and Singh *et al*. (2022). From these results it is evident that the performance of the technology demonstrated was found to be better than the farmer's practice under same environment conditions. The farmers were motivated by seeing the results in term of productivity and they are adopting the technologies. The yield of the front line demonstrations and potential yield of the crop was compared to estimate the yield gaps which were further categorized into technology index and technology gap.

Economic returns

In order to find the economic feasibility of the demonstration technologies over and above the control, some economic indicators like cost of cultivation, net return and B: C ratio was worked out. The economic viability of improved demonstrated practices over farmer's practices was calculated depending on prevailing price of inputs and outputs cost and represented in terms of B: C ratio (Table 3). It was found that the cost of production of tomato under demonstration practices varied from of Rs.1,51,200 to 1,92,200 per ha with an average of Rs.1,71,167 per ha as against Rs.1,42,300 to 1,84,500 per ha with an average Rs.1,63,650 per ha under farmer's practice. The additional cost increased in demonstration was mainly due to more cost involved in balanced fertilizer application, procurement of improved hybrid seed and IPM practices. The cultivation of tomato under improved technologies gave higher net return of Rs. 2,91,737 per ha, Rs. 3,46,500 per ha and Rs. 4,43,876 per ha in the year 2019-20, 2022-23 and 2023-24 respectively with an average net return of Rs. 3,60,704 per ha while in farmer's practices it was Rs. 2,86,789 per ha. The average additional net return was Rs. 73,915 per ha and the benefit cost ratio of tomato ranged from 2.92 to 3.30 in demonstration practice plots and from 2.66 to 2.85 in farmer's practice plots during three years of demonstration with an average of 3.08 in demonstration and 2.74 under farmer's practices. This may be due to higher yield obtained and lower cost of cultivation under improved technologies compared to farmer's practice. The B: C ratio was recorded to be higher under demonstration against control during all

the years of study. Extension agencies in the district need to provide proper technical support to the farmers through different extension methods to reduce the extension gap for better tomato production in the Jaitaran block of Pali (Rajasthan). These results are in accordance with findings of Singh *et al.,* (2018), Rai *et al.* (2019), Yadav and Tripathi (2019), Misra *et al.* (2019), Parmar *et al.* (2020), Rathod *et al.* (2022) and Singh *et al.* (2022) in different crops.

Table 2. Productivity and gap analysis of frontline demonstration of improved production technology of tomato

Years	Area (ha)	No. of farmers	Yield (q/ha)			Additional yield over FP (kg/ha)	Increase in yield (%)	EG (q/ha)	TG (q/ha)	TI (%)
			PY	DP	FP					
2019-20	3.00	15	750	402.67	351.78	5089.00	14.46	50.89	347.33	46.31
2022-23	2.40	12	750	430.50	364.50	6600.00	18.10	66.00	319.50	42.60
2023-24	3.60	18	750	454.34	376.40	7794.00	20.70	77.94	295.66	39.42
Average	-	-	750	429.17	364.23	6494.34	17.75	64.94	320.83	42.77

PY = Potential yield, DP = Demonstrated practice, FP = Farmer's practice, EG = Extension gap, TG = Technology gap, TI = Technology index

Table 3. Comparative B: C analysis of tomato under demonstration practice and farmer's practice

Years	Cost of cultivation		Gross return (Rs./ha)		Net return (Rs./ha)		Additional net return (Rs./ha)	B:C ratio	
	DP	FP	DP	FP	DP	FP		DP	FP
2019-20	1,51,200	1,42,300	4,42,937	3,86,958	2,91,737	2,44,658	47,079	2.92	2.71
2022-23	1,70,100	1,64,150	5,16,600	4,37,400	3,46,500	2,73,250	73,250	3.03	2.66
2023-24	1,92,200	1,84,500	6,36,076	5,26,960	4,43,876	3,42,460	1,01,416	3.30	2.85
Average	1,71,167	1,63,650	5,31,871	4,50,439	3,60,704	2,86,789	73,915	3.08	2.74

Average rate in 2019-20 = Rs. 1100/q, 2022-2023 = Rs. 1200/q and 2023-24 = Rs. 1400/q

Conclusion

The study clearly demonstrated that demonstration practice plots consistently achieved higher yields compared to farmer's traditional practices. This was primarily due to the adoption of a new variety, better knowledge dissemination, and the implementation of a complete package of improved practices. The FLDs significantly impacted productivity and profitability, highlighting the potential of modern technology in real farming conditions. The FLDs conducted by KVK, Pali, played a crucial role in the horizontal spread of improved tomato cultivation techniques. To further enhance technology adoption, targeted training programs on improved vegetable production techniques, coupled with multiple demonstrations, are essential. This approach can help overcome challenges in the existing technology transfer system in the Jaitaran block of Pali district, Rajasthan. The success of FLDs in tomato cultivation serves as an inspiration for non-tomato growers to embrace improved cultivation methods, ultimately contributing to the overall growth of vegetable farming in the region.

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Conflict of Interest

The authors have no conflict of interest.

Data Sharing

All relevant data are within the manuscript.

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Research Article

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Phytochemical analysis of *Momordica balsamina* L. fruits

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ABSTRACT

Momordica balsamina L., commonly known as balsam apple or African pumpkin, is a vegetable with high nutritional value. It contains bioactive compounds with medicinal properties and has been extensively used in traditional medicine to treat a range of ailments, including malaria, fevers, and diabetes. This study focused on analyzing the bioactive compounds in *Momordica balsamina* fruits using GC-MS/MS. A total of 100 bioactive compounds were detected in the green fruits, while 109 were identified in the ripe fruits. These compounds exhibit significant pharmacological properties and have the potential to be used in the treatment of various human diseases.

Introduction

The balsam apple, also known as the African pumpkin (*Momordica balsamina* L.), is considered one of the most significant medicinal plants, widely used as a source of life-saving treatments for people around the world (Hassan and Umar, 2006; Thakur *et al.*, 2009 and Souda *et al.*, 2018). This crop can be consumed as a vegetable to supplement protein and potassium in the diets of poor rural communities. Its high potassium content is also associated with the treatment of hypertension and other cardiovascular diseases (Souda *et al.*, 2018). *M. balsamina* is useful in the production of dietary supplements because of its high protein and fat content and low fibre level.

The extracts from *Momordica* spp. are known for their reputed bioactivities, including antidiabetic, antimicrobial, anthelmintic, abortifacient, antibacterial, and antiviral properties. Various parts of *Momordica* species are rich in both primary and secondary metabolites. The primary metabolites derived from *Momordica* spp. include sugars, proteins, and chlorophyll (Hassan and Umar, 2006 and Muronga *et al.*, 2021), while the secondary metabolites consist of alkaloids, flavonoids, and tannins (Madala *et al.*, 2016; Muronga *et al.*, 2021; Jadhav and Kamble, 2022 and Choudhary *et al.*, 2022). *Momordica* spp. contain "Momordin", a therapeutic agent known for its ability to inhibit the replication of HIV and other viruses. Additionally, it functions as both an anti-diabetic and anti-cancer agent (Thakur *et al.*, 2009).

Fully exploring the effectiveness of *Momordica balsamina* plant extracts could lead to new drug discoveries or enhance the use of indigenous herbal medicines in conventional treatments for certain diseases (Karumi *et al.*, 2003; Bhardwaj *et al.*, 2010 and Rahmatullah *et al.*, 2012). The aim of this study was to analyze the biochemical and medicinal properties of

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Momordica balsamina, assessing its potential as a promising and innovative source of natural bioactive compounds for future pharmaceutical applications and promotion/ popularization of this neglected crop.

Material and Methods

Mature, green and full-ripened fruits of *M. balsamina* genotype CIAHMB-1 were harvested at peak fruiting season during August 2020, from the Experimental Farm of the ICAR Central Institute for Arid Horticulture, Bikaner, Rajasthan, India, at a latitude of 28°N and longitude of 73°18'E and an altitude of 234.84 m amsl. After the fruits were harvested, they were sun-dried for 6 days at room temperature. After drying, they were ground into a powder and stored at -20°C until further analysis. The samples were analyzed in January 2021 in the Food Testing Laboratory, Department of Biotechnology, Junagadh Agricultural University, Junagadh, Gujarat.

For analysis, 200 mg of the dried and powdered fruit sample was homogenized in a pre-chilled mortar and pestle with 3 ml of ice-cold HPLC-grade methanol (100%). The homogenate was incubated in a water bath at 70°C for 10 minutes, with continuous shaking at 950 rpm. After that, the mixture was centrifuged at 11,000 g for 10 minutes. The supernatant resulting from this step was divided into two aliquots. One portion was used in TAA determinations, and the other portion was transferred to a Schott GL14 glass tube. To this, were added 1.5 ml pre-chilled chloroform and 3 ml ice-cold deionized water (4°C), and then mixed on a vortex for 20 seconds. After centrifugation at 2200 g for 15 minutes, the top (polar) and bottom (non-polar) phases were transferred into separate test tubes and dried to dryness under a stream of nitrogen. All reagents and chemicals used in this present study were standard make and quality. Ultrapure water of 18.2 MΩ·cm was used in all assays through the Milli-Q Simplicity, Millipore, France.

Bioactive compounds of the methanolic extract were quantified using GC-MS/MS (GCMSQP2010Plus, Shimadzu). An aliquot of 4 µl was injected into the DB 17MS capillary column 30 m x 0.25 mm. Injection temperature at 280°C with a 5-minute solvent delay and an initial GC oven temperature of 65°C, which was raised after 2 minutes to 290°C. The temperature of the ion source was 230°C. Helium was used as the carrier gas at a constant flow of 1 ml/min. Measurements were obtained with electron impact ionization at a setting of 70 eV and in full scan mode (m/z 50–900) at a rate of 2000. Tentative identification of the phytochemicals was determined based on the retention times and mass spectra of the standards compared to the NIST 14 library. Baseline correction, alignment, peak picking, and integration on the total ion chromatograms were performed using ACD/Spec Manager v. 12.00, while data analysis was done with CSV comma-delimited files.

Result and Discussion

The non-targeted phytochemical profiling of *Momordica balsamina* through GC-MS/MS analysis of methanolic extracts from dried fruits (both green and ripe) led to the detection and identification of numerous compounds. These compounds were verified by matching their mass spectra with entries from the NIST 14 library. Tables 1 and 2 provide detailed information on the compounds identified, including retention time (in minutes), peak area percentage, compound names, molecular formulas, and molecular weights. The height of each peak corresponds to the relative concentration of the bioactive compounds. The GC-MS/MS chromatograms revealed a total of 100 compounds in the green fruits (Table 1) and 109 compounds in the ripe fruits (Table 2).

The analysis identified several key phytochemicals in *Momordica balsamina* fruits, including 3',5'-Dimethoxyacetophenone, Ar-tumerone, Ascorbic acid, Heptadecanoic acid, Cyclopropanedecanoic acid 2-hexyl, Cyclononasiloxane (octadecamethyl-), 4,7,10-Hexadecatrienoic acid, 8,11,14-Eicosatrienoic acid (Z,Z,Z), 2-(Dimethylamino) ethyl adamantanecarboxylate, Dotriacontane, 2,6,10,14,18,22-Tetracosahexaene, 6,9,12,15-Docosatetraenoic acid, and Bisabolen-12-OL. According to the literature, many of these compounds exhibit biological activity and have been linked to nutraceutical and pharmaceutical applications, with potential therapeutic effects against major diseases such as cancer, diabetes, cardiovascular disorders, and other chronic conditions (Bot *et al.*, 2007; Spengler *et al.*, 2009; Thakur *et al.*, 2009; Souda *et al.*, 2018; Muronga *et al.*, 2021 and Choudhary *et al.*, 2022).

Among the detected compounds, a substantial portion (approximately 60%) consisted of fatty acids, including monounsaturated fatty acids (MUFA), polyunsaturated fatty acids (PUFA), and some unusual fatty acids with odd-numbered carbon chains. Many of these compounds have demonstrated notable bioactivities, including antimicrobial, anti-inflammatory, anticancer, analgesic, antipyretic, antidiabetic, hepatoprotective, cardiovascular, antioxidant, and anti-mutagenic effects (Souda *et al.*, 2018; Muronga *et al.*, 2021; Choudhary *et al.*, 2022). Several studies have also highlighted the bioactive compounds in *M. balsamina*, such as Cucurbitane glycoside and Kuguacin J, which have proven anti-diabetic properties (Spengler *et al.*, 2009), and Karavilagenin C, Balsaminagenin A, and Balsaminoside B, known for their anti-cancer potential (Ramalhete *et al.*, 2009). Additionally, Momordin I and Momordin II have been shown to possess antiviral activity (Thakur *et al.*, 2009), while Balsaminapentaol has demonstrated anti-malarial effects (Ramalhete *et al.*, 2009).

Table 1: Compounds detected in methanolic extract of *M. balsamina* (dried green fruits) through GC-MS/MS analysis

Peak No.	R. Time (minute)	Area (%)	Height (%)	Compound	Molecular formula	Molecular weight
1	3.778	0.22	0.29	3,3-Dimethoxy-2-butanone	$C_6H_{12}O_3$	132
2	5.162	0.04	0.07	1,2-Cyclopentanedione	$C_5H_6O_2$	98
3	6.158	0.49	0.21	Ethane, 1,1,1-triethoxy-	$C_8H_{18}O_3$	162
4	6.285	1.06	0.49	Phenol	C_6H_6O	94
5	6.688	0.44	0.28	Decane	$C_{10}H_{22}$	142
6	6.875	0.27	0.16	Glycerin	$C_3H_8O_3$	92
7	8.300	0.05	0.03	2-Pyrrolidinone	C_4H_7NO	85
8	8.775	0.05	0.05	3,3-Diethoxy-1-propyne	$C_7H_{12}O_2$	128
9	8.892	0.07	0.07	Phenol, 2-methoxy-	$C_7H_8O_2$	124
10	10.470	0.38	0.41	Silane, triethylmethoxy-	$C_7H_{18}OSi$	146
11	11.475	0.03	0.05	Dimethyl, fluoromethyl, phenylsilane	$C_9H_{13}FSi$	168
12	12.095	0.63	0.52	1,4:3,6-Dianhydro-.alpha.-d-glucopyranose	$C_6H_8O_4$	144
13	12.636	0.04	0.05	1-Dimethyl(octyl)silyloxypropane	$C_{13}H_{30}OSi$	230
14	13.081	0.17	0.17	1-Butanone, 1-phenyl-	$C_{10}H_{12}O$	148
15	14.175	0.13	0.11	Cyclohexasiloxane, dodecamethyl-	$C_{12}H_{36}O_6Si_6$	444
16	14.466	0.19	0.31	2-Methoxy-4-vinylphenol	$C_9H_{10}O_2$	150
17	15.861	0.22	0.31	DL-Proline, 5-oxo-, methyl ester	$C_6H_9NO_3$	143
18	15.953	0.07	0.11	Benzoic acid, 4-methoxy-, methyl ester	$C_9H_{10}O_3$	166
19	17.220	0.04	0.06	5-Dimethylsilyloxytetradecane	$C_{16}H_{36}OSi$	272
20	18.050	0.14	0.12	Cycloheptasiloxane, tetradecamethyl-	$C_{14}H_{42}O_7Si_7$	518
21	18.367	0.22	0.11	Ethanone, 1-(4-hydroxy-3-methoxyphenyl)-	$C_9H_{10}O_3$	166
22	18.612	0.65	0.25	1,6-Anhydro-.beta.-D-glucopyranose (levoglucosan)	$C_6H_{10}O_5$	162
23	18.879	0.03	0.05	Phenol, 2,4-bis (1,1-dimethylethyl)-	$C_{14}H_{22}O$	206
24	19.232	0.07	0.10	Dodecanoic acid, methyl ester	$C_{13}H_{26}O_2$	214
25	20.009	0.20	0.28	3',5'-Dimethoxyacetophenone	$C_{10}H_{12}O_3$	180
26	21.462	0.03	0.07	Cyclooctasiloxane, hexadecamethyl-	$C_{16}H_{48}O_8Si_8$	592
27	22.157	0.14	0.17	Ar-tumerone	$C_{15}H_{20}O$	216
28	22.389	0.08	0.15	1-Pentadecanol	$C_{15}H_{32}O$	228
29	22.667	0.04	0.08	Cycloheptasiloxane, tetradecamethyl-	$C_{14}H_{42}O_7Si_7$	518
30	22.821	0.16	0.22	E-7-Tetradecen-1-ol	$C_{14}H_{28}O$	212
31	23.139	0.22	0.37	Tridecanal	$C_{13}H_{26}O$	198
32	23.286	0.32	0.63	Methyl tetradecanoate	$C_{15}H_{30}O_2$	242
33	23.930	0.27	0.41	Tetradecanoic acid	$C_{14}H_{28}O_2$	228
34	24.401	0.03	0.06	Cyclononasiloxane, octadecamethyl-	$C_{18}H_{54}O_9Si_9$	666
35	25.170	0.30	0.45	Pentadecanoic acid, methyl ester	$C_{16}H_{32}O_2$	256
36	25.492	0.09	0.15	2-Pentadecanone, 6,10,14-trimethyl-	$C_{18}H_{36}O$	268

37	25.736	0.23	0.25	Cyclopropane, 1-methyl-1-(1-methylethyl)-2-nonyl-	$C_{16}H_{32}$	224
38	26.308	0.03	0.06	Hexadecanoic acid, methyl ester	$C_{17}H_{34}O_2$	270
39	26.509	0.06	0.11	9-Hexadecenoic acid, methyl ester, (Z)-	$C_{17}H_{32}O_2$	268
40	26.587	0.46	0.69	9-Hexadecenoic acid, methyl ester, (Z)-	$C_{17}H_{32}O_2$	268
41	26.756	0.03	0.06	2-Dodecen-1-yl(-)succinic anhydride	$C_{16}H_{26}O_3$	266
42	26.980	5.14	6.86	Hexadecanoic acid, methyl ester	$C_{17}H_{34}O_2$	270
43	27.221	0.20	0.14	Cyclopentadecanone, 2-hydroxy-	$C_{15}H_{28}O_2$	240
44	27.655	6.23	4.51	l-(+)-Ascorbic acid 2,6-dihexadecanoate	$C_{38}H_{68}O_8$	652
45	28.267	0.10	0.20	Cyclopropaneoctanoic acid, 2-hexyl-, methyl ester	$C_{18}H_{34}O_2$	282
46	28.668	0.26	0.48	Heptadecanoic acid, methyl ester	$C_{18}H_{36}O_2$	284
47	28.933	0.03	0.05	BISABOLEN-12-OL <BETA-> DB5-2242	$C_{15}H_{26}O$	222
48	29.003	0.19	0.35	Hexadecanoic acid, 2-hydroxy-, methyl ester	$C_{17}H_{34}O_3$	286
49	29.357	0.05	0.07	2-Cyclohexen-1-one, 4-hydroxy-3,5,5-trimethyl-4-(3-oxo-1-butenyl)-	$C_{13}H_{18}O_3$	222
50	29.822	8.25	8.62	9,12-Octadecadienoic acid (Z,Z)-, methyl ester	$C_{19}H_{34}O_2$	294
51	29.922	6.32	7.81	9-Octadecenoic acid, methyl ester, (E)-	$C_{19}H_{36}O_2$	296
52	30.316	3.84	5.75	Octadecanoic acid, methyl ester	$C_{19}H_{38}O_2$	298
53	30.499	7.23	5.30	9,12-Octadecadienoic acid (Z,Z)-	$C_{18}H_{32}O_2$	280
54	30.597	7.68	5.51	Octadec-9-enoic acid	$C_{18}H_{34}O_2$	282
55	30.834	0.74	0.99	9,12,15-Octadecatrienoic acid, ethyl ester, (Z,Z,Z)-	$C_{20}H_{34}O_2$	306
56	30.929	3.12	2.08	Octadecanoic acid	$C_{18}H_{36}O_2$	284
57	31.182	0.82	0.87	Hexadecanamide	$C_{16}H_{33}NO$	255
58	31.442	0.61	0.55	Cyclopropanedecanoic acid, 2-hexyl-, methyl ester	$C_{20}H_{38}O_2$	310
59	31.507	0.42	0.56	9-Hexadecenoic acid, methyl ester, (Z)-	$C_{17}H_{32}O_2$	268
60	31.575	0.53	0.55	Cyclononasiloxane, octadecamethyl-	$C_{18}H_{54}O_9Si_9$	666
61	31.667	0.96	0.62	9-Octadecenoic acid (Z)-, methyl ester	$C_{19}H_{36}O_2$	296
62	32.057	11.01	8.74	9,12,15-Octadecatrienoic acid, methyl ester	$C_{19}H_{32}O_2$	292
63	32.242	0.95	1.10	4,7,10-Hexadecatrienoic acid, methyl ester	$C_{17}H_{28}O_2$	264
64	32.595	3.18	2.68	8,11,14-Eicosatrienoic acid, (Z,Z,Z)-	$C_{20}H_{34}O_2$	306
65	32.714	3.00	3.03	9,12,15-Octadecatrienoic acid, methyl ester	$C_{19}H_{32}O_2$	292
66	32.901	0.51	0.64	Hexadecanoic acid, 2-hydroxy-1,3-propanediyl ester	$C_{35}H_{68}O_5$	568

67	32.992	1.19	1.34	11-Eicosenoic acid, methyl ester	$C_{21}H_{40}O_2$	324
68	33.272	0.28	0.30	Dodecanoyl chloride	$C_{12}H_{23}ClO$	218
69	33.366	1.16	1.53	Eicosanoic acid, methyl ester	$C_{21}H_{42}O_2$	326
70	33.770	0.53	0.80	METHYL LINOLEATE DB5-2915 (=METHYL (Z,Z)-9,12-OCTADECADIENOATE)	$C_{19}H_{34}O_2$	294
71	33.856	0.40	0.55	9-Octadecenamide, (Z)-	$C_{18}H_{35}NO$	281
72	34.211	0.23	0.16	Octadecanamide	$C_{18}H_{37}NO$	283
73	34.679	0.20	0.16	9,12-Tetradecadien-1-ol, acetate, (Z,E)-	$C_{16}H_{28}O_2$	252
74	34.800	0.13	0.17	Heneicosanoic acid, methyl ester	$C_{22}H_{44}O_2$	340
75	34.907	0.14	0.21	1,9,12,15-Octadecatetraene, 1-methoxy-	$C_{19}H_{32}O$	276
76	35.073	0.17	0.22	1H-Benzocyclohepten-7-ol, 2,3,4,4a,5,6,7,8-octahydro-1,1,4a,7-tetramethyl-, cis-	$C_{15}H_{26}O$	222
				77 35.152		
77	35.152	0.39	0.55	Carbamic acid, 2-(dimethylamino) ethyl ester	$C_5H_{12}N_2O_2$	132
78	35.225	0.15	0.26	Ethanamine, 2,2'-oxybis[N,N-dimethyl-	$C_8H_{20}N_2O$	160
79	35.378	0.25	0.42	Bicyclo[10.1.0]tridec-1-ene	$C_{13}H_{22}$	178
80	35.445	0.23	0.33	Cyclopentadecanone, 2-hydroxy-	$C_{15}H_{28}O_2$	240
81	35.807	0.75	0.88	Octadecane, 1-chloro-	$C_{18}H_{37}Cl$	288
82	35.944	1.47	1.51	Hexadecanoic acid, 2-hydroxy-1-(hydroxymethyl) ethyl ester	$C_{19}H_{38}O_4$	330
83	36.182	0.82	1.30	Docosanoic acid, methyl ester	$C_{23}H_{46}O_2$	354
84	36.516	0.15	0.13	Methyl (Z)-5,11,14,17-eicosatetraenoate	$C_{21}H_{34}O_2$	318
85	36.912	0.08	0.12	2-Oxabicyclo[3.3.0]oct-7-en-3-one, 7-(1-hydroxypentyl)-	$C_{12}H_{18}O_3$	210
86	37.056	0.28	0.26	2-(Dimethylamino) ethyl adamantanecarboxylate	$C_{15}H_{25}NO_2$	251
87	37.192	0.14	0.12	Cyclononasiloxane, octadecamethyl-	$C_{18}H_{54}O_9Si_9$	666
88	37.287	0.14	0.15	Docosahexaenoic acid, 1,2,3-propanetriyl ester	$C_{69}H_{98}O_6$	1022
89	37.509	0.28	0.53	Tricosanoic acid, methyl ester	$C_{24}H_{48}O_2$	368
90	37.918	0.17	0.20	Docosanoic acid, 2-hydroxy-, methyl ester	$C_{23}H_{46}O_3$	370
91	38.309	2.10	2.98	9,12-Octadecadienoic acid (Z,Z)-, 2,3-dihydroxypropyl ester	$C_{21}H_{38}O_4$	354
92	38.356	2.90	2.97	Oleic anhydride	$C_{36}H_{66}O_3$	546
93	38.657	0.66	0.78	Octadecanoic acid, 2,3-dihydroxypropyl ester	$C_{21}H_{42}O_4$	358
94	38.791	0.81	1.33	Tetracosanoic acid, methyl ester	$C_{25}H_{50}O_2$	382
95	39.467	0.26	0.12	7,10-Octadecadienoic acid, methyl ester	$C_{19}H_{34}O_2$	294
96	39.667	0.17	0.10	Dotriacontane	$C_{32}H_{66}$	450

97	39.863	0.86	1.24	2,6,10,14,18,22-Tetracosahexaene, 2,6,10,15,19,23-hexamethyl-, (all-E)-	$C_{30}H_{50}$	410
98	40.064	2.10	1.02	6,9,12,15-Docosatetraenoic acid, methyl ester	$C_{23}H_{38}O_2$	346
99	40.406	0.25	0.15	BISABOLEN-12-OL <BETA-> DB5-2242	$C_{15}H_{26}O$	222
100	40.502	0.39	0.52	Docosanoic acid, 2-hydroxy-, methyl ester	$C_{23}H_{46}O_3$	370

Table 2: Compounds detected in methanolic extract of *M. balsamina* (dried ripe fruits) through GC-MS/MS analysis

Peak No.	R. Time (minute)	Area (%)	Height (%)	Compound	Molecular formula	Molecular weight
1	3.752	0.46	0.44	3,3-Dimethoxy-2-butanone	$C_6H_{12}O_3$	132
2	3.908	0.29	0.24	1H-Pyrrole, 2,4-dimethyl-	C_6H_9N	95
3	5.000	0.09	0.11	Pyrrolidine	C_4H_9N	71
4	5.172	0.47	0.29	1,2-Cyclopentanedione	$C_5H_6O_2$	98
5	5.542	0.13	0.08	DL-Arabinitol	$C_5H_{12}O_5$	152
6	6.165	0.54	0.24	Glycerin	$C_3H_8O_3$	92
7	6.297	0.79	0.54	Phenol	C_6H_6O	94
8	8.333	0.08	0.06	2-Pyrrolidinone	C_4H_7NO	85
9	8.910	0.11	0.12	Phenol, 2-methoxy-	$C_7H_8O_2$	124
10	9.142	0.57	0.36	4-Pyridinol	C_5H_5NO	95
11	9.767	0.14	0.09	Hexyl n-valerate	$C_{11}H_{22}O_2$	186
12	9.967	0.09	0.09	Methylene asparagine	$C_5H_8N_2O_3$	144
13	10.306	0.41	0.52	4H-Pyran-4-one, 2,3-dihydro-3,5-dihydroxy-6-methyl-	$C_6H_8O_4$	144
14	10.481	0.24	0.33	Silane, diethoxymethyl-	$C_5H_{14}O_2Si$	134
15	10.692	0.11	0.11	Benzoic Acid	$C_7H_6O_2$	122
16	11.076	0.11	0.09	3-[N'-(3H-Indol-3-ylmethylene)-hydrazino]-5-methyl-[1,2,4]triazol-4-ylamine	$C_{12}H_{13}N$	255
17	11.308	0.16	0.17	(S)-(+)-2',3'-Dideoxyribonolactone	$C_5H_8O_3$	116
18	11.519	0.13	0.18	1,2-Benzenediol	$C_6H_6O_2$	110
19	12.048	0.14	0.17	3-Thiopheneethanol	C_6H_8OS	128
20	12.140	0.61	0.86	Benzofuran, 2,3-dihydro-	C_8H_8O	120
21	12.271	0.31	0.38	2-Furancarboxaldehyde, 5-(hydroxymethyl)-	$C_6H_6O_3$	126
22	12.682	0.10	0.10	1-Dimethyl(octyl)silyloxypropane	$C_{13}H_{30}OSi$	230
23	12.830	0.12	0.10	Silane, [3-(2,3 epoxypropoxy)propyl] ethoxydimethyl-	$C_{10}H_{22}O_3Si$	218
24	13.864	0.14	0.10	2-Decanynoic acid	$C_{10}H_{16}O_2$	168
25	14.163	0.18	0.21	Cyclohexasiloxane, dodecamethyl-	$C_{12}H_{36}O_6Si_6$	444
26	14.472	0.28	0.39	2-Methoxy-4-vinylphenol	$C_9H_{10}O_2$	150
27	15.862	0.55	0.82	DL-Proline, 5-oxo-, methyl ester	$C_6H_9NO_3$	143
28	16.484	0.14	0.21	3,8-Dioxa-2,9-disiladec-5-ene, 2,2,9,9-tetramethyl-, (E)-	$C_{10}H_{24}O_2Si_2$	232

29	17.233	0.19	0.14	Cyclooctanecarboxylic acid, 4,5-dimethyl-, methyl ester	$C_{12}H_{22}O_2$	198
30	17.342	0.16	0.23	2-Formyl-9-[.beta.-d-ribofuranosyl] hypoxanthine	$C_{11}H_{12}N_4O_6$	296
31	17.482	0.59	0.30	Xanthosine	$C_{10}H_{12}N_4O_6$	284
32	17.969	0.29	0.17	Cycloheptasiloxane, tetradecamethyl-	$C_{14}H_{42}O_7Si_7$	518
33	19.108	0.06	0.05	Quinoline, 5,6,7,8-tetrahydro-	$C_9H_{11}N$	133
34	20.013	0.09	0.14	3',5'-Dimethoxyacetophenone	$C_{10}H_{12}O_3$	180
35	20.508	0.23	0.13	6-Methyl-2-pyrazinylmethanol	$C_6H_8N_2O$	124
36	20.679	0.24	0.18	Ethyl N-(o-anisyl)formimidate	$C_{10}H_{13}NO_2$	179
37	21.200	0.61	0.32	Ethyl .alpha.-d-glucopyranoside	$C_8H_{16}O_6$	208
38	21.458	0.08	0.13	Cyclooctasiloxane, hexadecamethyl-	$C_{16}H_{48}O_8Si_8$	592
39	22.385	0.07	0.07	Trichloroacetic acid, undecyl ester	$C_{13}H_{23}Cl_3O_2$	316
40	22.825	0.11	0.19	E-10-Methyl-11-tetradecen-1-ol propionate	$C_{18}H_{34}O_2$	282
41	23.284	0.08	0.18	Methyl tetradecanoate	$C_{15}H_{30}O_2$	242
42	23.919	0.15	0.22	Tetradecanoic acid	$C_{14}H_{28}O_2$	228
43	23.992	0.12	0.14	5-Ethoxy-2-oxiran-2-yl-pyridine	$C_9H_{11}NO_2$	165
44	25.406	0.18	0.32	2-Dodecen-1-yl(-)succinic anhydride	$C_{16}H_{26}O_3$	266
45	26.959	2.25	4.53	Hexadecanoic acid, methyl ester	$C_{17}H_{34}O_2$	270
46	27.383	1.23	0.57	.alpha.-D-Glucopyranoside, .alpha.-D-glucopyranosyl	$C_{12}H_{22}O_{11}$	342
47	27.572	2.70	3.45	l-(+)-Ascorbic acid 2,6-dihexadecanoate	$C_{38}H_{68}O_8$	652
48	27.704	0.36	0.33	10-Octadecynoic acid, methyl ester	$C_{19}H_{34}O_2$	294
49	28.058	2.12	0.93	.beta.-D-Mannofuranoside, 1-O-(10-undecenyl)-	$C_{17}H_{32}O_6$	332
50	28.744	0.82	0.57	26,27-Dinorergost-5-ene-3,24-diol, (3.beta.)-	$C_{26}H_{44}O_2$	388
51	28.841	0.48	0.52	.alpha.-D-Glucopyranoside, .alpha.-D-glucopyranosyl	$C_{12}H_{22}O_{11}$	342
52	29.023	1.49	0.71	4,8,13-Cyclotetradecatriene-1,3-diol, 1,5,9-trimethyl-12-(1-methylethyl)-	$C_{20}H_{34}O_2$	306
53	29.235	1.12	0.60	Stigmasta-5,24(28)-dien-3-ol, (3.beta.)-	$C_{29}H_{48}O$	412
54	29.400	0.30	0.35	Cyclooctasiloxane, hexadecamethyl-	$C_{16}H_{48}O_8Si_8$	592
55	29.500	0.47	0.31	L-Lyxose	$C_5H_{10}O_5$	150
56	29.775	1.73	3.38	9,12-Octadecadienoic acid (Z,Z)-, methyl ester	$C_{19}H_{34}O_2$	294
57	29.879	3.91	6.63	11,14,17-Eicosatrienoic acid, methyl ester	$C_{21}H_{36}O_2$	320
58	30.014	0.73	0.78	Tricyclo[4.2.1.0(2,5)]non-3-en-9-endo-ol, 9-exo-ethyl-, endo-	$C_{11}H_{16}O$	164
59	30.301	1.92	3.00	Octadecanoic acid, methyl ester	$C_{19}H_{38}O_2$	298
60	30.488	4.15	3.72	cis,cis,cis-7,10,13-Hexadecatrienal	$C_{16}H_{26}O$	234
61	30.717	0.38	0.62	Cyclohexadecanone	$C_{16}H_{30}O$	238
62	30.784	0.46	0.69	Benzeneacetic acid, phenylmethyl ester	$C_{15}H_{14}O_2$	226
63	30.848	1.39	1.04	Octadecanoic acid	$C_{18}H_{36}O_2$	284
64	31.112	0.67	0.64	Hexadecanamide	$C_{16}H_{33}NO$	225

65	31.247	1.69	0.71	9,19-Cycloergost-24(28)-en-3-ol, 4,14-dimethyl-, acetate, (3.beta.,4. alpha.,5.alpha.)-	$C_{32}H_{52}O_2$	468
66	31.700	1.95	0.83	beta.-Sitosterol	$C_{29}H_{50}O$	414
67	31.908	0.36	0.42	Formamide, N-phenyl-	C_7H_7NO	121
68	31.989	0.99	1.38	Hexadecatrienoic acid, methyl ester	$C_{17}H_{28}O_2$	264
69	32.086	0.92	1.36	Hexadecatrienoic acid, methyl ester	$C_{17}H_{28}O_2$	264
70	32.304	0.68	1.08	Hexadecatrienoic acid, methyl ester	$C_{17}H_{28}O_2$	264
71	32.567	0.35	0.35	Hexadecatrienoic acid, methyl ester	$C_{17}H_{28}O_2$	264
72	32.698	1.82	2.72	1,9,12,15-Octadecatetraene, 1-methoxy-	$C_{19}H_{32}O$	276
73	32.886	0.93	1.46	Hexadecanoic acid, 2-hydroxy-1,3-propanediyl ester	$C_{35}H_{68}O_5$	568
74	32.983	0.28	0.36	Tetrapentacontane, 1,54-dibromo-	$C_{54}H_{108}Br_2$	914
75	33.092	0.09	0.14	Hexadecanoic acid, 2-propyl-, methyl ester	$C_{20}H_{40}O_2$	312
76	33.158	0.06	0.11	Palmidrol	$C_{18}H_{37}NO_2$	299
77	33.247	0.15	0.24	3-(2-Oxocyclohexyl)propionaldehyde	$C_9H_{14}O_2$	154
78	33.358	0.26	0.40	Eicosanoic acid, methyl ester	$C_{21}H_{42}O_2$	326
79	33.572	0.78	0.37	Naphthalene, decahydro-1,6-dimethyl-4-(1-methylethyl)-	$C_{15}H_{28}$	208
80	33.837	0.54	0.48	Methyl (Z)-5,11,14,17-eicosatetraenoate	$C_{21}H_{34}O_2$	318
81	34.189	0.46	0.18	9-Octadecenamide, (Z)-	$C_{18}H_{35}NO$	281
82	34.670	0.14	0.18	9,12,15-Octadecatrienoic acid, methyl ester, (Z,Z,Z)-	$C_{19}H_{32}O_2$	292
83	34.773	0.15	0.19	Nonanoic acid, 9-(3-hexenylidenecyclopropylidene)-, 2-hydroxy-1-(hydroxymethyl)ethyl	$C_{21}H_{36}O_4$	352
84	34.983	0.24	0.14	Vitamin A aldehyde	$C_{20}H_{28}O$	284
85	35.245	0.90	1.26	Carbamic acid, 2-(dimethylamino)ethyl ester	$C_5H_{12}N_2O_2$	132
86	35.375	1.09	1.68	9,12-Octadecadienoyl chloride, (Z,Z)-	$C_{18}H_{31}ClO$	298
87	35.455	3.54	2.72	1-Heptatriacotanol	$C_{37}H_{76}O$	536
88	35.796	0.67	1.08	1-Octanol, 2-butyl-	$C_{12}H_{26}O$	186
89	35.941	4.80	5.62	Hexadecanoic acid, 2-hydroxy-1-(hydroxymethyl)ethyl ester	$C_{19}H_{38}O_4$	330
90	36.176	0.37	0.48	Docosanoic acid, methyl ester	$C_{23}H_{46}O_2$	354
91	36.534	0.43	0.31	.beta.-Sitosterol	$C_{29}H_{50}O$	414
92	36.849	2.22	1.16	Olean-12-en-28-oic acid, 3-(acetyloxy)-, methyl ester, (3.beta.)-	$C_{33}H_{52}O_4$	512
93	36.967	0.48	0.84	Olean-12-en-28-oic acid, 3-oxo-, methyl ester	$C_{31}H_{48}O_3$	468
94	37.061	1.03	0.78	1,2,6a,6b,9,9,12a-Heptamethyl-1,3,4,5,6,6a,6b,7,8,8a,9,12,12a,12b,13,14b-hexadecahydro-2H	$C_{31}H_{48}O_2$	452
95	37.343	2.22	1.02	Androstan-3-one, 17-hydroxy-1,17-dimethyl-, (1.alpha.,5.alpha.,17.beta.)-	$C_{21}H_{34}O_2$	318
96	37.499	0.31	0.50	Tricosanoic acid, methyl ester	$C_{24}H_{48}O_2$	368

97	37.974	2.55	1.90	5,8,11,14-Eicosatetraenoic acid, ethyl ester, (all-Z)-	$C_{22}H_{36}O_2$	332
98	38.292	2.72	4.26	9,12-Octadecadienoic acid (Z,Z)-, 2,3-dihydroxypropyl ester	$C_{21}H_{38}O_4$	354
99	38.404	8.27	6.12	Methyl (Z)-5,11,14,17 eicosatetraenoate	$C_{21}H_{34}O_2$	318
100	38.654	3.69	4.46	Octadecanoic acid, 2,3 dihydroxypropyl ester	$C_{21}H_{42}O_4$	358
101	38.785	0.45	0.70	Tetracosanoic acid, methyl ester	$C_{25}H_{50}O_2$	382
102	38.867	0.61	0.50	Alloaromadendrene oxide-(1)	$C_{15}H_{24}O$	220
103	39.193	1.63	0.67	Rhodopin	$C_{40}H_{58}O$	554
104	39.496	1.42	0.74	9,19-Cyclolanostan-3-ol, acetate, (3.beta.)-	$C_{32}H_{54}O2$	470
105	39.667	0.15	0.21	Tetratetracontane	$C_{44}H_{90}$	618
106	39.855	1.24	1.62	2,6,10,14,18,22-Tetracosahexaene, 2,6,10,15,19,23-hexamethyl-, (all-E)-	$C_{30}H_{50}$	410
107	40.116	3.87	1.81	1H-Indene, 1-ethylideneoctahydro-7a-methyl-, cis-	$C_{12}H_{20}$	164
108	4.391	1.64	1.09	1R,3Z,9s-4,8,11,11-Tetramethylbicyclo[7.2.0]undeca-3,7-diene	$C_{15}H_{24}$	204
109	40.823	1.42	1.71	11,11-DIMETHYL-SPIRO[2,9] DODECA-3,7-DIEN	$C_{14}H_{22}$	190

Conclusion

The methanolic extract of *Momordica balsamina* fruits, analyzed via Gas Chromatography-Mass Spectrometry (GC-MS/MS), revealed a diverse range of bioactive phytochemicals with notable therapeutic potential. These compounds include alkaloids, phenols, flavonoids, saponins, tannins, terpenoids, cardiac glycosides, carbohydrates, and steroids. In total, 100 bioactive compounds were identified in the green fruits and 109 in the ripe fruits. The detected constituents exhibit a wide array of pharmacological properties, establishing *M. balsamina* as a promising candidate for the treatment of various human diseases. This comprehensive phytochemical analysis not only underscores the plant's medicinal value but also highlights its potential in pharmaceutical development. Additionally, the non-targeted profiling provides a foundation for future targeted studies of specific bioactive compounds in *M. balsamina*, allowing for a deeper investigation into its therapeutic applications.

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Conflict of Interest

The authors have no conflict of interest.

Data Sharing

All relevant data are within the manuscript.

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Research Article

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Effect of hand defoliation on advancing fruiting in sugar apple (*Annona squamosa* L.) cv. Balanagar

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ABSTRACT

Sugar apple cultivar 'Balanagar' comes to harvest during August-September under mild tropical climate, which coincides with monsoon rains resulting in poor fruit quality and high susceptibility to anthracnose and fruit fly. An attempt was made to advance the fruiting in this cultivar through hand defoliation in 2017-18 at ICAR-IIHR, Bengaluru. The effects of defoliation on flowering and fruiting were compared. Early sprouting, flowering and fruit harvest were recorded in the defoliated trees. Earliest fruits were harvested during 3rd week of April (216 days) from the defoliated trees while the un-defoliated trees come to harvest during the 1st week of August (313 days) (P=0.01). Fruits with higher pulp per cent (61.6) and lesser seeds (31) were harvested from the defoliated trees (P=0.05). Defoliation did not affect the fruit weight and acidity significantly. Comparatively, higher yield per tree and fruit TSS (7.2 kg; 24.7°B) were recorded for the un-defoliated trees over the defoliated trees (6.8 kg; 23.1°B) (P=0.05). Results revealed that fruiting could be advanced by three months to April from the normal season of August-September with comparable fruit quality by defoliating the trees during 3rd week of September.

Introduction

Sugar apple (*Annona squamosa* L.) is predominantly cultivated in the arid and semi-arid regions of India, notably in the states of Andhra Pradesh, Maharashtra, Karnataka, Bihar, Odisha, Assam, Tamil Nadu, and Rajasthan. The fruit is primarily consumed fresh, although products like custard powders and ice creams are also made from its pulp. It is a high-energy fruit, an excellent source of vitamin C and manganese, and is rich in thiamine, vitamin B6, iron, magnesium, phosphorus, and potassium in moderate amounts. Among the various sugar apple cultivars grown across India, such as Balanagar, Mammoth, Mahaboobnagar, Washington, Red Sitaphal, British Guinea, and Kakarlapahad, Balanagar is the most commonly cultivated. Under natural conditions, the fruits mature and are harvested during September-October, coinciding with the monsoon season. This often results in poor fruit quality and a high incidence of anthracnose disease and fruit fly infestations. As a deciduous fruit crop, sugar apple flowers on the current season's growth in March-April after leaf fall in the preceding winter. Advancing the fruiting season through crop regulation practices could enhance fruit quality during summer.

In sugar apple, flower buds are formed extra-axillary, opposite

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the leaves, and are limited to early shoot development (George and Nissen, 1987; Chander *et al.*, 2022). The cultivar Balanagar, with its short branches and sub-petiolar lateral buds, offers an opportunity to induce flowering through defoliation or bud break promotion, which could be more practical than pruning (Chander *et al.*, 2019). While various chemicals like urea, dormex, ethrel, potassium iodide (KI), and naphthalene acetic acid (NAA) have been used to defoliate trees effectively, the physical removal of leaves through hand defoliation is an alternative that avoids chemical applications. Several studies on different fruit crops, including guava (Sahay & Singh, 2001; Atawia *et al.*, 2017), lasora (Kumar *et al.*, 2018), peach (Lloyd and Firth, 1990), and vine (Sabbatini and Howell, 2010), have demonstrated the potential of hand defoliation to induce flowering. Watson and Casper (1984) and Watson (1986) noted that defoliation interrupts flowering responses by altering the partitioning of assimilates within the tree.

Defoliation of leaves in lasura in the month of December-January produces early flowering and fruiting in arid region (Sharma *et al.*, 2013). The competition for assimilates depends on branch autonomy, balancing reproductive and vegetative growth. In sugar apple, axillary buds are concealed at the leaf petiole base, which acts as a physical barrier to sprouting. Manipulating the timing of defoliation could thus influence flowering and fruiting periods (Chander *et al.*, 2022). Given the limited information on crop regulation in sugar apple, this study aims to explore the potential of hand defoliation as a sustainable, small-scale method to induce early flowering and fruiting and enhance the fruit's marketability. Although challenging and not economical on a large scale, this approach offers valuable insights into targeted and environmentally friendly crop management practices.

Material and Methods

The investigation was carried out on 8-year-old healthy trees of sugar apple cv. Balanagar maintained at orchard of ICAR-IIHR, Bengaluru (India) during 2017-18. Thirty-two uniform and healthy trees were selected for the present study. Hand defoliation (T_1) (Picture 1) was undertaken during third week of September while control trees kept un-defoliated (T_2). Standard package of practices were adopted for maintenance of trees during the experimentation. The number of days required for sprouting and flowering was assessed by recording the days taken for the emergence of first sprout and flower respectively after the treatment imposition. The duration of the harvest was calculated from the date of imposing the treatments to the first fruit harvest. The total fruit yield per tree was recorded at harvest by measuring weight of fruit harvested and values were expressed in kilogram. Fruit weight (g) and peel weight (g) was recorded using electronic balance. The number of seeds per fruit was calculated by counting the number of seeds. The total soluble solids (TSS) were measured using digital refractometer and expressed as degree Brix. The titrable acidity was estimated by adopting the titrimetric method of AOAC (1975) using phenolphthalein indicators and values were expressed in terms of percentage tartaric acid equivalent. Pulp per cent was calculated by using the following formula: Pulp (%) = (Pulp weight / Fruit weight) × 100.

The experiment was laid out in a randomized block design (RBD) with two treatments (T_1 and T_2), each comprising 16 trees. Statistical analysis was performed using the t-test at a significance level of P = 0.05 as well as P = 0.01 to compare the means of the treatments. The critical difference (CD) at 5% & 1% was calculated to determine significant differences between the treatments. Data analysis was conducted using WASP (2.0) software (Jangam and Thali, 2004).

Results and Discussion

Sprouting and flowering

Hand defoliation significantly influenced the timing of sprouting and flowering in sugar apple. Early sprouting was observed in defoliated trees compared to the control. This early sprouting led to the initiation of flowering earlier than usual, with flowers emerging approximately 216 days after defoliation. In contrast, flowering in the un-defoliated trees began much later, with fruit development taking 313 days. The advancement of sprouting and flowering in defoliated trees could be attributed to the removal of physical and hormonal barriers posed by the leaf petioles (Chander *et al.*, 2019). This manipulation allowed better exposure of the dormant buds, stimulating early bud break. Manual defoliation resulted in the earliest sprouting as well as flowering in lasora (Kumar *et al.*, 2018). Similar findings have been reported in other fruit crops, where defoliation induced early flowering by redistributing assimilates and activating dormant meristems (Nanra *et al.*, 2001; Olesen and Muldoon, 2012; Sharma *et al.*, 2013; Boora *et al.*, 2016). Advancing the flowering period can provide significant advantages, as it shifts the fruiting period to a more favourable season, reducing susceptibility to monsoon-related fruit quality issues such as anthracnose and fruit fly infestation.

Fruit weight

The average fruit weight was marginally lower in the defoliated trees (237.7 g) compared to the control (246.1 g). However, this difference was statistically non-significant, indicating that hand defoliation did not have a measurable impact on fruit size. However, Sahay *et al.* (2001) found that application of urea at 15% + hand de-blossoming in guava,

and Chander *et al*. (2019) in Annona cv. Balanagar (urea @ 15%) increased the fruit size over control. The present study achieved an earlier harvest of fruits which was an notable advancement, nevertheless the fruit size remained non-significant which may be due to the weather conditions particularly lower temperature during the growth period of fruit.

Peel weight

Peel weight was significantly lower in the defoliated trees (79.7 g) compared to the control (84.4 g) (P=0.05). This reduction could be attributed to the early development of fruits under defoliated conditions, which potentially influenced fruit composition and peel thickness. Lower peel weight was advantageous as it increases the proportion of edible pulp, improving overall fruit quality (Chander *et al.*, 2019).

Pulp content (%)

Hand defoliation significantly increased pulp content to 61.6%, compared to 59.9% in the control (P=0.05). The higher pulp percentage in defoliated trees could be due to better resource allocation during fruit development under induced flowering conditions. This improvement in edible yield is a critical quality parameter for commercial production, as higher pulp content enhances consumer preference. Chander *et al.* (2019) stated that lower peel weight was advantageous as it increases the proportion of edible pulp, improving overall fruit quality of sugar apple cv. Balanagar.

Number of seeds

Defoliated trees produced fruits with significantly fewer seeds (31.3) compared to the control (41.8) (P=0.05). Reduced seed numbers are desirable in fruits, as it enhances the fruit's marketability and consumer acceptance. The reduction may result from altered assimilate partitioning caused by defoliation, as well as prevailing weather conditions during fruit set and development period which prioritizes pulp development over seed formation. Similar, result was observed by Gonzalez *et al.* (2013) in defoliated cherimoya and Chander *et al*. (2019) in sugar apple cv. Balanagar.

Fruit yield per tree

The yield per tree was slightly lower in the defoliated trees (6.8 kg) compared to the control (7.2 kg) (P=0.05). While the reduction in yield may be attributed to the physiological stress caused by defoliation, it is noteworthy that the quality parameters of the fruits from defoliated trees were superior. This trade-off between yield and quality warrants further investigation to optimize defoliation practices. The findings of Khan *et al.* (2013) in guava and Chander *et al.* (2019) in sugar apple, where no significant effect of defoliation treatments was seen on fruit yield.

Total soluble solids (TSS) and acidity

The TSS of fruits from defoliated trees was slightly lower (23.1°B) than that of the control (24.7°B) (P=0.05). Despite this reduction, the TSS remained within acceptable ranges for high-quality sugar apple fruits. Acidity levels were not significantly affected by defoliation, suggesting that the manipulation did not compromise the fruit's flavour profile. Similar result was reported by Chander *et al*. (2019) in sugar apple cv. Balanagar.

Harvest time

Defoliation advanced fruit harvest by approximately three months, with fruits from defoliated trees maturing in the third week of April (216 days), compared to the first week of August (313 days) for the control (P=0.01) (Picture 2a and 2b). This advancement aligns with the goal of shifting fruiting to a period with reduced rainfall, thereby improving fruit quality by minimizing the incidence of anthracnose and fruit fly infestation. Similar results were recorded in guava (Amador *et al.*, 1992; Khan *et al.*, 2013), lasora (Sharma *et al.*, 2013; Kumar *et al.*, 2018) and annona (Gonzalez *et al.*, 2013; Chander *et al.*, 2019).

Table 1. Effect of defoliation on fruit and yield characteristics

Treatments	Fruit weight (g)	Peel weight (g)	Pulp content (%)	Number of seeds	Fruit yield/ tree (kg)	TSS (°B)	Acidity (%)
Hand defoliation (T_1)	237.7	79.7	61.6	31.3	6.8	23.1	0.18
Control (Un-defoliated) T_2	246.1	84.4	59.9	41.8	7.2	24.7	0.19
CD @ 5%	-	2.0	2.0	2.1	2.0	2.1	-
T-test (P=0.05)	NS	S	S	S	S	S	NS

NS=Non-significant; S=Significant

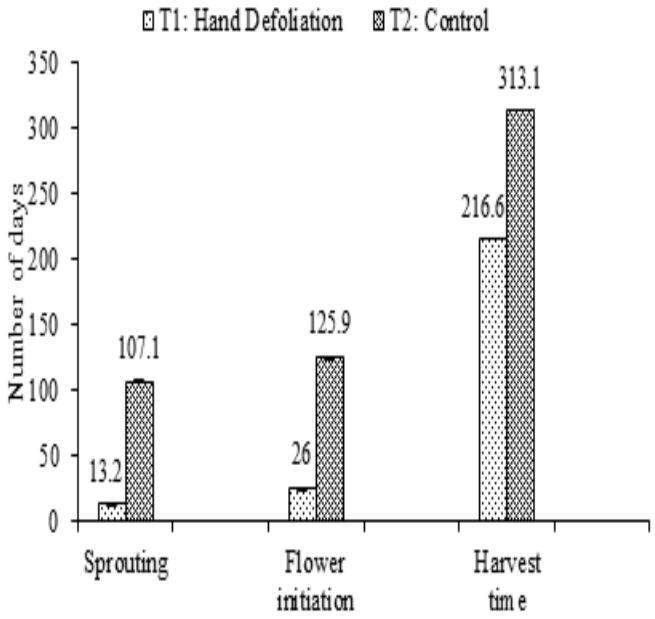

Figure 1. Effect of defoliation on sprouting, flower initiation and harvest time

Picture 1. Defoliated tree

Picture 2a. Harvested fruits (April, 2018)

Picture 2b. Harvested fruits (August, 2018)

Conclusion

The study demonstrated that hand defoliation during the third week of September effectively advanced the harvest season of sugar apple cultivar 'Balanagar' by three months, yielding fruits of comparable or improved quality. Although yield and TSS were slightly lower in defoliated trees, the higher pulp percentage, fewer seeds, and reduced peel weight highlight the potential of this practice for enhancing fruit quality of sugar apple cv. Balanagar. Further studies are recommended to refine defoliation techniques and evaluate their long-term impacts on productivity and profitability.

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Conflict of Interest

The authors have no conflict of interest.

Data Sharing

All relevant data are within the manuscript.

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Research Article

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Influence of organic and inorganic source of nutrients on physico-chemical attributes of fig (*Ficus carica* L.) cv. Dinkar

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ABSTRACT

A field experiment was conducted during 2023-24 to evaluate the impact of organic and inorganic nutrients on the physico-chemical attributes of fig cv. Dinkar. The highest fruit size, including length (5.31 cm), width (5.42 cm), fresh fruit weight (44.38 g), dry fruit weight (21.48 g), fruit volume (45.70 cc), and specific gravity (0.970), were observed in plants applied with 75% NPK + 25% Poultry manure. Additionally, the highest values for TSS (18.86° Brix), reducing sugars (17.60%), non-reducing sugars (1.29%), total sugar (18.90%), and TSS/Acid ratio (93.21) were also recorded with 75% NPK + 25% Poultry manure. The lowest acidity (0.24%) was found in plants treated with 75% NPK + 25% Poultry manure, while the highest acidity (0.24%) was noted in those treated with 75% NPK + 25% Vermicompost. The highest ascorbic acid content (13.30 mg/100g pulp) was also observed in the 75% NPK + 25% Poultry manure treatment.

Introduction

The fig (*Ficus carica* L.) belongs to the family Moraceae. It is among the earliest cultivated fruit trees in the world (Solomon *et al*., 2006). Although the fig tree is native to central Asia, it has spread throughout the Mediterranean region where it is well-adapted to several types of soils and climates due to its tolerance to salinity and drought. Consequently, figs are grown in many parts of the world where the climate is moderate (Crisosto *et al*., 2011). The common fig is a gynodioecious plant species with two different genders: female trees that produce syconia with female flowers that will develop into edible seeded figs (syconium with multiple one-seed fruits or drupelets) and caprifigs that produce syconia with male and female flowers that present a style shorter than the fruit of female trees. Pollen is only produced by caprifigs, so the reproductive system is functionally dioecious (Kjellberg *et al*., 1987). Three edible types of female figs are grown commercially viz., the common fig type that develops fruit parthenocarpically, the Smyrna type that needs pollination from caprifigs (caprification) to develop fruit, and the San Pedro type that produces a first crop (breba) parthenocarpically and a second or main crop (fig) only after caprification. Common-type figs can produce one (unifera types) or two crops (bifera types) (Fleishman *et al*., 2008).

Fig is an important crop worldwide for dry and fresh consumption. A per the Dietary Reference Intakes (DRI) data (Goswami *et al*., 2015) and the nutrient composition of dried

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figs (Hazarika *et al*., 2019), fig is a superior source of minerals and vitamins, providing iron (30%), calcium (15.8%), potassium (14%) thiamin (vitamin B_1) (7.1%), riboflavin (vitamin B_2) (6.2%) and ascorbic acid (15.65 mg/100 g fruit pulp). Figs are sodium free as well as fat and cholesterol free (Hazarika *et al*., 2019 and Kjellberg *et al*., 1987). Fig fruits contain at least 17 types of amino acids, among which aspartic acid and glutamine are the highest ones (Kjellberg *et al*., 1987). The dried figs also contain relatively high amounts of crude fibers (5.8%, w/w) which is higher than all other common fruits (Hazarika *et al*., 2019). More than 28% of the fiber is of the soluble type, which has been shown to aid in the control of blood sugar and blood cholesterol and in weight loss. Dried figs also contain one of the highest concentrations of polyphenols among the commonly consumed fruits and beverages (Hazarika *et al*., 2019 and Kumar *et al*., 1998). Keeping in view, the importance of fig in human diet, an experiment was conducted on effect of organic and inorganic source of nutrients on physico-chemical attributes of fig cv. Dinkar.

Material and Methods

A field experiment was conducted during 2023-24 at the Fruit Orchard, College of Horticulture, Banda University of Agriculture and Technology, Banda. The experiment was laid out in Randomized Block Design with three replications. The treatments comprised of Control (T_0), 100% NPK (T_1), RDF, 75%NPK + 25% Poultry manures (T_2), 75%NPK + 25% Vermicompost (T_3), 75%NPK + 25% Mushroom Waste (T_4), 50%NPK + 50% Poultry manures (T_5), 50%NPK + 50%Vermicompost (T_6), 50%NPK + 50% Mushroom Waste (T_7), 25%NPK + 75% Poultry manures (T_8), 25%NPK + 75% Vermicompost (T_9) and 25%NPK + 75% Mushroom Waste (T_{10}). Five fruits were randomly harvested from each treatment having uniform shape and size. Six morphological or physical characters and seven chemical attributes of fig fruits were studied during the study.

Results and Discussion

The physical characters of fig viz., fruit length, fruit width, fruit weight and fruit volume were found to be significantly influenced with organic and inorganic sources of nutrients (Table 1). The fruit length ranged from 4.37 to 5.31 cm. Maximum fruit length (5.31cm) was observed in the plants treated with 75%NPK + 25% Vermicompost, followed by (5.27 cm) treatment 75%NPK + 25% Mushroom waste. The maximum fruit width (5.42 cm) was observed in the plants treated with 75%NPK + 25% Vermicompost, followed by (5.37 cm) treatment 75%NPK + 25% Mushroom waste. The minimum fruit width (4.81 cm) was recorded in control. It is evident from the result the treatment 75%NPK + 25% Vermicompost had significant effect on fresh fruit weight (44.38 g), which was statistically at par with 75%NPK + 25% Mushroom waste (43.32 g) and 75%NPK + 25% Poultry manure (42.38 g). The dry weight of fruit was found highest (21.48 g) in treatment 75%NPK + 25% Vermicompost which was statistically at par with treatment 75%NPK + 25% Mushroom waste, (20.32 g) and treatment 75%NPK + 25% Poultry manure (19.99 g). While the lowest dry weight of fruit (14.21 g) was recorded in control. The result indicated that the treatment 75%NPK + 25% Vermicompost had the maximum fruit volume 45.70 cc which was significantly superior over all other treatment, followed by treatment 75%NPK + 25% Mushroom waste (44.58 cc) and treatment 75%NPK + 25% Poultry manure (43.90 cc) respectively. The minimum fruit volume (30.09 cc) was found with treatment T_0. A similar report has also been provided by Ratna *et al.* (2011 & 2019) in guava, Sharma *et al.* (2013) in guava, and Singh *et al.* (2017) in strawberry.

Table 1. Influence of organic and inorganic source of nutrients on physical fruit traits of fig

	Treatments	Fruit length (cm)	Fruit width (cm)	Fresh fruit weight (g)	Dry fruit weight (g)	Fruit volume (cc)	Fruit specific gravity
T_0	Control	4.37	4.81	29.72	14.21	31.09	0.950
T_1	100% NPK (RDF)	5.10	5.08	36.94	17.56	38.33	0.961
T_2	75%NPK + 25% Poultry manures	5.19	5.31	42.64	19.99	43.90	0.963
T_3	75%NPK + 25% Vermicompost	5.31	5.42	44.38	21.48	45.70	0.970
T_4	75%NPK + 25% Mushroom waste	5.27	5.37	43.32	20.32	44.58	0.967
T_5	50%NPK + 50% Poultry manures	5.12	5.28	38.89	17.79	40.29	0.963

T_6	50%NPK + 50%Vermicompost	5.15	5.14	39.12	18.46	40.52	0.963
T_7	50%NPK + 50% Mushroom waste	4.84	5.21	34.93	16.36	36.30	0.960
T_8	25%NPK + 75% Poultry manures	4.61	4.99	33.57	15.21	34.66	0.957
T_9	25%NPK + 75% Vermicompost	4.63	4.91	33.57	15.62	34.84	0.960
T_{10}	25%NPK + 75% Mushroom waste	4.47	4.85	33.17	14.66	34.38	0.957
	SEm±	0.04	0.01	1.07	0.69	1.057	0.003
	CD at 5%	0.12	0.03	3.20	2.05	3.141	NS

The fruit quality parameters of fig were found to be significantly affected by the application of inorganic nutrients and organic manures (Table 2). The maximum TSS (18.86° Brix) was recorded with the treatment T_3 followed by T_2 which had TSS of 18.63° Brix. The study affirms with the studies conducted by Gawande *et al.* (1998), Majunnatha *et al.* (2006) and Pereira and Mitra (1999) in guava. The response to treatment T_3 gave the highest titratable acidity (0.24%) and was statistically at par with T_4 and T_2, *i.e.,* 0.23, 0.23% respectively. However, the lowest titrable acidity (0.18%) was recorded in T_0. The similar results were observed by Kurubar *et al.* (2017) and Sharma *et al.* (2013) in fig and Ennab (2016) in Eureka Lemon Trees (*Citrus limon* L.).

Among all the treatment, significantly maximum TSS/ acid ratio was observed in treatment T_9 (93.21) followed by treatment T_0 (91.37). While, minimum TSS/acid ratio was recorded in treatment T_3 (78.59). This study is supported by the findings Singh and Banik (2011), Hazarika *et al.* (2019) in mandarin and Kurubar *et al.* (2017) in fig. The maximum percentage of total sugar in fig fruit pulp was found with treatment T_3 (18.90%) followed by T_4 (18.78%).

The maximum percentage of reducing sugar (17.60%) was found with 75%NPK + 25% Vermicompost (T_3) While, it was minimum T_0 (15.20%). Application of 75%NPK + 25% Vermicompost (T_3) resulted in minimum percentage of non-reducing sugar (1.29%) whereas, it was maximum in T_1 (2.41%) followed by T_0 (2.12%). The similar results were also reported by Kurubar *et al.* (2017) in fig cv. Poona Fig. Kumar *et al.* (1998), Shukla *et al.* (2014) and Sharma *et al.* (2013) in guava also confirmed the present findings. The treatment 75%NPK + 25% Vermicompost (T_3) resulted in significantly higher ascorbic acid content (13.30 mg/100g pulp) over all other treatments. These results are in conformity to the findings reported by Yadav *et al.* (2011), Shukla *et al.* (2014) and Goswami *et al.* (2015).

Table 2. Influence of organic and inorganic source of nutrients on fruit quality parameters of fig

	Treatments	TSS (°Brix)	Titratable acidity (%)	TSS/ Acid ratio	Total sugars (%)	Reducing sugar (%)	Non-reducing sugar (%)	Ascorbic acid (mg/100g fruit pulp)
T_0	Control	16.45	0.18	91.37	17.32	15.20	2.12	5.26
T_1	100% NPK (RDF)	18.17	0.22	81.41	18.38	15.98	2.41	9.17
T_2	75%NPK + 25% Poultry manures	18.63	0.23	81.00	18.70	17.18	1.52	12.60
T_3	75%NPK + 25% Vermicompost	18.86	0.24	78.59	18.90	17.60	1.29	13.30
T_4	75%NPK + 25% Mushroom waste	18.48	0.23	79.24	18.78	17.33	1.45	11.79
T_5	50%NPK + 50% Poultry manures	18.29	0.21	85.77	18.59	16.94	1.64	10.11
T_6	50%NPK + 50%Vermicompost	18.41	0.22	84.99	18.69	17.15	1.54	10.96

T_7	50%NPK + 50% Mushroom waste	18.16	0.21	85.15	18.31	16.51	1.80	8.39
T_8	25%NPK + 75% Poultry manures	17.60	0.20	86.76	17.51	15.66	1.85	6.59
T_9	25%NPK + 75% Vermicompost	17.71	0.19	93.21	17.70	15.71	1.99	7.52
T_{10}	25%NPK + 75% Mushroom waste	16.96	0.19	87.66	17.36	15.34	2.02	5.26
	SEm±	0.089	0.004	1.692	0.046	0.063	0.051	0.23
	CD at 5%	0.265	0.012	5.026	0.136	0.188	0.151	0.69

Conclusion

From the results, it is concluded that the application of inorganic nutrients combined with organic manures. 75%NPK + 25% Vermicompost (T_3) resulted in the highest fruit length, width, weight, volume, and dry weight, along with superior fruit quality attributes, including TSS, titratable acidity, total sugars, and ascorbic acid content. Among all treatments, 75%NPK + 25% Vermicompost (T_3) showed the most consistent and positive effects on the physical and biochemical properties of fig, demonstrating its potential for enhancing fruit quality in fig cultivation.

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Conflict of Interest

The authors have no conflict of interest.

Data Sharing

All relevant data are within the manuscript.

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Research Article

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Performance of the bottle gourd variety Thar Avani for morphological traits under rainfed semi-arid conditions

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ABSTRACT

Among 29 physiological traits in bottle gourd varieties, highly significant correlation in FL and FW with FG and NFMF, respectively (r=+1.00; P≤0.05) were observed. Likewise, FW with FL (r=+1.00; P≤0.001). High magnitude and significant correlation were found between IL and LPL (r=+1.00; P≤0.01). Similarly, high magnitude and significant correlation links were noticed in NFFF (r=+1.00; P≤0.01), DFFA (r=+1.00; P≤0.05) and NMFP (r=+1.00; P≤0.01) with LW; DFFA with NFFF(r=+1.00; P≤0.05); NMFP with NFFF (r=+1.00; P≤0.05); NFP with NFFP with NFFF (r=+1.00; P≤0.01); SYH with SYP with NFFF (r=+1.00; P≤0.001); FYP with SYP (r=+0.99; P≤0.01), SYH (r=+0.99; P≤0.01) and PL (r=+1.00; P≤0.01); FYH with SYP (r=+0.99; P≤0.01), SYH (r=+0.99; P≤0.01), PL (r=+1.00; P≤0.01) and FYP (r=+1.00; P≤0.001); RT with NSF (r=+1.00; P≤0.05); AAS with NSF (r=+1.00; P≤0.05) and RT (r=+1.00; P≤0.001); SYF with NSF (r=+1.00; P≤0.05). The findings indicate that Thar Avani demonstrates strong performance under semi-arid conditions, showcasing substantial variability that can be leveraged in breeding programs focused on yield improvement.

Introduction

Bottle gourd [*Lagenaria siceraria* (Mol.) Standl.], also known as Calabash, white flower gourd, or locally as *ghia* or *lauki*, is one of the most significant cucurbitaceous vegetables cultivated in tropical and subtropical regions worldwide (Yadav *et al.*, 2023). The chromosome number of bottle gourd is 2n = 22 (Reddy *et al.*, 2017). The fruit shape of bottle gourd can vary from flat to round, oval, oblong, and long. The name "bottle gourd" is believed to have originated from the mature, harvested fruits, which were traditionally used as bottles, utensils, or pipes (Yadav *et al.*, 2023). The term "Calabaza" is thought to have come from the Arabic *qar'a yabisa* (meaning "dry gourd"), Persian *kharabuz* (used for various large melons), or from pre-Roman Iberian *calapaccia* (Basu *et al.*, 2017). In India, bottle gourd is known by several names, including Lauki, Ghia, Doodhi, Jatilao, Sorakaya, and Bhopla, among others. Historical Sanskrit texts also reference bottle gourd as *alabu*, and it was used to make musical instruments such as the *tambura*, *veena*, and *kamandalu* (Yadav *et al.*, 2023).

Bottle gourd is primarily grown for its tender leaves and young fruit, which are consumed as vegetables, usually cooked. The mature fruit's hard shell is used to create a variety of domestic items, including musical instruments, jugs, utensils for storing liquids and food, and floats for fishing nets (Chadha,

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2001). The fruit colour can range from green to cream or yellow, with the flesh typically being white. Uniquely, bottle gourd undergoes anthesis in the evening, and the flowers are white, earning it the moniker "white flowered gourd". Its fruit possesses medicinal properties and is traditionally used as a cardiotonic, aphrodisiac, hepatoprotective, analgesic, anti-inflammatory, expectorant, diuretic, and antioxidant.

Considered one of the oldest crops in the tropics, bottle gourd is believed to be indigenous to Africa and has a wide adaptability to arid and semi-arid regions. In India, significant cultivation areas include Uttar Pradesh, Rajasthan, Haryana, Punjab, Delhi, Andhra Pradesh, Bihar, and Karnataka (Yadav *et al.*, 2023). Local varieties are often grown in backyard gardens or kitchen gardens, particularly in semi-arid tribal regions, where they exhibit significant morphological diversity.

Considering its broad potential and importance, efforts to improve bottle gourd were initiated at the ICAR - Central Horticultural Experiment Station, Godhra, Gujarat, through hybridization, followed by selection from the segregating population of promising lines LS-4 x LS3-2. This work led to the development of the variety Thar Avani, which was advanced to the F_8 generation between 2016 and 2020 and identified at the institute level in 2021. Thar Avani is a highly vigorous variety with dense foliage. Male and female flowers appear from the 7th and 11th nodes, respectively. Each plant produces 24-32 female flowers and sets harvestable-sized fruits between 57 and 62 days after sowing. The fruit is round in shape, with an average length of 22.8 cm, a girth of 39.4 cm, and weighs between 750-860 g. Additionally, the variety shows a TSS of 8.1-8.7° Brix, a content of 21.6 mg/100g AAF, and an average fruit weight of 12.91 kg per plant.

Material and Methods

The comparative evaluation of bottle gourd variety Thar Avani along with Pusa Sandesh and Arka Bahar was carried out for different traits. The experiment was laid out during rainy season of 2021 and 2022 in randomized block design with eight replications under rainfed semi-arid condition at Vegetable Experiment Block, ICAR- Central Horticultural Experiment Station, Godhra, Gujarat situated at latitude 22°41"38" N, longitude 73°33′ 38" E and elevation of 113-115 m above mean sea level.

The cultural practices and production technology were followed according to (Yadav *et al.*, 2023). The mean maximum and minimum temperature varied between 28.5 to 47.1°C and 12.6 to 26.8°C, respectively, and total annual minimum and maximum rainfall of ranged from 462.46 mm to 764.82 mm with relative humidity 27.55-92.50 per cent during the period under study. The crop was raised during rainy season of 1st week of July, 2021 and 2022.

The physiological traits of bottle gourd varieties were recorded by following standard methods (Yadav *et al.*, 2022; Yadav *et al.*, 2019) and morphological traits according to (Biodiversity International for Cucurbitaceae, 2007). Ascorbic acid content was determined in accordance with the dinitrophenylhydrazine (DNPH) method. The value was expressed as mg/100 g FW (Yadav *et al.*, 2019).

The best linear unbiased predictor (BLUP) was calculated to eliminate the different environmental effect, using the Meta-R program V.6.0 (Alvarado *et al.*, 2020). Variability package in the R program (Team R.C.R., 2017) was used for the analysis of variance (ANOVA), where varieties and environments were treated as fixed and random effects, respectively. Statistical significance was determined at $P < 0.001$. Descriptive statistics including mean, range, and standard error, were estimated using the same package of the R program (Alvarado *et al.*, 2020). Additionally, the biplot PCA graphs were generated by aggregating mean data of all traits with the assistance of *FactoMineR* and *factoextra* packages in R program (Team R.C.R., 2017).

Results and Discussion

Among 29 physiological traits in bottle gourd genotypes, highly significant correlation in FL and FW with FG and NFMF, respectively (r=+1.00; P≤0.05) were observed. Likewise, FW with FL (r=+1.00; P≤0.001). High magnitude and significant correlation was found between IL and LPL (r=+1.00; P≤0.01). Similarly, High magnitude and significant correlation links were noticed in NFFF (r=+1.00; P≤0.01), DFFA (r=+1.00; P≤0.05) and NMFP (r=+1.00; P≤0.01) with LW; DFFA with NFFF(r=+1.00; P≤0.05); NMFP with NFFF (r=+1.00; P≤0.05); NFP with NFFP with NFFF(r=+1.00; P≤0.01); SYH with SYP with NFFF(r=+1.00; P≤0.001); FYP with SYP (r=+0.99; P≤0.01), SYH (r=+0.99; P≤0.01) and PL (r=+1.00; P≤0.01); FYH with SYP (r=+0.99; P≤0.01), SYH (r=+0.99; P≤0.01), PL (r=+1.00; P≤0.01) and FYP (r=+1.00; P≤0.001); RT with NSF (r=+1.00; P≤0.05); AAS with NSF (r=+1.00; P≤0.05) and RT (r=+1.00; P≤0.001); SYF with NSF (r=+1.00; P≤0.05).

High magnitude and non-significant correlation were found between NFMF with FG (r=+0.97) and NFMF (r=+0.99); FL and FW with LL (r=+0.96); LW, NFFF, DFFA, NMFP with LL (r=+0.95; r=+0.95; r=+0.93 and r=+0.92, respectively); SYP and SYH with NFFP and NFP (r=+0.95); PL with SYP and SYH (r=+0.96); FYP and FYH with NFFP, NFP (r=+0.94) and SYP and SYH (r=+0.99), respectively; DMFA with FYP and FYH (r=+0.90); PtL and SKW with DMFA (r=+0.90); NSF with SKW (r=+0.92) and FT (r=+0.97); RT with SKW (r=+0.86), FT (r=+0.90) and NSF (r=+0.96); AAF with SKW (r=+0.90) and FT (r=+0.96); SYF with DMFA (r=+0.84), PL (r=+0.91), SKW (r=+0.94) and FT (r=+0.95); SW with DMFA (r=+0.87), PL (r=+0.93), SKW (r=+0.96), FT (r=+0.93), NSF (r=+0.99), RT (r=+0.99) and AAF (r=+0.99); AATL with DMFA (r=+0.84), PL (r=+0.94), SKW (r=+0.96), FT (r=+0.93), NSF (r=+0.99), RT (r=+0.98) and AAF (r=+0.98).

Subsequently, NFFP and NFP conveyed strong negative connections with LL (r=-1.0; P≤0.05). Likewise, SYP and SYH with LW (r=-1.0; P≤0.01), NFFF (r=-1.0; P≤0.001), DFFA (r=-1.0; P≤0.05) and NMFP (r=-1.0; NS), respectively; PL with PdL (r=-1.0; P≤0.05); FYP and FYH with DFFA (r=-1.0; P≤0.05) and NMFP (r=-1.0; P≤0.01), respectively; DMFA with LPL (r=-1.0; P≤0.01).

Besides this, LPL, IL, PdL, LW, NFFF, DFFA and NMFP are found negatively correlated with SKW, PtL, DMFA, FYH, FYP, PL, SYH, SYP, NFP and NFFP, which are not useful (LPL, IL, PdL, LW, NFFF, DFFA and NMFP) as they governed by non-additive gene action. Among all 29 traits of bottle gourd varieties in combined form, FYP and S were highly and significantly correlated with FYH (r=+1.00; P≤0.001). Similarly, S with FYP and S (r=+1.00; P≤0.001); SYH and SYP with S (r=+1.00; P≤0.001); SYP with SYH (r=+1.00; P≤0.05); NFFP with NFP (r=+1.00; P≤0.05); TP with NFP and NFFP (r=+1.00; P≤0.001); SW with AATL (r=+1.00; P≤0.01); SYF with AATL and SW (r=+1.00; P≤0.05); NSF with SYF (r=+1.00; P≤0.05); AAF and RT with NSF and ViC (r=+1.00; P≤0.05); RT with AAF (r=+1.00; P≤0.001); Fe, Ca and FRAP with PL (r=+1.00; P≤0.05); SKW, Ca with Fe (r=+1.00; P≤0.05); FRAP with Ca (r=+1.00; P≤0.05); K with FRAP and DMFA (r=+1.00; P≤0.05); LW, Mn and DFFA with NFFF (r=+1.00; P≤0.01 and P≤0.05); Mn and DFFA with LW (r=+1.00; P≤0.05); DFFA with NMFP (r=+1.00; P≤0.05); FL with FW (r=+1.00; P≤0.01); NFMF and FG with FW and FL (r=+1.00; P≤0.05); Mg with LL (r=+1.00; P≤0.01). In this context, same link was observed non-significant correlation among the different traits in combination form viz. SYH and SYP with FYH and FYP (r=+0.99).

Similarly in PL, NFP, NFFP and TP with FYH, FYP, S, SYH, SYP and PL; P, TF and AATL with DPPH, CUPRAC and P. the same link was also noticed in NSF, AAF, Rt, FT, PtL, Fe, SKW, Cu, Ca, FRAP, DMFA and K with AATL, SW, SYF, NSF, AAF, RT, Ft, PtL, and SKW, FW and FG also indicated positive non-significant correlation with NFFF, LW, Mn and DFFA. The symmetrical links were also noticed in LL and Mg with FW, FL, NFMF and FG. The remaining traits showed negatively non-significant results. Simultaneously, NFFF and LW mapped strong negative connections with SYH and SYP (r=-1.0; P≤0.01); S (r=-1.0; P≤0.05). The traits indicated similar finding like Mn with SYH and SYP (r=-1.0; P≤0.05). The NMFP with FYH and FYP (r=-1.0; P≤0.01); DFFA with FYH, FYP, SYH and SYP (r=-1.0; P≤0.05); PdL with PL (r=-1.0; P≤0.05); LL with NFP (r=-1.0; P≤0.05) and TP (r=-1.0; P≤0.01); Mg with TP (r=-1.0; P≤0.05). Rest of the traits showed negatively non-significant results. These results are in support with the findings of Bharathi et al. (2014) in M. dioca and (Yadav et al. (2023), Sirohi et al. (1988) and Samadia (2002) in bottle gourd.

High correlation between core collection (CC) entire population of the bottle gourd germplasm (r= 0.974 for normalized Mantel Statistic Z) demonstrated that CC represents the most of total genetic variation with minimum redundancy in of Turkey (Tas et al., 2019). Surprising findings of correlations were noticed for seed weight with seed length (R^2 = 0.259) by (Sari et al., 2020). A strong positive correlation among morphological traits was observed by Samadia (2002) in bottle gourd genotypes. The results pertaining to this study are in support with the findings of (Gürcan et al., 2015).

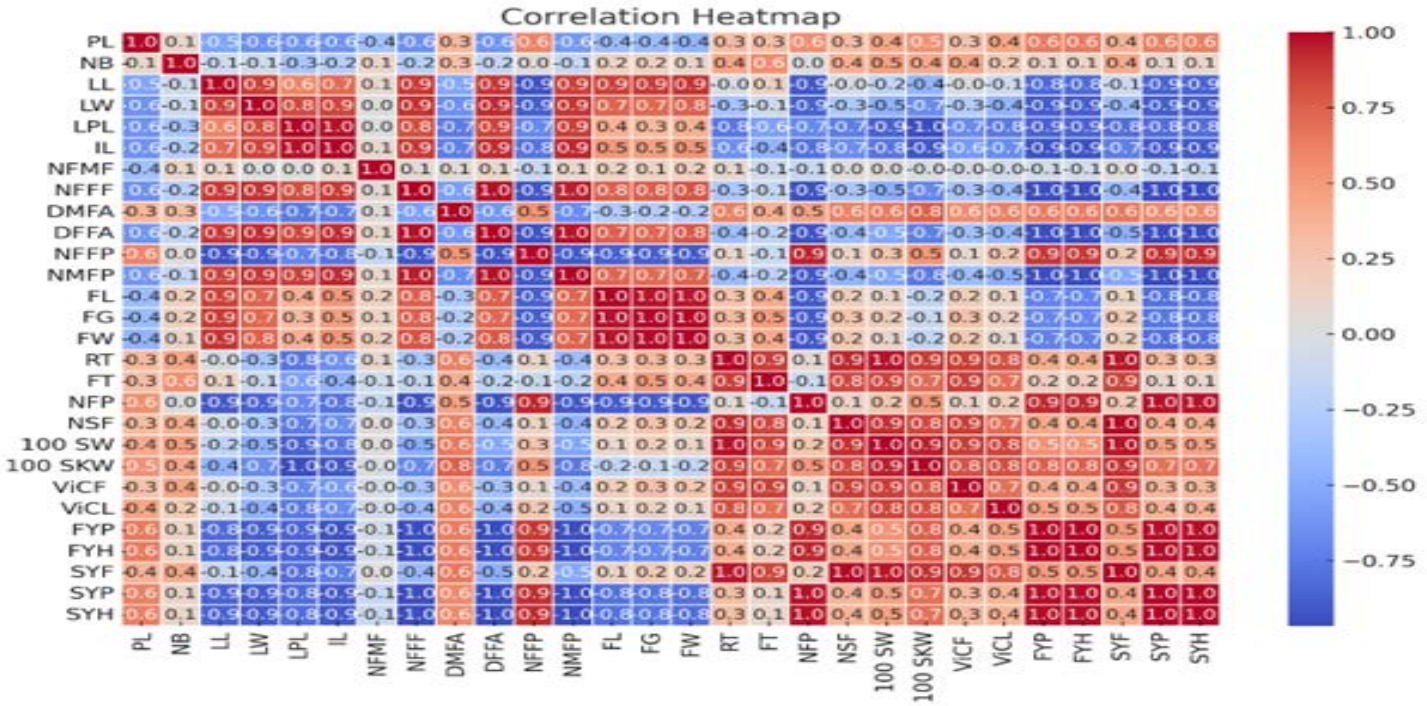

Fig. 1. Pearson correlation coefficients among all 28 traits calculated by using the BLUPs. (Significant at * P≤0.05, ** P≤0.01, and *** P≤0.001)

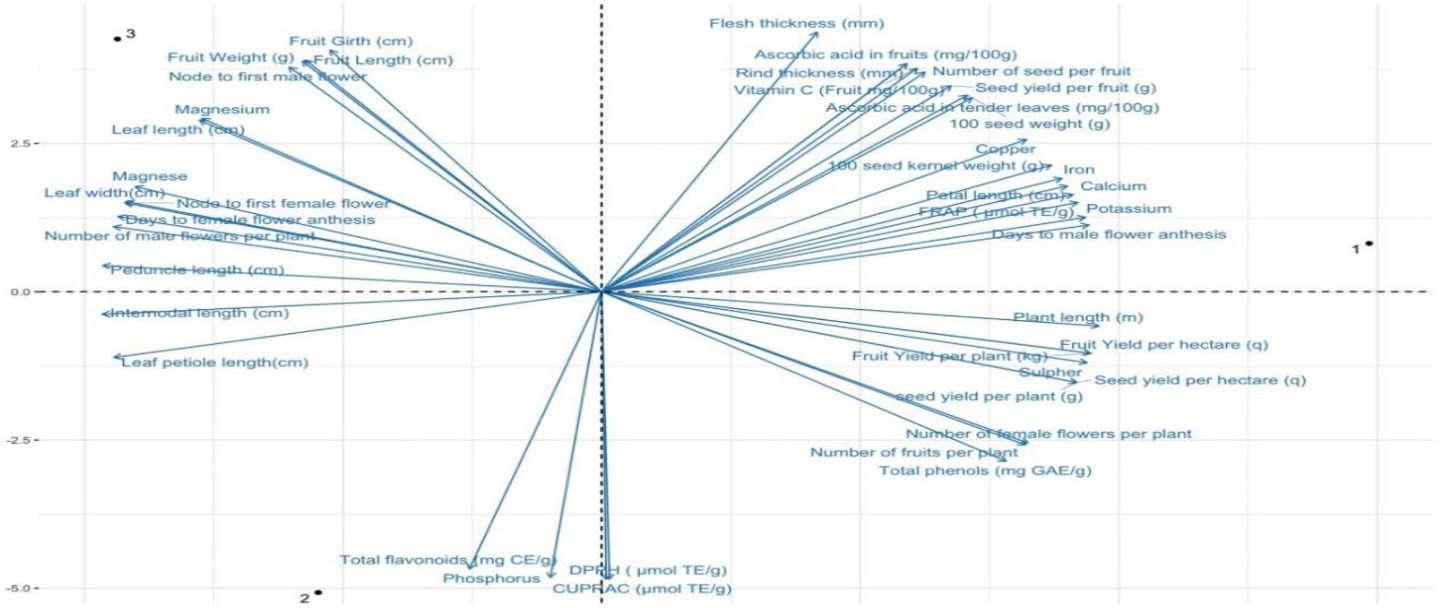

Fig. 2. Genotype by trait biplot graphical display of the measured traits in bottle gourd genotypes

The analysis of variance (ANOVA) revealed significant variation among the three varieties of bottle gourd for 29 different physiological traits were depicted in Table 1. This indicated the presence of high degree of variation within the genotypes. One of the ways by which variability is assessed through a simple approach of examining the range of variations. Range of variation observed for all the traits is presented in Table 2.

Various genetic traits like phenotypic and genotypic coefficient of variability (PCV, GCV), heritability, genetic advance (GA) and genetic advance as per cent of mean (GAM) for the 29 traits like PL-Plant length (m), LL-Leaf length (cm), LW-Leaf width (cm), LP-Leaf petiole length (cm), IL-Internodal length (cm), NFMF-Node to first male flower, NFFF-Node to first female flower, DMFA-Days to male flower anthesis, DFFA-Days to female flower anthesis, NFFP-Number of female flowers per plant, NMFP-Number of male flowers per plant, PtL-Petal length (cm), FL-Fruit length (cm), FG-Fruit girth (cm), PdL-Peduncle length (cm), FW-Fruit weight (g), RT-Rind thickness (mm), FT-Flesh thickness (mm), NFP-Number of fruits per plant, NSF-Number of seed per fruit, SW-100 seed weight (g), SKW-100 seed kernel weight (g) AAF-Ascorbic acid in fruits (mg/100g), AATL-Ascorbic acid in tender leaves (mg/100g), FYP-Fruit yield per plant (kg), FYH-Fruit yield per hectare (q), SYF-Seed yield per fruit (g), SYP-Seed yield per plant (g), SYH-Seed yield per hectare (q), High PCV and GCV were recorded for the TF (24.5398 and 25.1457), LPL (32.812 and 33.0797), IL (26.4953 and 26.6832), NFFF (44.5599 and 45.3748), DFFA (23.9766 and 24.1199), NFFP (60.336 and 61.3191), NMFP (26.8572 and 27.2372), FL (30.2802 and 31.4789), FG (50.0635 and 50.2592), FW (66.1507 and 66.3744), NFP (66.752 and 68.1876), FYP (37.6462 and 38.6746), FYH (37.6464 and 38.6749), SYP (74.9487 and 76.552) and SYH (74.9489 and 76.5522).

The findings revealed that the estimates of the phenotypic PCV were greater than the GCV for all the evaluated traits which signifying that the apparent variation is not only due to the genetic effects but also due to environmental effects. While, the variation between PCV and GCV for most of the traits were small, indicating high possibility of genetic progress through selection, apparently, the environmental impact on any trait is indicated by magnitude of the differences between the GCV and PCV. Whereas large differences reflect a large environmental influence, while small differences show a high genetic influence, and these findings conformed with the findings of (20). Moderate PCV and GCV were recorded for antioxidants including TP (16.1697 and 16.3706), CUPRAC (17.4532 and 18.1472), FRAP (18.232 and 18.9743), AAF (10.8654 and 10.9649); mineral content including Ca (10.9894 and 13.2227); and physiological traits including PL (11.5225 and 15.8628), LL (10.3656 and 10.8454), LW (11.6807 and 12.3559), PdL (10.8469 and 11.4547), RT (19.414 and 19.6855), FT (10.2848 and 10.748), AAF (10.5699 and 11.5069), and SYF (14.5073 and 14.7749). Besides this, remaining traits exhibited lower PCV and GCV.

These results explained the existence of limited variability or low genetic variability in the bottle gourd varieties for the trait. High heritability (>60%) was noticed for all the characters during study except DPPH P, Mg, S, Mn, PL and DMFA). Which indicates that these traits are less influenced by environmental factors and are under the control of additive gene effect and selection for improvement of such characters would be rewarding. GCV along with heritability estimates would provide a better picture of the amount of advance expected by phenotypic selection. High heritability coupled with high genetic gains is more effective and dependable in

predicting the improvement through selection. High genetic advance as per cent mean was observed for TP (22.07%), AAF (21.38%), K (67.44%), DFFA (29.85%), NFFP (23.64%), FL (25.74%), FG (49.31%), FW (99.61%), FT (22.68%), NFP (25.29%), NSF (22.62%), FYP (26.93%), FYH (23.88%) and SYP (45.51%) indicating that these traits are controlled by additive gene action.

Further selection for these characters will improve the different traits. Moderate genetic advance as per cent mean was observed for P (14.72%), Ca (14.42%), LPL (10.07%), NFFF (12.13%), SYP (17.69%) and SYH (15.22%). Whereas, low genetic advance as per cent mean was observed for CUPRAC, FRAP, DPPH, Mg, S, Fe, Mn, PL, LL, LW, IL, DMFA, NMFP, PtL, PdL, RT, SW, SKW, AAF and AATL governed by non additive gene action and selection for these traits is not useful. Similar results were reported by (Gürcan et al., 2015; Sirohi et al. 1988; Samadia, 2002; Yadav et al. 2022; Yadav et al. 2023; Yadav et al., 2023).

Table 1. Analysis of Variance (ANOVA) for 28 different physiological traits in bottle gourd genotypes

Sources of Variation	Mean Square		
	Replication (DF=7)	Genotypes (DF=2)	Residuals (DF=14)
Plant length (m)	2.04	3.175**	2.237
Leaf length (cm)	0.074	14.520***	0.169
Leaf width (cm)	0.454	35.808***	0.525
Leaf petiole length(cm)	0.456	194.514***	0.398
Internodal length (cm)	0.03	45.191***	0.08
Node to first female flower	2.47	289.042***	1.327
Days to male flower anthesis	4.571	70.542***	5.589
Days to female flower anthesis	8.85	1702.17***	2.55
Number of female flowers per plant	2.95	1093.04***	4.47
Number of male flowers per plant	34.6	8368***	29.7
Petal length (cm)	0.003	0.395***	0.027
Fruit length (cm)	1.5	509.81***	5.09
Fruit girth (cm)	1.3	4624.8***	4.5
Peduncle length (cm)	0.093	6.361***	0.090
Fruit weight (g)	8490	438652***	3711
Rind thickness (mm)	0.0233	5.073***	0.018
Flesh thickness (mm)	29.08	1071.63***	12.2
Number of fruits per plant	2.74	462.18***	2.5
Number of seed per fruit	14.64	1087.88***	15.02
100 seed weight (g)	0.007	10.257***	0.008
100 seed kernel weight (g)	0.001	0.849***	0.002
Ascorbic acid in fruits (mg/100g)	0.915	35.896***	0.812
Ascorbic acid in tender leaves (mg/100g)	2.64	62.711***	2.25
Fruit yield per plant (kg)	0.681	96.11***	0.661
Fruit yield per hectare (q)	756	106790***	734
Seed yield per fruit (g)	0.586	116.32***	0.539
Seed yield per plant (g)	2369	412087***	2215
Seed yield per hectare (q)	2.63	457.87***	2.46

Significant at *P≤0.05, ** P≤0.01, and *** P≤0.001

Table 2. Maximum, minimum, mean, GCV, PCV, heritability (h^2) and genetic advance of 28 traits

Character	Max	Min	Grand Mean	GCV	PCV	h^2 (%)	Genetic Advance %
PL	5.1	2.9	3.666	11.523	15.863	52.76	0.63

LL	14.8	11	12.920	10.366	10.845	91.35	2.64
LW	20.7	14.8	17.979	11.681	12.356	89.37	4.09
LPL	19.8	9.1	15.012	32.812	33.080	98.39	10.07
IL	10.8	6	8.962	26.495	26.683	98.6	4.86
NFFF	22	6	13.458	44.560	45.375	96.44	12.13
DMFA	53	42	47.833	5.957	7.740	59.23	4.52
DFFA	79	44	60.791	23.977	24.120	98.82	29.85
NFFP	32	5	19.333	60.336	61.319	96.82	23.64
NMFP	163	80	120.208	26.857	27.237	97.23	5.58
PtL	14.4	13.5	13.937	1.539	1.944	62.67	0.35
FL	37.3	19.2	26.231	30.280	31.479	92.53	25.74
FG	77.6	29.79	48.002	50.064	50.259	99.22	49.31
PdL	9.2	6.8	8.162	10.847	11.455	89.67	1.73
FW	2118	583	1118.915	66.151	66.374	99.33	99.61
RT	4.87	3.04	4.094	19.414	19.686	97.26	1.62
FT	126	93.8	111.890	10.285	10.748	91.57	22.68
NFP	21	2.84	11.355	66.752	68.188	95.83	25.29
NSF	146	112	130.25	8.891	9.376	89.93	22.62
SW	21.19	18.71	20.046	5.646	5.664	99.36	2.32
SKW	8.84	8.09	8.431	3.860	3.900	97.96	0.66
AAF	23.1	17.1	19.812	10.570	11.507	84.38	3.96
AATL	35.6	24.7	31.3	8.783	10.005	77.06	4.97
FYP	14.03	5.10	9.175	37.646	38.675	94.75	26.93
FYH	467.66	170.02	305.843	37.646	38.675	94.75	23.88
SYF	30.92	21.35	26.223	14.507	14.775	96.41	17.69
SYP	649.3	74.23	302.005	74.948	76.552	95.86	45.51
SYH	21.65	2.47	10.066	74.948	76.552	95.86	15.22

Conclusions

The physiological traits in bottle gourd showed significant variation among genotypes, with strong positive correlations observed for traits like fruit length and weight, and yield-related traits. Genetic analysis revealed high heritability and genetic advance for traits such as fruit yield and fruit girth, indicating their potential for selection. Overall, traits with high genetic advance, such as fruit yield, are most suitable for selection, while those influenced by non-additive gene action are less responsive to selection. Keeping in view of the above significant findings and wider adaptability of the variety Thar Avani under rainfed semi-arid conditions recommended for commercial cultivation with its important characters viz. earliness in female flowering and round fruit shape.

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Conflict of Interest

The authors have no conflict of interest.

Data Sharing

All relevant data are within the manuscript.

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Research Article

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Genetic variability analysis in onion (*Allium cepa* L.) genotypes

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ABSTRACT

Genetic variability parameters *viz*., GCV, PCV, heritability, genetic gain, mean performance and range were assessed among twelve varieties of onion during *Kharif*, 2023 with adopting randomized block design and three replications. Results revealed that phenotypic coefficient of variation were higher than corresponding genotypic coefficient of variation for all the traits studied and magnitude of various variability parameters were significantly differ among the genotypes studied. Highest heritability (98.95%) has been reported in fresh weight of bulb, followed by dry weight of bulb (98.83%) and neck diameter (96.64%). Bhima Dark Red was best performing variety on the basis of yield per ha, followed by Bhima Super and Bhima Red yielding 298.24, 279.64 and 266.84 q/ha, respectively.

Introduction

Onion (*Allium cepa* L.) is the second most widely cultivated vegetable crop belonging to *Alliaceae* family. In India, it occupies an area of 1.64 million hectare with the production of 26.83 million tonnes and productivity is 16.36 tonnes per ha (Anonymous, 2021). It has predominantly expansion as a food source and value addition for a range of meals, it can be eaten raw (salads) or cooked as well as in processed form *e.g.,* flakes.

The availability of sufficient genetic variability is of immense importance in a crop improvement programme as high variability is required for effective selection. Therefore, it is essential for a plant breeder to assess the variability parameters mean performance, range, phenotypic coefficient of variation, genotypic coefficient of variation, heritability, genetic advance and genetic gain, as these parameters give the information regarding the availability of genetic variability for different characters in available germplasm. Hence, study of genetic variability of bulb yield and its contributing traits among different genotypes provides a strong basis for selection of desirable genotypes for augmentation of yield and other yield attributing characters (Amerullah *et al*., 2021).

Material and Methods

The present study was conducted at open field of Hi-tech Unit, Department of Horticulture, Rajasthan College of Agriculture, and Udaipur from June, 2023 to February, 2024 located at 24°34' 50.0556" N latitude and 73° 42' 19.8648" E longitude. Twelve onion varieties *viz.,* Bhima Super, Bhima Raj, Bhima Safed, Bhima Dark Red, Bhima Shubhra, Bhima Red, N-241, Bhima Light Red, Agrifound Light Red, Baswant-780 and Desi Tejas were planted with row-to-row distance of 30 cm and plant-to-plant distance of 10 cm in

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Randomized Block Design (RBD) with three replications. All the recommended cultural practices for raising healthy crop and plant protection measures were followed as per POP of Zone IV a (Kaushik and Ameta, 2014). Observations were recorded from five randomly selected plants in each plot for various quantitative characters and the data were subjected to statistical analyses. Suitable methodology was followed for recording various traits *i.e.* days to emergence was recorded by counting days from sowing to germination, plant height (cm) was measured from base of plant to top of longest leaf with the help of measuring scale and average was worked out, number of leaves per plant was counted in five randomly selected and tagged plant at maturity, leaf length (cm) was measured from joint of the leaf lamina to the tip of the leaf at maturity with the help of measuring scale, leaf width (mm) was measured from broad part of the leaf at maturity with the help of measuring scale, days to maturity was recorded by counting days from the day of planting seedlings till the bulbs reached to maturity, neck thickness (mm) was measured with the help of vernier caliper from below the joint of leaf lamina, bulb length (cm) was recorded by measuring the length between two polar ends of the bulb with the help of vernier caliper at harvest, bulb diameter (cm) was recorded from the point of maximum width of the bulb across the polar length, fresh weight of bulb (g) was recorded by selecting five randomly tagged plant from each treatment and replication and weighed after harvest with the help of digital electronic balance and expressed in grams and average was worked out, similarly dry weight of bulb (g) was recorded after curing and bulb yield (q/ha) was recorded by weighing all the bulbs of each experimental plot and derived yield per ha in quintals. The phenotypic coefficient of variation (PCV), genotypic coefficient of variation (GCV), heritability (h^2), genetic advance (GA) and the expected genetic gain (GG) for different characters were analyzed as per procedure suggested by Burton and Devane (1953) and Johnson *et al* (1955), respectively.

Result and Discussion

Analysis of variance for twelve characters indicated that there is considerable variation in respect to all the traits, the results of mean, range, GCV, PCV, heritability, genetic advance and genetic gain for various characters of 12 genotypes of onion (Table 1) clearly showed higher magnitude of genetic variability in material studied.

High magnitude of genotypic as well as phenotypic coefficient of variation were recorded for traits *viz*., leaf width (22.07% and 22.62%), neck diameter (23.00% and 23.39%), fresh weight of bulb (34.95% and 35.13%), dry weight of bulb (35.16% and 35.37%) and bulb yield (23.41% and 24.80%). Similar results were reported by Dwivedi *et al*. (2017) for neck thickness, Amerullah *et al*. (2021) for fresh weight of bulb and Hosamani *et al*. (2010) for bulb yield while working with onion, further, phenotypic coefficient of variation were higher than corresponding genotypic coefficient of variation for all the traits studied, indicating influence of environment on expression of traits in various genotypes. The earlier findings of Chattopadhyay *et al*. (2013), Khosa and Dhatt (2013), Lakshmi (2015), Dwivedi *et al*. (2017), Dangi *et al*. (2018), Manjunath and Hiremath (2022), Prakash *et al*. (2022) and Yogita *et al*. (2023) were in same line as they also reported high PCV then corresponding GCV while working on onion.

The majority of the traits had high heritability and it was recorded highest for fresh weight of bulb (98.95%), dry weight of bulb (98.83%), neck diameter (96.64%), days to maturity (96.43%), leaf width (95.20%), bulb diameter (92.74%), bulb yield (89.08%) and number of leaves per plant (83.08%). However, moderate heritability was observed for bulb length (58.28%), leaf length (44.48%), days to emergence (44.16%) and plant height at maturity (30.30%). The highest heritability was recorded for fresh weight of bulb which was cent percent followed by dry weight of bulb, neck diameter, days to maturity, leaf width, bulb diameter, bulb yield and number of leaves per plant were showed high heritability (>60 per cent). High heritability have also been reported by Amerullah *et al*. (2021) for fresh weight of bulb, Hosamani *et al*. (2010) for bulb yield, whereas Parmar *et al*. (2018) observed high heritability for neck diameter, similarly, Khosa and Dhatt (2013), Dangi *et al*. (2018), Singh *et al*. (2011) and Bharti *et al*. (2011) reported high heritability for days to maturity, leaf width, bulb diameter and bulb weight, respectively, while working with onion. The moderate heritability (30-60 per cent) were observed for bulb length, leaf length, days to emergence and plant height at maturity. These findings are in close conformity of Parmar *et al*. (2018) as they reported moderate heritability for bulb length and plant height of onion. High heritability coupled with high genetic advance (89.08% and 103.40%) was recorded for bulb yield q/ ha, (98.95% and 86.06%) for fresh weight of bulb and (98.83% and 78.14%) for dry weight of bulb.

The highest genetic advance as percent of mean (genetic gain) was recorded for the dry weight of bulb (72.01%), fresh weight of bulb (71.61%), neck diameter (46.57%), bulb yield (45.51%), leaf width (44.36%), bulb diameter (37.03%) and number of leaves per plant (22.38%). Genetic gain is an important parameter of genetic variability as it shows potentialities of the improvement for particular trait in breeding programme. Seven characters out of twelve showed higher genetic gain *i.e*. more than 20 per cent (Johnson *et al.,* 1955), these characters are dry weight of bulb, fresh weight of bulb, neck diameter, bulb yield, leaf width, bulb diameter and number of leaves per plant, further heritability for above traits was also high, indicated that these characters are governed by additive gene action and thus direct selection of these traits could be effective in crop improvement programme, hence

these traits can be keep on priority for selection. Higher genetic gain accompanied with high heritability has also been reported by Parmar *et al.* (2018) for bulb yield, weight of bulb, number of leaves per plant and neck thickness of bulb, similarly Prakash *et al.* (2023) seen same trend for fresh bulb weight, dry bulb weight and bulb yield while working with onion. The findings of Srivastav *et al.* (2017), Dangi *et al.* (2018), Solanki *et al.* (2015) and Dwivedi *et al.* (2017) was in conformity with the results of present investigation as they also reported high genetic advance as per cent of mean along with high heritability for the various traits of onion.

Mean performance (Table 2 & 3) for various traits in genotypes showed that the highest bulb yield (298.24 q/ha) and fresh weight of bulb (183.92 g) was recorded for genotype Bhima Dark Red which was at par with genotype Bhima Super (279.64 q/ha and 176.10 g, respectively). Further, mean values for most of traits were higher and in desirable direction for Bhima Dark Red and Bhima Super varieties. A wide range of bulb yield (105.91-298.24 q/ha), fresh weight of bulb (66.02-183.92 g), dry weight of bulb (59.64-164.75 g), days to maturity (105-127 days), leaf length (43.45-55.49 cm) and plant height at maturity (49.48-59.08 cm) were observed for various genotypes.

Table 1. Range, mean, GCV, PCV, heritability, genetic advance and genetic gain in *kharif* onion

S.No.	Characters	Range	Mean	GCV	PCV	ECV	h^2	GA	GG
1	Days to emergence	4.80-6.07	5.58	6.60	9.94	7.43	44.16	0.50	9.04
2	Plant height at maturity (cm)	49.48-59.08	52.85	4.32	7.85	6.56	30.30	2.59	4.90
3	Number of leaves/ plant	8.20-11.53	9.61	11.92	13.08	5.38	83.08	2.15	22.38
4	Leaf length (cm)	43.45-55.49	48.33	6.28	9.42	7.02	44.48	4.17	8.63
5	Leaf width (mm)	4.75-11.38	8.74	22.07	22.62	4.95	95.20	3.88	44.36
6	Neck diameter (mm)	7.30-15.85	11.35	23.00	23.39	4.29	96.64	5.28	46.57
7	Bulb length (cm)	5.42-6.87	6.26	7.49	9.81	6.34	58.28	0.74	11.78
8	Bulb diameter (cm)	5.40-9.31	7.02	18.66	19.38	5.22	92.74	2.60	37.03
9	Days to maturity	105.43-127.27	116.45	6.02	6.13	1.16	96.43	14.19	12.18
10	Fresh weight of bulb (g)	66.02-183.92	120.17	34.95	35.13	3.60	98.95	86.06	71.61
11	Dry weight of bulb (g)	59.46-164.75	108.51	35.16	35.37	3.83	98.83	78.14	72.01
12	Bulb yield (q/ ha)	105.91-298.24	88.05	23.41	24.80	8.19	89.08	103.40	45.51

Table 2. Mean values for days to emergence and vegetative parameters of *kharif* onion

S. No.	Genotype	Days to emergence	Plant height at maturity (cm)	Number of leaves/ plant	Leaf length (cm)	Leaf width (mm)	Neck diameter (mm)
1	Bhima Super	6.07	57.98	11.13	53.88	10.82	14.52
2	Bhima Raj	5.80	51.54	9.20	47.59	9.02	11.28
3	Bhima Safed	5.87	51.25	8.70	46.90	8.28	10.62
4	Bhima Dark Red	5.67	59.08	11.53	55.49	11.38	15.85
5	Bhima Shubhra	6.03	49.48	8.20	43.45	4.75	7.30
6	Bhima Red	4.83	52.68	10.33	47.42	9.65	12.38
7	N-241	5.57	51.79	9.43	47.57	9.29	11.46
8	Bhima Light Red	5.60	50.88	8.67	46.38	8.07	9.78
9	N-53	5.73	53.17	10.53	49.89	9.67	12.86
10	Agrifound Light Red	4.80	50.27	8.40	44.34	6.09	7.80
11	Baswant-780	5.10	55.11	10.73	50.93	10.46	13.40
12	Desi Tejas	5.93	50.93	8.43	46.12	7.43	8.91
GM		5.58	52.85	9.61	48.33	8.74	11.35
SEm±		0.24	2.00	0.30	1.96	0.25	0.28
CD at 5%		0.70	5.87	0.88	5.74	0.73	0.82

CD at 1%	0.95	7.98	1.19	7.81	1.00	1.12
CV (%)	7.43	6.56	5.38	7.02	4.95	4.29

Table 3. Mean values for days to maturity, bulb traits and bulb yield of *kharif* onion

S. No.	Genotype	Bulb length (cm)	Bulb diameter (cm)	Days to maturity	Fresh weight of bulb (g)	Dry weight of bulb (g)	Bulb yield (q/ ha)
1	Bhima Super	5.42	8.66	118.47	176.10	157.73	279.64
2	Bhima Raj	6.35	6.50	122.33	110.52	97.42	243.01
3	Bhima Safed	6.87	6.34	105.43	93.15	84.03	212.46
4	Bhima Dark Red	6.56	9.31	121.67	183.92	164.75	298.24
5	Bhima Shubhra	6.29	5.44	120.57	66.02	59.46	105.91
6	Bhima Red	5.43	7.82	113.73	133.80	122.88	266.84
7	N-241	6.38	6.85	108.50	123.22	113.30	246.38
8	Bhima Light Red	6.83	6.09	110.23	86.18	78.41	214.22
9	N-53	6.54	7.52	122.70	151.45	137.09	257.99
10	Agrifound Light Red	6.51	5.40	118.60	71.97	63.22	161.87
11	Baswant-780	5.48	8.51	127.27	166.75	152.73	251.57
12	Desi Tejas	6.48	5.76	107.87	78.96	71.04	188.61
GM		6.26	7.02	116.45	120.17	108.51	227.23
SEm±		0.23	0.21	0.78	2.50	2.40	10.75
CD at 5%		0.67	0.62	2.29	7.33	7.03	31.52
CD at 1%		0.91	0.84	3.11	9.97	9.56	42.88
CV (%)		6.34	5.22	1.16	3.60	3.83	8.19

Conclusion

From the results of investigation, it is concluded that the Bhima Dark Red was found best performer for bulb yield per hectare followed by Bhima Super, Bhima Red and N-241. Further a wide range of variability were observed for most of traits in varieties in terms of GCV, PCV, heritability and genetic gain which could be utilized in breeding programmes to develop desirable genotypes of *kharif* onion.

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Conflict of Interest

The authors have no conflict of interest.

Data Sharing

All relevant data are within the manuscript.

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Research Article

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Accessing the higher production with integrated nutrient management in Custard apple cv. Arka Sahan

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ABSTRACT

The experiment evaluated different levels of recommended doses of fertilizers (RDF), farmyard manure (FYM), vermicompost, and biofertilizers like Azotobacter (AZB) and Phosphorus Solubilizing Bacteria (PSB) in custard apple cv. Arka Sahan. Treatment 75% RDF + VC @ 2 kg/plant + AZB + PSB (T_9) showed the highest plant height (400 cm), plant spread (East-West 409.96 cm, North-South 481.36 cm), number of branches (58.92), fruits per plant (31.5), and yield per plant (9.08 kg). The highest fruit weight (350.65 g) was recorded with 50% RDF + VC @ 4 kg/plant + AZB + PSB (T_{10}), while 100% RDF + 20 kg FYM (T_2) resulted in the maximum stem girth (37.49 cm). These findings underline the efficacy of integrated nutrient management in enhancing the growth and yield of custard apple cv. Arka Sahan, offering valuable insights for its cultivation under Madhya Pradesh's conditions.

Introduction

Custard apple (*Annona squamosa* L.) is a tropical fruit tree, also known as sugar apple, sweetsop, sharifa, sitaphal and noi-na in different parts of growing regions. It is part of the Annonaceae family, with over 120 species and 40 genera, only five of which are edible. The custard apple is known to have originated from the West Indies and South America. Currently, custard apple cultivation is practised in several countries including Australia, Brazil, Chile, Egypt, India, Israel, Philippines, Spain, Sri Lanka and the USA (Nakasone and Paull, 1998).

Among the Annonas, the Sugar apple (*Annona squamosa* L.) holds important value. Custard apple is a small, semi-deciduous, much branched shrub or small tree 3 to 8 m tall with a broad, open crown or irregularly spreading branches and a short trunk, not buttressed at base. Branches with light brown bark and visible leaf scars, inner bark is light yellow in colour and slightly bitter, twigs become brown with light brown dots. Custard apple is considered as a crop for a wasteland and successfully grown in sandy, rocky gravel and heavy soil, even in sandy loam soils. Custard apple can tap a considerable volume of soil with its extensive root system under natural habitat. However, the natural fertility of soils is rarely sufficient to give economic yields.

In sand culture grown custard apple saplings had nitrogen deficiency that was characterized by restricted growth of plants with pale green to yellowish leaves.

Phosphorus deficiency leads to growth reduction, appearance of brown necrotic bands at the tips and margin of leaves, while potassium deficiency produces marginal scorching of leaves (Sadhu and Ghosh, 1976). However, in most of the orchard, poor nutrition is one of the major causes of low productivity. Plants need sufficient nutrients

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in proper balance for normal growth and development. Depletion soil nutrients pose a major threat to sustainability of fruit production and underline the need for maintaining it by tapping other plant nutrient sources. The reduction in the soil fertility has resulted in low productivity of the crop. Besides, the increasing cost of fertilizers and their negative effect on soil health has led to intensified attempts to the use of biofertilizers and organic matter along with inorganic fertilizers.

Integrated Nutrient Management (INM) is an approach to managing nutrient requirements in crops that aims to optimize nutrient use efficiency, improve soil health, and enhance crop productivity. This approach involves the judicious use of organic and inorganic fertilizers, along with other agronomic practices, to meet the nutrient needs of the crop while minimizing environmental impacts. This approach is necessary to achieve high crop yields and maintain optimal nutrient levels in the soil, thereby ensuring the production of high-quality fruits (Ganeshamurthy *et al*., 2015).

Material and Methods

The experiment was conducted during crop season 2021-22 and 2022-23 with nine-year-old plants of custard apple cultivar Arka Sahan in the experimental orchard of AICRP-Arid Zone Fruit at Jabalpur center under the Department of Horticulture, Jawaharlal Nehru Krishi Vishwa Vidyalaya, Jabalpur (M.P.). The experimental location is located at 23°22'16.6"N latitude and 79°96'85.9"E longitude with an altitude of 1273 feet. Experiment was conducted comprising 10 treatments namely T_1 (100 % RDF), T_2 (100 % RDF + 20 kg FYM), T_3 (75% RDF + FYM @ 5 kg/plant), T_4 (50% RDF+FYM @ 10 kg/plant), T_5 (75% RDF + FYM @ 5 kg/plant + AZB + PSB), T_6 (50% RDF + FYM @ 10 kg/plant + AZB + PSB), T_7 (75% RDF + Vermi compost @ 2 kg/plant), T_8 (50% RDF + Vermicompost @ 4 kg/plant), T_9 (75% RDF + Vermicompost @ 2 kg/plant+ AZB+PSB), T_{10} (50% RDF +Vermi compost @ 4 kg/plant+ AZB + PSB).

The recommended dose of fertilizer (RDF) consisted of 250g of nitrogen (N), 125g of phosphorus (P2O5), and 125g of potassium (K2O).

The recommended dose of fertilizer (RDF) per plant consisted of 250 g of N, 125 g of P_2O_5, and 125 g of K_2O. Additionally, 80g of Azotobacter (AZB) and 80g of phosphorus solubilizing bacteria (PSB) were applied per tree. The soil condition of experimental orchard is vertisol having dark colour and is described as having a medium to deep depth. The experiment was carried out in Randomized block design (RBD) with three replications having single plant per treatment. Orchard was established in high density planting following 6m x 6m of spacing for plant to plant and row to row.

The physical attributes of the plants were meticulously measured using standardized techniques. Plant height was determined using an altimeter and expressed in centimetre, while stem girth was measured with vernier callipers. Plant spread was recorded at noon using a meter tape, capturing measurements in both East-West and North-South directions, and the average plant spread in each direction were calculated in meters. The average fruit weight was assessed using an electronic weighing machine and recorded in grams. Additionally, the number of branches, fruits per plant and yield per plant were counted and recorded.

For statistical analysis, pooled mean data from two consecutive years were used to ensure the robustness and reliability of the results. Analysis of variance (ANOVA) was performed using the R statistical package to evaluate the significance of the treatment effects.

Result and Discussion

The data obtained from present study showed that various treatment doses of integrated nutrients are significantly affecting plant height, stem girth, plant spread in east-west and north-south direction, number of branches, average weight of fruit, number of fruits per plant and yield per plant. Growth parameters viz, plant height, stem girth, plant spread in east-west and north-south directions, and number of branches are presented in Table 1 and Table 2 in which the treatments differ significantly from each other.

The maximum plant height (360.39 cm, 440.5 cm and 400.45 cm), maximum plant spread in east-west (450.45 cm, 531.46 cm and 490.96 cm), in north- south direction (431.25 cm, 530.46 cm and 481.36 cm) and higher number of branches (54.2, 63.63 and 58.92) was observed under T_9 (75% RDF + Vermicompost @ 2 kg/plant+ AZB + PSB) in the year 2021-22, 2022-23 and pooled data, respectively. The significant increase in plant height is due to the improvement of physical properties of soil, higher nutrient uptake, increased activity of microorganisms with the vermicompost can improve plant growth, reduce nitrogen losses which were manifested in the form of enhanced growth as also confirmed by Kumar *et al*. (2008).

Phosphate solubilizing bacteria (PSB) play an essential role in P cycling and promoting plant growth by increasing its P uptake in rhizosphere soils. Most PSB produces indole-3-acetic acid (IAA) which enables plant cells to grow, RNA/protein synthesis thus increasing plant growth. Canopy spread is more in east-west direction. Nitrogen, Phosphorus and Potassium in combination with vermicompost, AZB, PSB fertilizer can enhance vegetative growth in plants. Plants often have a greater exposure to sunlight when positioned in an east-west direction, since this alignment enables them to harness solar radiation from the eastern horizon at sunrise to the western horizon during sunset, so maximising their daily light intake. Positive response of Azotobacter and PSB were also reported in mango by Yadav *et al*. (2011). These

results are in close conformity with the findings of Singh *et al*. (2009).

The data presented in Table 3 and Fig. 1 clearly demonstrate the significant impact of INM treatments on fruit weight, number of fruits per plant, and yield per plant of custard apple cv. Arka Sahan. Higher number of fruits per plant (27, 36 and 31.5) and maximum yield/ plant (5.86 kg, 12.3 kg and 9.08 kg) were also reported in T_9 (75% RDF + Vermicompost @ 2 kg/plant+ AZB + PSB) in the year 2021-22, 2022-23 and pooled data, respectively. The observed increase in the number of fruits per plant might potentially be attributed to the favourable impact of INM on the extraction of nutrients from the soil by crops, as well as the solubilization effect of plant nutrients via the addition of vermicompost, Azotobactor and Phosphate solubilizing bacteria (Subbiah *et al*., 1982).

The rise in number of fruits can potentially be attributed to the increase in nutrient levels in the assimilating area of the crop, which is a result of the rational partitioning of dry matter to the economic sink. This allocation of resources has led to an increase in the yield attributes. These findings are consistent with the research conducted by Dalal *et al*. (2004), who observed a higher number of fruits per plant and yield per plant through the integrated application of nutrients in sapota. In their study, Mandal and Chattopadhyay (1993) observed that higher dosages of fertilisers resulted in increased yields in custard apple. This effect was ascribed to the promotion of strong vegetative growth, as well as enhanced development and reproduction of the plant.

Maximum stem girth (32.55 cm, 42.42 cm and 37.49 cm) was recorded under T_2 (RDF 100% + 20 kg FYM) in the year 2021-22, 2022-23 and pooled, respectively. Nitrogen, phosphorus, and potassium are vital elements that have an impact on the augmentation of stem diameter via the facilitation of cellular division, elongation and the establishment of robust with well organized stems. Optimal potassium levels are associated with enhanced water absorption and translocation mechanisms inside the plant, hence facilitating an increase in stem diameter and promoting overall plant development (Marschner, 2012).

Maximum average fruit weight (261.27 g, 440.04 g and 350.65 g) was recorded in T_{10} (50% RDF + VC @ 4 kg/ plant+ AZB+PSB) in the year 2021-22, 2022-23 and pooled data, respectively. Improvement in fruit weight in response to organic source of nutrients, also have been reported by Yadav *et al*. (2007) in aonla and Yadav *et al*. (2011) in mango. The optimal delivery of plant nutrients is crucial in ensuring the right quantity of nutrients is available during the whole time of fruit development. This eventually leads to the accumulation of greater photosynthesis, resulting in increased fruit weight and other physical characteristics (Lal and Dayal, 2014).

Table 1. Effect of INM on plant height and stem girth of custard apple

Treatments	Plant height (cm)			Stem girth (cm)		
	2021-22	2022-23	Pooled	2021-22	2022-23	Pooled
RDF 100% (T_1)	298.16	324.6	311.38	19.20	26.56	22.88
RDF 100% + 20 kg FYM (T_2)	306.34	376.6	341.47	32.55	42.42	37.49
75% RDF + FYM @ 5 kg/plant (T_3)	222.01	290.26	256.14	23.97	30.50	27.24
50% RDF + FYM @10 kg/plant (T_4)	350.32	431.16	390.74	25.40	32.50	28.95
75% RDF + FYM @ 5 kg/plant +AZB+PSB (T_5)	234.3	294.26	264.28	27.63	35.47	31.55
50% RDF + FYM @ 10 kg/plant +AZB+PSB (T_6)	268.31	328.33	298.32	24.83	32.98	28.90
75% RDF + VC @ 2 kg/plant (T_7)	302.77	390.46	346.62	24.25	31.75	28.00
50% RDF + VC @ 4 kg/plant (T_8)	258.39	320.7	289.55	19.75	25.56	22.66
75% RDF + VC @ 2 kg/plant+ AZB+PSB (T_9)	360.39	440.5	400.45	24.60	31.60	28.10
50% RDF + VC @ 4 kg/plant+ AZB+PSB (T_{10})	313.29	374.06	343.68	23.45	30.50	26.98
SEm±	5.06	0.68	1.14	1.0	0.58	0.26
CD (p=0.05)	15.02	2.03	3.27	2.97	1.73	0.74

Table 2. Effect of IMN on plant spread and number of branches of custard apple

Treatments	Plant spread East- West (cm)			Plant spread North-South (cm)			Number of branches		
	2021-22	2022-23	Pooled	2021-22	2022-23	Pooled	2021-22	2022-23	Pooled
RDF 100% (T_1)	255.31	322.60	288.96	316.52	370.50	343.51	34.60	41.53	38.07

RDF 100% + 20 kg FYM (T_2)	386.60	450.60	418.60	338.08	408.49	373.29	32.31	41.47	36.89
75% RDF + FYM @ 5 kg/plant (T_3)	358.67	415.60	387.14	311.52	370.83	341.18	22.04	30.34	26.19
50% RDF + FYM @10 kg/plant (T_4)	371.54	444.30	407.92	337.06	406.20	371.63	19.09	26.29	22.69
75% RDF + FYM @ 5 kg/plant +AZ-B+PSB (T_5)	377.08	454.60	415.84	383.21	445.43	414.32	40.20	50.20	45.20
50% RDF + FYM @ 10 kg/plant +AZ-B+PSB (T_6)	390.36	470.50	430.43	348.15	410.53	379.34	26.41	32.77	29.59
75% RDF + VC @ 2 kg/plant (T_7)	421.74	491.36	456.55	392.04	460.62	426.33	38.21	48.53	43.37
50% RDF + VC @ 4 kg/plant (T_8)	401.50	470.93	436.22	359.82	410.76	385.29	27.81	35.50	31.66
75% RDF + VC @ 2 kg/plant+ AZ-B+PSB (T_9)	450.45	531.46	490.96	431.25	531.46	481.36	54.20	63.63	58.92
50% RDF + VC @ 4 kg/plant+ AZ-B+PSB (T_{10})	422.52	492.43	457.48	418.61	492.43	455.52	49.25	56.02	52.64
SE(m)±	13.95	0.55	3.12	11.62	0.63	2.61	1.64	0.57	0.39
CD (p=0.05)	41.44	1.63	8.95	34.53	1.87	7.46	4.87	1.71	1.12

Table 3. Effect of INM on fruit traits and yield of custard apple

Treatments	Fruit weight (g)			Number of fruits/ plant			Yield/ plant (kg)		
	2021-22	2022-23	Pooled	2021-22	2022-23	Pooled	2021-22	2022-23	Pooled
RDF 100% (T_1)	238.74	301.30	270.02	16	22	19	3.81	6.63	5.22
RDF 100% + 20 kg FYM (T_2)	204.82	301.77	253.30	22	33	27.5	4.50	9.96	7.23
75% RDF + FYM @ 5 kg/ plant (T_3)	223.18	329.97	276.58	21	30	25.5	4.68	9.90	7.29
50% RDF + FYM @10 kg/ plant (T_4)	203.22	321.84	262.53	24	32	28	4.86	10.30	7.58
75% RDF + FYM @ 5 kg/ plant +AZB+PSB (T_5)	234.10	333.30	283.70	18	27	22.5	4.18	9.00	6.59
50% RDF + FYM @ 10 kg/ plant +AZB+PSB (T_6)	225.68	339.41	282.54	19	29	24	4.28	9.83	7.06
75% RDF + VC @ 2 kg/ plant (T_7)	217.00	323.69	270.35	26	35	30.5	5.65	11.33	8.49
50% RDF + VC @ 4 kg/ plant (T_8)	230.03	353.16	291.60	15	21	18.5	3.44	7.77	5.60
75% RDF + VC @ 2 kg/ plant+ AZB+PSB (T_9)	217.23	341.47	279.35	27	36	31.5	5.86	12.30	9.08
50% RDF + VC @ 4 kg/ plant+ AZB+PSB (T_{10})	261.27	440.04	350.65	16	24	20	4.17	10.56	7.37

SEm±	8.05	7.71	2.49	0.71	0.54	0.2	0.09	0.3	0.07
CD (p=0.05)	23.93	22.91	7.15	2.12	1.61	0.57	0.29	0.89	0.21

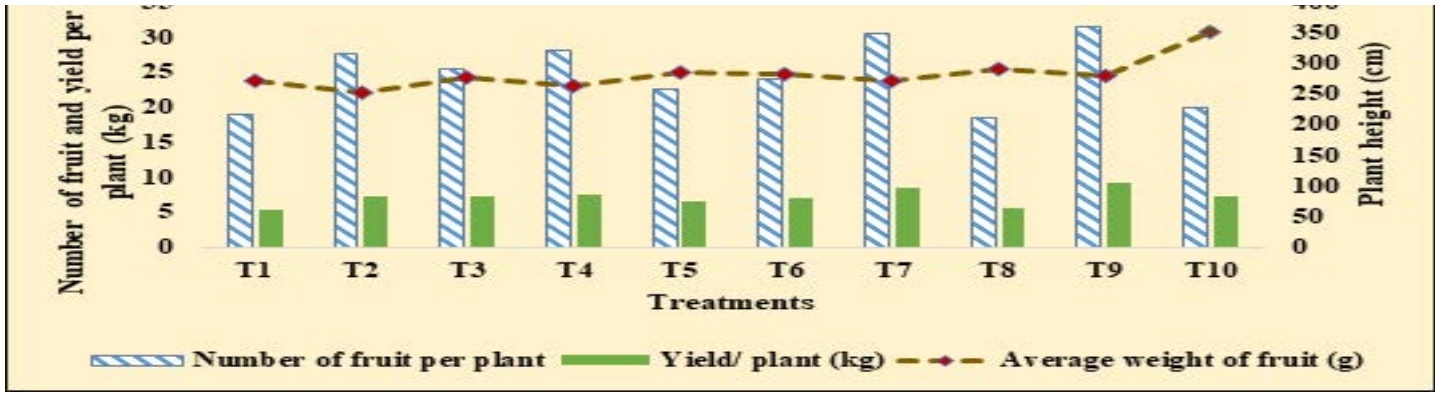

Fig. 1. Effect of INM on fruit traits and yield of custard apple (pooled data)

Conclusion

In conclusion, integrated nutrient management treatments significantly enhanced plant growth and yield parameters of custard apple. The treatment consisting of 75% RDF + vermicompost @ 2 kg/plant + AZB + PSB (T_9) consistently improved vegetative growth, including plant height, canopy spread, branch number, fruit count, and yield per plant. Additionally, the application of 50% RDF + vermicompost @ 4 kg/plant + AZB + PSB (T_{10}) resulted in the highest fruit weight. These findings underscore the effectiveness of combining reduced chemical fertilizers, organic inputs, and biofertilizers, reinforcing INM as a sustainable strategy to enhance custard apple yield.

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Conflict of Interest

The authors have no conflict of interest.

Data Sharing

All relevant data are within the manuscript.

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Short note

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A case study of crown bending disorder in date palm cv. Barhee and the impact of cyclone Biparjoy

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Introduction

Date palm (*Phoenix dactylifera*) is one of the oldest cultivated fruit crops in the world, and in India, it is believed to have been cultivated for more than four and a half centuries, where a significant area under cultivation is under the Kachchh district of Gujarat, which accounts for more than 85 % of the total area and production of the country (Muralidharan *et al.*, 2022). Unlike other countries, harvesting of the date fruits is mostly done in the *khalal* stage while most countries harvest at their *tamar* stage due to the climatic conditions (incidence of early rainfall). Among the various cultivars evaluated, cultivar Barhee was recommended by Date Palm Research Station, Mundra, under AICRP-Arid Zone Fruits for cultivation in Gujarat in 2002 (Sharma *et al.*, 2019), and since then Barhee has become the primary cultivar of the country, with a population of more than three lakh plants in Gujarat itself (data collected through personal communication).

Internationally, Barhee is a popular cultivar that initially originated in Iraq owing to its golden yellow-coloured fruits, round shape, prolific bearing, and non-astringent fruits at the *Khalal* stage. While, it is globally popular, it is uniquely susceptible to a disorder known as crown bending, also referred to as "Barhee disorder" due to its particular occurrence in this cultivar (Muralidharan *et al.*, 2019). This condition manifests as an abnormal bending of the palm's crown, typically towards the east or south-east.

The earlier reports of crown bending disorder of Barhee variety were from California (USA) by Darley *et al.* (1960), Al Basra (Iraq) by Hussain (1974), and at the Kibbutz Yotvata (Israel) by Zaid (1996). Affected palms were observed to bend primarily towards the south and occasionally towards the southwest. However, previous studies in the Kachchh region of India revealed that date palms that exhibited crown bending had a significantly higher number of bunches as well as distribution of bunches in the east when compared with date palms without bending disorder (Muralidharan, 2019). The proportion of bending is dependent on height and has been noted by Muralidharan *et al.* (2019). They noted that the presence of crown bending is observed in plants having a height of more than 5 m, and chances of bending increase with the

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increase in height. However, they also noted that the pattern was noted in the Barhee alone and not on other cultivars. The severity of the bending can be understood by the verge of bending, and we classified them as acute bending (< 90° bending with respect to the crown portion) and severe bending (>90° bending with respect to the crown portion). Similar observations were also done at Kibbutz Kineret in Israel, where this phenomenon was particularly severe, with the bending sometimes reaching angles as extreme as 90°.

In Israel, this bending disorder was also observed in the Dayri variety, and literature indicates that it affects the Jahla and Aguellid varieties as well (Djerbi, 1983). At Yotvata Kibbutz, growers have been addressing the issue by attaching a heavy iron bar to the side opposite the bending fruit bunches. The bunches are then tied to the bar to counterbalance the weight. It appears that within two to three years, this method successfully corrected the bending which suggests bunch handling has been found to be an effective solution (Yost, 1968). However, the factors driving crown bending remain poorly understood, based on earlier reports it suggesting that wind direction, height, and bunch weight distribution are possible contributing factors (Muralidharan, 2019). There is still no permanent solution for this disorder, emphasizing the need for further studies to understand its root causes and management.

Material and Methods

Based on the earlier suggestion by Darley *et al.* (1964) and earlier observations with the moderate crown bending disorder, a trial was initiated to straighten the plant terminal part (crown) in the year 2022 at Date Palm Research Station, Mundra-Kachchh, Gujarat, India. Due to limited availability of the similar bending plants, twelve plants of similar bending were identified and were treated with two treatments, replicated six times with one plant in each replication. The treatments were T_1 = The outer canopy leaves were pruned, terminal leaves were tied with rope and pulled in the opposite direction in the month of April and kept tied with the plant in the opposite direction of the bending (Fig. 1a, 2b), and T_2 = control. The direction of the bending was South-East for all the experimented plants. The comparison was made using a T-test. Their observations with respect to bending were noted after one year.

Results and Discussion

After one year of treatment, there were significant differences in bending between the treated and untreated palms (Table 1). The treated plants have largely recovered from the bending from 28.16° to 7.16° after a year (Fig. 1c), while in the untreated plants the bending were slowly rising (Fig. 1).

Table 1: Crown bending in treated and untreated plants

Treatment	Average angle of bending (Before treatment) (April, 2022)	Average angle of bending (one year after treatment) (April, 2023)	Average angle of bending (After cyclone) (June, 2023)
Treated crown	28.16° [a]	7.16° [b]	44.00° [b]
Untreated crown	26.83° [a]	28.66° [a]	144.16° [a]

Cyclone Biparjoy and its effects on date palm cv. Barhee

Cyclone Biparjoy, categorized as a "very severe cyclonic storm," impacted the Kachchh region on June 15–16, 2023, with wind speeds reaching up to 125 km/h, accompanied by heavy rainfall (Anonymous, 2023). After the cyclone their impact on the crown bending were recorded at the Date Palm Research Station in Mundra, situated close to the coastal area of the Gulf of Kachchh. The cyclone had a pronounced effect on Barhee palms, particularly those exhibiting crown bending disorder. Treated palms, which had initially shown a reduction in bending angle from an average of 28.16° to 7.16° between April 2022 and April 2023, exhibited renewed bending post-cyclone, albeit to a lesser degree (around 44°) (Fig. 1d). In contrast, untreated palms showed severe bending angles of over 90° (Fig. 2a), with some experiencing complete crown detachment or trunk breakage (Fig. 2b). As terminal leaves play a significant role in the palm trees as the new leaves emerge from there, separation of the terminal leaves destroys the meristematic part responsible for the regeneration of new leaves and ultimately leads to mortality (Fig. 2c). Among the various cultivars and germplasms, no other germplasms were showing any such bending symptoms. The presence of crown bending not only impacts the tree's structural integrity but also affects its overall health and productivity. Due to crown bending, it might be possible that there is a rise in the stress in the internal stress withing the trunk which potentially impeding nutrient and water transport, which may weaken the meristematic tissue at the crown, vital for new leaf growth and overall plants regenerative capacity. This may ultimately leads to mortality, especially severe bending leads to crown detachment. Such physiological implications underline the importance of addressing crown bending disorder in Barhee, both for enhancing plant survival and sustaining productivity.

Fig.1 (a) Date palm with crown bending disorder (July 2021) (b) Pruned date palm with tied terminal leaves (April 2022) (c) Treated date palm with a straight crown (April 2023) (d) Bending of the crown after cyclone (June 2023)

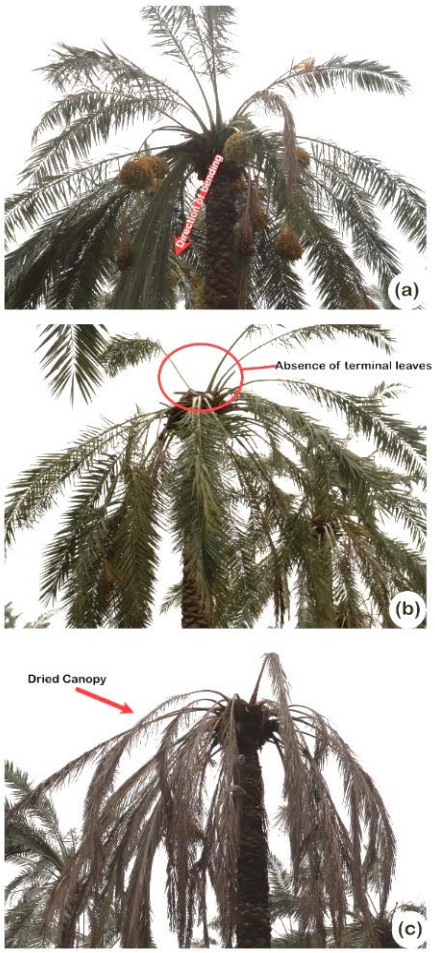

Fig. 2 (a) Untreated date palm with severe bending (b) Untreated date palm with broken crown (June, 2023) (c) Untreated date palm after one month of broken crown (July, 2023)

Conclusion and future directions

The impact of the cyclone highlights the vulnerability of the Barhee palms and its susceptibility to high wind conditions. The combined forces of heavy wind and imbalanced bunch weights suggests both environmental and physiological factors contributing to the disorder. This infers to the following conclusions: (i) crown bending is typical for the cultivar Barhee; (ii) it can be influenced by heavy wind; (iii) wind can influence and alter the direction and proportion of bending; (iv) excessive wind may result in breakage of terminal leaves and may result in death of the plant.

This case study illustrates the importance of understanding localized disorders and their interactions with environmental stressors, particularly in regions like Kachchh region of Gujarat where date palm cultivation plays a significant economic role. Exploring agronomic practices, such as enhanced support structures, regulated pruning, and targeted bunch management, may also offer practical solutions for mitigating bending severity. In addition, a more extensive study examining the biomechanical properties of crown bending across different heights and canopy structures could provide valuable data to inform cultivation strategies. Since, Barhee is a popular cultivar in the regions and is under extensive cultivation, developing practical solutions for crown bending is needful to support productivity and sustainability for growers across cyclone-prone areas.

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Conflict of Interest

The authors have no conflict of interest.

Data Sharing

All relevant data are within the manuscript.

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