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Review Article

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Advances in date palm (*Phoenix dactylifera* L.) research programme in Morocco: A review

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ABSTRACT

Date palm (*Phoenix dactylifera* L.) is a woody monocot tree producing fruits with high nutritional and economic importance. Currently, date production suffers from several biotic and abiotic stresses that reduce its profitability and menace its productivity. Moreover, climate change due to global warming has negatively affected crop production worldwide including dates. Despite the high resilience of the date palm tree, there is a need to develop new cultivars that are higher yielding and possess highly valued agronomic traits, such as fruit quality, disease and pest resistance and tolerance of extreme environmental conditions. In order to address this uncomfortable situation, Morocco has undertaken a huge research program for control of Bayoud disease basically, based on selection or creation of tolerant genotypes. To this end, several breeding approaches have been pursued, including: mass selection, cross breeding and induced mutagenesis. Obtained results have permitted to select many tolerant genotypes to Bayoud disease including cv. Nejda that has been largely propagated. In order to rehabilitate devastated palm groves with selected genotypes, plant tissue culture techniques were developed including organogenesis and somatic embryogenesis both from offshoot shoot tips or inflorescence plant material. Developed propagation technologies were transferred to private labs for mass propagation of date palm. Collaboration between public and private sectors has permitted production of three million vitro plants up to 2020 and a new plant production program is launched to produce five million plants in the horizon of 2030. Overall, the major research aspects of the national strategy to develop date palm sector in Morocco are presented.

Introduction

The date palm (*Phenix dactylifera* L.) plays a vital role in the ecology and economy of the arid and semi-arid regions. The total area of palm groves in Morocco is around 67 000 ha including 7.2 million palms, characterized by a high genetic diversity with

453 varieties and more than 2.7 million of natural hybrids. Average annual dates production is around 130000 MT for the last five years (Anonymous, 2025). However, Moroccan date palm sector suffers from many abiotic and biotic challenges particularly prolonged drought periods and Bayoud disease (*Fusarium oxysporum* f.sp. *albedinis*) (Foa). As direct consequences, Morocco is among the great importer of dates worldwide. Besides, there is a need of millions of date palm plants to rehabilitate the palm groves. To alleviate this uncomfortable situation, Morocco has undertaken a research program for control of Bayoud disease basically, based on selection or creation of tolerant genotypes and development of micropropagation techniques. Several breeding approaches have been pursued, including: mass selection, cross breeding and induced mutagenesis. In addition, the ongoing research program 2025-2028 is focused on

i. Selection, characterization, conservation and valorization of palm genetic resources
ii. Optimization of date palm production, protection and post-harvest techniques
iii. Evaluation of agro-ecological practices, improvement of organic production and innovative uses of dates and by-products of palm groves.

Mass selection

Plant breeding success depends on the collection and selection of germplasm, its evaluation for desired characters, multiplication, release and distribution of new varieties to farmers. Date palm is one of the oldest cultivated crops and up to now, most countries have followed conventional breeding methods (Al-Khayri *et al.*, 2018). The selection program conducted in Morocco aimed to find Bayoud resistant varieties and producing fruits of great commercial value. Taking into account that these two characters were never found together at the highest level in the most widespread cultivars, the first research works were focused on the selection of average quality dates in varieties with a high level of resistance then partial resistance for high fruit quality varieties (Saaidi, 1990). The selection first took place within common varieties, then among natural populations of individuals from natural hybridization. Such a program required a rigorous methodology, especially at the stage of the Bayoud resistance assessment. The main results of this research program was the selection of many tolerant genotypes particularly: Nejda, Deraouia, Sedrat, Al-Amal, Al-Fayda, Bourihane, Mabrouk and INRA-3010 genotypes. Moreover, two male cultivars: Nebch-Bouskri and Nebch-Boufeggous have been selected and characterized as resistant genotypes (Sedra, 2015). More details on genetic improvement to produce value-added date palm cultivars are available in a recent publication chapter of CABI (Abahmane, 2023).

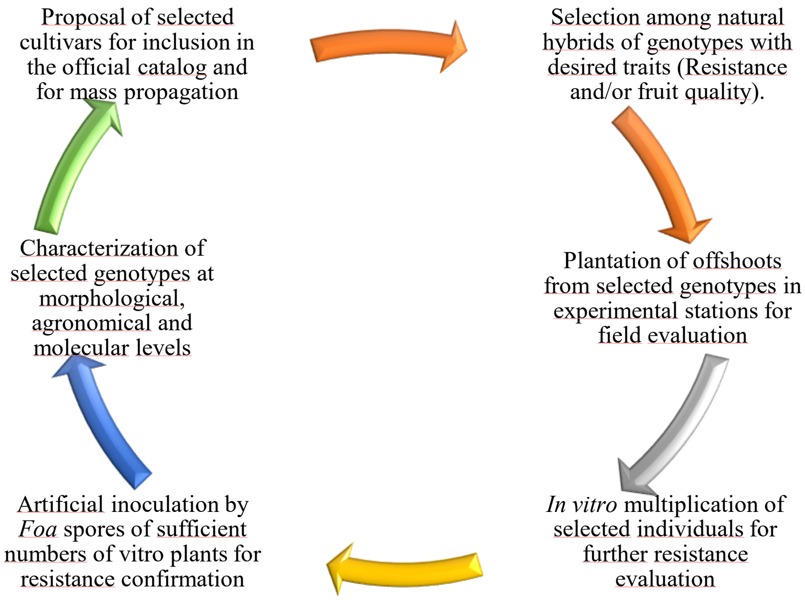

Fig. 1. Selection steps for Bayoud disease resistant genotypes implemented in Moroccan context (Sedra, 2011)

Cross breeding

Despite the great spontaneous diversity that can be found in landraces, simply applying selection to pre-existing diversity is an eroding process that eventually comes to a limit. The true creative power of plant breeding resides in promoting recombination for shuffling favourable alleles. The combination of different alleles in many loci results in a virtually infinite number of genotypes (Breseghello and

Coelho, 2013). The controlled hybridization program aimed creating palm trees resistant to Bayoud disease and producing dates of better quality than those resulting from natural hybridization. The descendants of controlled crosses were assessed for their resistance to Bayoud by artificial inoculation and by planting in naturally infested lands. The survivors were maintained until fruit set for examining the quality of dates (Sedra, 2011). As much as possible, hybridization programs were started using wide range of parents, especially palm trees with a high level of resistance and palm trees producing fruits of good quality. Male parents are selected based on their pollen production, timing of flowering, metaxenic effects and tolerance to biotic stresses (Saaidi, 1990). This program resulted in creation of 15 hybrids of good fruit quality and not showing symptoms of Bayoud disease. Although, cross breeding can make certain crosses between genotypes that can never occur naturally, this technique has some limitations. Indeed, it is time consuming, requiring 30 years to complete three backcrosses due to the palm's long-life cycle. Another constraint is gender identification, which is not possible before the onset of fruiting, 5-7 years after planting.

Induced mutagenesis

Natural genetic diversity is gradually eroded and consequently there is a loss of valuable genetic resources, which hinders genetic improvement of crops including date palm. Moreover, the rate of spontaneous mutations is extremely slow and makes the availability of wide genetic diversity more limited for plant breeders (Jain, 2010). Induced mutations hasten the rate of genetic diversity in a short period and are readily available to plant breeders for developing new cultivars. Mutation breeding can be described as the purposeful application of mutations in plant breeding (Carimi *et al.*, 2012). A mutation is a sudden heritable change in the DNA of a living cell that is not caused by genetic segregation or genetic recombination (van Harten, 1998). Mutations are generally induced by physical and chemical mutagenic treatments in seed and vegetatively propagated crops including the date palm. Gamma irradiation breaks DNA into small fragments and secondly the DNA starts a repair mechanism. During this second step, new variations develop or mutations occur. Mutation induction in date palm is feasible now due to a reliable plant regeneration system via somatic embryogenesis and organogenesis techniques. The optimal mutagen dose is determined empirically in order to minimize the impact on growth and fertility. Thus, induced mutations can provide useful alternative or complement to natural variation as well as to hybridization (Chakraborty and Paul, 2013). The undertaken research program in Morocco resulted in selection of 10 putative mutant plants that were later transferred to the "hot spot" in the field infested with Bayoud disease at the experimental station in Zagora. The performance of these mutant plants is under evaluation for other agronomic traits (Bougerfaoui *et al.*, 2006). Mutation breeding has many drawbacks. One issue is that large mutant populations must be generated due to the random nature of mutation induction. Theoretical calculations of the population size needed to find induced mutations in diploid plants suggest that tens of thousands of M1 lines are necessary (Roychowdhury and Tah, 2013). Producing and maintaining such numbers of line progenies can sometimes be impractical for species that are large or long-lived.

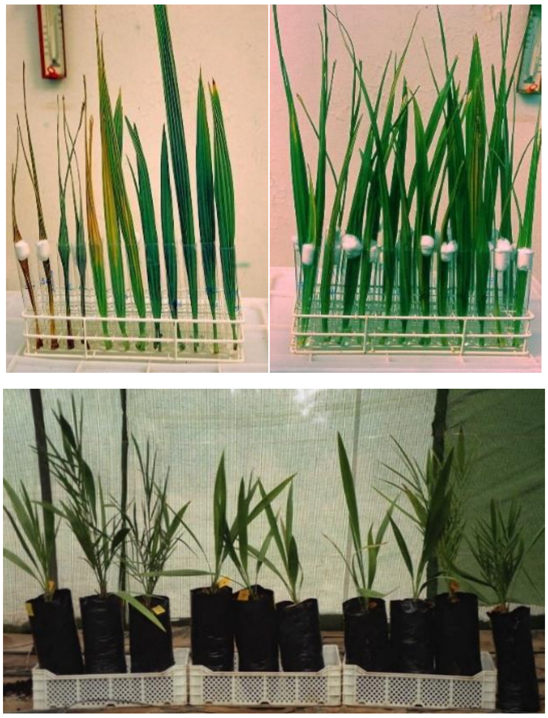

Fig. 2. Screening of plant material produced from irradiated Boufeggous variety (Gamma radiation at 20 gray) by using fungus toxins at 100 µg/ ml

Micropropagation techniques

Vegetative propagation by offshoots is the only conventional method to maintain genetic integrity of date palm cultivars. Due to its slow growth and limited number of offshoots, plant tissue culture (PTC) remains the ideal means for propagating and disseminating selected cultivars among oasis growers (Abahmane, 2011). The application of PTC techniques to date palm has many advantages: propagation of healthy selected female cultivars, large-scale multiplication, avoiding seasonal effects, production of genetically uniform plants, fast and secure exchange of plant material, and economical profitability. Looking at these advantages, micropropagation techniques have been developed for rapid mass propagation of date palm (Abahmane, 2017). In order to rehabilitate devastated palm groves with selected genotypes, organogenesis technique from both offshoot and inflorescence plant materials was used. Produced propagation technology was transferred to private labs for mass propagation of date palm.

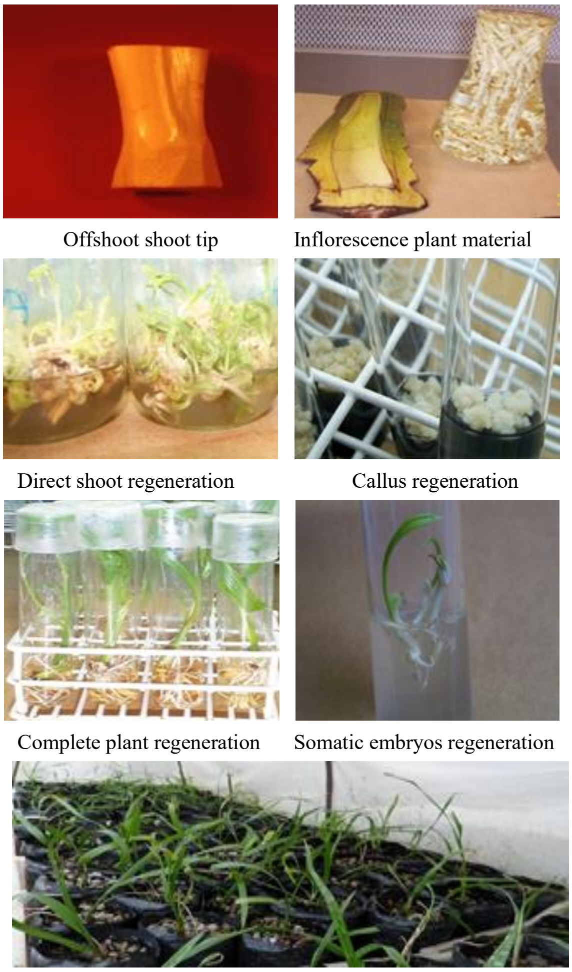

Fig. 3. Multiplication techniques for date palm micropropagation

Collaboration between public and private sectors has permitted production of three million vitro plants up to 2020. A new plant production program is launched to produce five million plants in the framework of Green Generation plan 2020-2030. Produced palm plant material is used for traditional palm groves rehabilitation as well as for creation of modern palm plantations.

Conclusion

Obtaining Plant material resistant to Bayoud disease through conventional breeding is a time-consuming and tedious process. There is enormous scope in the transformation technology to introduce disease-resistant traits in the date palm. The combined application of conventional breeding and applied biotechnological tools can generate genetic variations and produce fast propagating new genotypes with superior fruit quality, and resistance to biotic and abiotic stresses. Well-established plant tissue culture systems can be used for the rapid propagation of elite cultivars, to establish germplasm banks and for mutagenic studies *in vitro* to isolate useful mutants. Application of protoplast techniques can be utilized in somatic hybridization and introduction of desired genes through genetic transformation of date palm.

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Conflict of Interest

The author declares that there is no conflict of interest.

Data sharing

No data was used for the research described in the article.

Informed Consent

Not applicable.

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Review Article

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Applications of molecular markers in onion (*Allium cepa* L.) improvement - A review

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ABSTRACT

Molecular markers have emerged as indispensable tools in onion improvement, facilitating precise assessment of genetic diversity, trait mapping, and marker-assisted selection. Their application has accelerated the development of cultivars with desirable characteristics, including enhanced bulb quality, disease resistance, and stress tolerance. Molecular breeding has revolutionized onion improvement by providing reliable, environment-independent methods for germplasm characterization, genetic diversity analysis, and the construction of high-density genetic maps for gene tagging and selection. These markers are extensively employed in DNA fingerprinting for varietal identification, breeding line validation, and plant variety protection. They also play a crucial role in identifying and transferring resistance genes against biotic and abiotic stresses. Onion breeding has particularly benefited from marker-assisted improvement of traits such as male sterility, earliness, bulb color, yield, total soluble solids, and bolting resistance. Recent advancements have enabled the mapping of genes controlling bulbing and flowering, as well as loci associated with disease resistance and hybrid development. Overall, molecular marker technologies have greatly enhanced the efficiency of onion breeding, supporting the rapid development of elite, high-performing cultivars.

Introduction

Onion (*Allium cepa* L.), a diploid species (2n = 2x = 16) of the family Alliaceae, is one of the most extensively cultivated and consumed vegetables worldwide (Mohamed & Tawfik, 2025). Renowned for its culinary versatility, medicinal value, and pharmacological significance, onion ranks among the top five vegetable crops worldwide in both production and economic importance. Onion ranks as the second most important vegetable crop in India, cultivated over 17.40 lakh ha with an annual production of 30.21 million tons; however, its productivity (17.36 t/ha) remains below the global average (FAOSTAT, 2023). Believed to have originated in Central Asia, with the Mediterranean region serving as a secondary center of diversity (Gupta *et al.*, 2024a), onion cultivation sustains millions of smallholder farmers and contributes substantially to export earnings in many developing nations (Ahmed & Sallam, 2020).Onions are rich in bioactive constituents such as flavonoids, sulfur-containing compounds, and antioxidants that impart anti-inflammatory, antimicrobial, and anti-carcinogenic properties (Sagar *et al.*, 2022). In addition to their nutritional and health benefits, onions play a vital role in ensuring year-round food availability and enhancing dietary diversity across regions. However, productivity and quality are severely affected by biotic stresses including stemphylium blight (*Stemphylium vesicarium*), purple blotch (*Alternaria porri*), Fusarium basal rot (*Fusarium oxysporum* f. sp. *cepae*), and insect pests like thrips (*Thrips tabaci*) (Cramer, 2000; Gill *et al.*, 2015; Khar *et al.*, 2022a; Gupta *et al.*, 2024a; Gupta *et al.*, 2025). Additionally, abiotic challenges such as drought, salinity, and temperature extremes intensified by climate change further contribute to yield instability (Ratnarajah & Gnanachelvam, 2021). Postharvest issues, including sprouting and bulb decay, also cause considerable storage and market losses (Muhie, 2022).

While conventional breeding has improved onion performance, its progress is hindered by the crop's biennial growth cycle, cross-pollinated nature, and large genome (~16.4 Gb), all of which slow genetic gain (Havey, 2018). Moreover, complex inheritance patterns and strong genotype × environment interactions limit the efficiency of selection (Khosa *et al.*, 2016). Modern molecular tools such as marker-assisted selection (MAS), quantitative trait loci (QTL) mapping, and genome-wide association studies (GWAS) now provide more precise and rapid means to identify genes controlling disease resistance, bulb quality, and stress tolerance (Sharma *et al.*, 2024). Integrating these molecular approaches with conventional breeding offers a robust pathway towards accelerating genetic improvement and achieving sustainable onion productivity.

Onion genomics and molecular markers

Molecular markers have become indispensable tools in onion research, facilitating germplasm characterization, genetic diversity analysis, cultivar identification, phylogenetic studies, genome mapping, and gene tagging (Yang *et al.*, 2015). Commonly employed marker systems include restriction fragment length polymorphism (RFLP), random amplified polymorphic DNA (RAPD), inter-simple sequence repeats (ISSR), simple sequence repeats (SSR or microsatellites), amplified fragment length polymorphism (AFLP), sequence-characterized amplified regions (SCAR), sequence-related amplified polymorphism (SRAP), cleaved amplified polymorphic sequences (CAPS), and single nucleotide polymorphisms (SNPs) (Semagn *et al.*, 2006; Chinnappareddy *et al.*, 2013).

Onion possesses an exceptionally large genome (~16.4 Gb), composed predominantly (>95%) of repetitive elements, particularly long terminal repeat (LTR) retrotransposons (Pearce *et al.*, 1996; Kelly & Leitch, 2011). This high level of repetitiveness complicates accurate genome assembly, leading molecular breeding efforts to depend largely on linkage maps and marker-based approaches. The first molecular marker study in onion was conducted by De Vries *et al.* (1992), yet genomic resources for the crop remain less developed compared to many other species. While related crops such as garlic (16.2 Gb) and asparagus (1.1 Gb) have complete genome assemblies, onion comparative genomics remains limited due to reduced synteny (Harkess *et al.*, 2017; Sun *et al.*, 2020).

Recent technological advances, including RNA sequencing (RNA-seq), QTL mapping, and specialized databases like the Onion Genomic Resource (OGR), have enhanced gene discovery related to bulb development, stress tolerance, and male sterility (Shukla *et al.*, 2016; Sharma *et al.*, 2024). High-quality genome assemblies represent a major milestone: a 12.77 Gb chromosomal-level assembly comprising 65,730 annotated genes (Finkers *et al.*, 2021) and a 14.9 Gb assembly predicting 540,925 genes, of which 47,066 are supported by RNA-seq data (Cho *et al.*, 2025). These genomic resources are poised to accelerate functional genomics research and advance molecular breeding in onion.

Varietal identification and genetic diversity in onion

Molecular markers are widely used for onion varietal identification and diversity analysis due to their reproducibility and stability. Several marker systems, including RFLP, RAPD, AFLP, SSR, ISSR, SRAP, SNPs, and intron length polymorphic (ILPs), have been employed with varying levels of efficiency (Table 1).

RAPD markers have differentiated onion cultivars into distinct genetic clusters, though often showing weak correlation with phenotype (Tanikawa *et al.*, 2002; Sangeeta *et al.*, 2006). RAPD and ISSR analyses further grouped cultivars into major clusters, while SRAP markers revealed higher breeding utility (Sudha *et al.*, 2019; Mansour *et al.*, 2020). SSR markers have proven particularly effective, enabling fine-scale discrimination within accessions (McCallum *et al.*, 2008; Gupta *et al.*, 2020). Studies using EST-SSR and genomic SSRs reported high polymorphism, unique alleles, and strong genetic differentiation across accessions, useful for seed purity testing and breeding (Anandhan *et al.*, 2014;

Hanci & Gokce 2016; Lyngkhoi *et al.*, 2021). SNP-based approaches, derived from next generation sequencing (NGS) and ddRAD-seq, have provided reliable cultivar authentication and insights into domestication (Khar *et al.*, 2011; Lee *et al.*, 2018; Villano *et al.*, 2019).

More recently, ILP markers have been used for the genetic grouping of onion genotypes, assisting in the selection of parental lines for breeding (Khade *et al*., 2022; Gupta *et al.*, 2024b). SSR-based studies continue to reveal moderate diversity and additive gene action, supporting hybrid development strategies (Raj *et al.*, 2022). Additionally, analysis of the *atp6* gene highlighted phylogenetic clustering of *A. cepa* and *A. sativum*, with implications for male sterility restoration (Gowd *et al.*, 2023). Overall, molecular markers particularly SSRs and SNPs have greatly enhanced onion diversity analysis, varietal identification, and breeding efficiency.

Table 1. Applications of molecular markers in onion crop improvement

Markers	Applications
RFLP	Genetic diversity analysis, comparative mapping and synteny analysis, genetic mapping, hybrid confirmation
RAPD	Hybrid confirmation, genetic diversity analysis, bulk segregant analysis
SSR	DNA fingerprinting and genetic identity test, gene mapping, genetic diversity analysis, comparative mapping and synteny analysis, hybrid confirmation, MAS
ISSR	DNA fingerprinting and genetic identity test, genetic diversity analysis, hybrid confirmation
SNP	Genetic diversity analysis, genetic mapping, hybrid confirmation, MAS, genomic selection
AFLP	DNA fingerprinting, studying diversity and germplasm characterization

Genetic linkage mapping and QTL identification in onion

Genetic linkage maps are critical for marker-trait associations and marker-assisted selection (MAS). In onion, mapping has been constrained by its large (~16 Gb) genome and biennial nature (Jo *et al.,* 2017). Nevertheless, molecular markers have enabled the construction of saturated maps and the identification of QTLs for key traits.

Early work mapped ESTs using CAPs in the 'Brigham Yellow Globe' × 'Ailsa Craig' population, showing that SSCP/duplex analysis is a cost-effective approach for gene mapping (McCallum *et al.*, 2001). Initial linkage maps for *A. fistulosum* were developed using AFLP, SSR, and CAPS markers, covering 775-947 cM (Takayoshi *et al.*, 2005), while SSR-based maps from F_2 populations identified 17 linkage groups, later aligned with *A. cepa* chromosomes using alien addition lines (Tsukazaki *et al.*, 2008).

Trait mapping has revealed markers linked to purple blotch resistance (Chand *et al.*, 2018), QTLs for pungency associated with *SiR* and *ATPS* on chromosome 3 (McCallum *et al.*, 2007), and genomic regions controlling flower stalk height and heterosis (Li *et al.*, 2023). The latter study generated the most detailed map to date, comprising 10,584 SLAF markers across eight linkage groups (928.32 cM, 0.09 cM spacing), validated by localizing the male fertility (*Ms*) locus. Beyond trait mapping, genome-wide analyses have identified transcription factor families such as 24 GATA genes, implicated in development, hormone regulation, and stress tolerance (Bose *et al.*, 2025). These advances highlight the increasing resolution of onion maps and their growing utility in functional genomics and breeding applications.

Molecular markers in onion trait improvement

Molecular markers have significantly advanced onion breeding by enabling precise improvement of complex traits. Bulb color, determined by flavonoids such as anthocyanins, is controlled by major loci (I, C, G, L, L2, R) and genes like *DFR* and *ANS*, with functional markers aiding selection (Kim *et al*., 2005; Kim *et al.,* 2009a). A major QTL on chromosome 7 near *DFR* explains ~88% of anthocyanin variation (Choi *et al.*, 2020). Regulatory factors, including *AcB2* (C locus) and *AcMYB1*, as well as allelic variations in *ANS* and novel genes such as *AcLAC12*, further shape pigmentation (Zhang *et al.*, 2018; Kim *et al.,* 2017; Han *et al*., 2023).

For yield traits, SNPs linked to bulb weight have been identified, with machine learning achieving >80% prediction accuracy, while bulb shape variation is largely explained by PC1 (height-to-diameter ratio) and a QTL on chromosome 5 (Sekine *et al*., 2023; Choi *et al*., 2024). Bulb quality traits such as soluble solids, sugars, fructans, and pungency have also been mapped using SSRs, ILPs, and NGS tools. QTLs for sugar metabolism and sulfur-related genes (*SiR, ATPS*) regulate sweetness and pungency (McCallum *et al*., 2007; Havey *et al*., 2004), while *LFS* on chromosome 5 is a key determinant of lachrymatory factor (Masamura *et al*., 2012). High-TSS genotypes identified using SSRs and ILPs support breeding for processing quality (Singh *et al*., 2021; Mahajan *et al*., 2025).

Bolting in onion is characterized by high photosynthesis, low respiration, and carbohydrate accumulation, which induces flowering hormones such as vernalin or florigen, promoting flower bud formation and shortening the bulb's life cycle (Gupta *et al*., 2015). Bulbing and flowering responses, regulated by photoperiod and vernalization, are controlled by *FT* homologs (*AcFT1*, *AcFT4*, *AcFT2*), with QTLs associated with bolting and bulbing traits (Lee *et al*., 2013; Marediya, 2020; Havey, 2024).

Collectively, structural, regulatory, and QTL-based markers for bulb color, yield, quality, bulbing, and flowering constitute a powerful toolkit for accelerating onion breeding programs (Fig. 1).

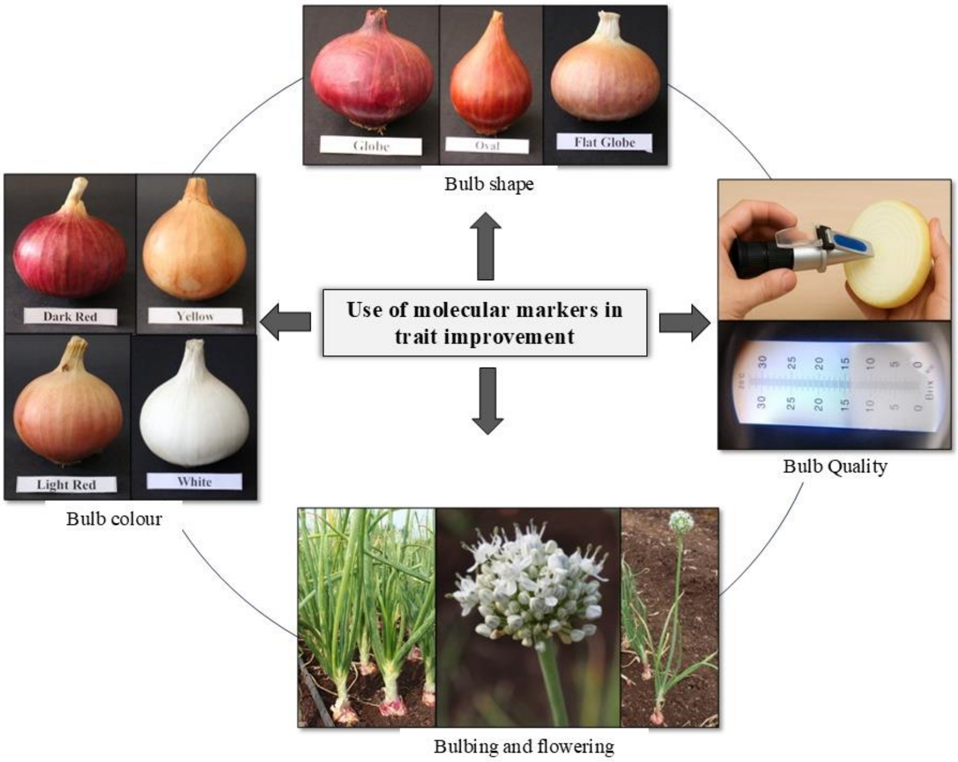

Fig. 1. Use of molecular markers in onion trait improvement

Molecular markers for male sterility and hybrid breeding in onion

Male sterility plays a crucial role in hybrid seed production in onion, and the use of molecular markers has significantly advanced the detection of different cytoplasmic types and identification of maintainer lines (Table 2). Early investigations differentiated normal (N) and sterile (S, T) cytoplasms through PCR-based and cytoplasm-specific markers (Havey, 1995; Engelke *et al*., 2003). The subsequent discovery of the chimeric mitochondrial gene *orf725* present in CMS-S and CMS-T lines but absent in N cytoplasm facilitated the development of reliable PCR markers for cytoplasm classification (Kim *et al*., 2009b).

Markers linked to the nuclear *Ms* locus are equally important for detecting maintainer genotypes. RAPD, CAPS and SSR-derived markers such as jnurf13, jnurf05 and ACms.1100 showed close linkage with *Ms* (Bang *et al*., 2013; Park *et al*., 2013; Kim & Kim 2015), allowing early identification of maintainers and elimination of fertile contaminants. However, marker reliability varies across germplasm, highlighting the need for additional validation (Khar & Saini 2016).

Population studies have revealed high frequencies of S cytoplasm in Indian germplasm (Saini *et al*., 2015), while authentic T cytoplasm remains rare in commercial programs (Havey & Kim 2021). Gupta *et al*. (2016) identified two naturally occurring male-sterile lines from red onions (RGP-4-Sel and Arka Kalyan), and Khar *et al*. (2022) reported five S-cytoplasm sterile plants out of eight in Arka Kalyan. However, these lines still need full characterization to clarify their S or T cytoplasmic status. Khar *et al*. (2022b) identified T cytoplasm in Indian accessions for the first time, expanding resources for hybrid development.

Chloroplast and mitochondrial polymorphism analyses (Von *et al*., 2013), together with nuclear-cytoplasmic gene expression studies (Yuan *et al*., 2018), have provided deeper insights into the mechanisms of cytoplasmic male sterility (CMS). Structural rearrangements in mitochondrial genomes that generate novel ORFs are key contributors to CMS diversity (Hanson & Bentolila, 2004). The advent of PCR-based assays has enabled rapid and precise identification of sterile (A), maintainer (B), and restorer (R) lines, significantly shortening breeding cycles from years to months compared with traditional progeny testing (Satoh *et al*., 1993; Havey, 2000). Moreover, the utilization of alternative CMS sources from *A. galanthum* and *A. roylei* has expanded the genetic base available for hybrid onion breeding (Havey, 1999; Vu *et al*., 2011).

Overall, CMS- and *Ms*-linked markers have transformed onion hybrid breeding, ensuring efficient line development, genetic purity, and improved hybrid seed productivity.

Table 2. Molecular markers linked to male sterility and fertility restoration in onion

Cytoplasm/ *MS* locus trait	Marker/ gene	References
CMS-S	AcMSH1, 5'cob, orfA501, orf725, orfA501	Mainkar *et al*., 2023; Saini *et al*., 2015; Abbasi, 2023
CMS-S & CMS-T	Orf725	Kim *et al*., 2009b
N Cytoplasm	IGS insertion marker orfA501, orf725	Abbasi, 2023
Cytoplasm - T	accD & MKFR	Khar *et al*., 2022b
Ms locus (fertility restoration)	jnurf13 & jnurf610, jnurf12 & jnurf13	Kim & Kim 2015; Kim, 2014
Ms locus	AcPMS1 & AcSKP1	Khar *et al*., 2022b; Kim *et al*., 2015; Huo *et al*., 2015

Molecular markers in biotic stress management

A significant yield gap is attributed to foliar, root, and bulb diseases that impair plant health and postharvest storability (Cramer, 2000). Consequently, breeding for disease resistance has become a major priority, with molecular markers emerging as valuable tools for tagging resistance genes and enabling marker-assisted selection (MAS) (Ashkani *et al*., 2015). Although progress in onion remains relatively modest (Scholten *et al*., 2007), recent advances underscore the growing potential of molecular approaches.

For purple blotch (*Alternaria porri*), mapping populations derived from ‘Arka Kalyan’ × ‘Agrifound Rose’ identified the SSR marker *AcSSR7* and STS marker *ApR-450* linked to *ApR1*, facilitating MAS (Chand *et al*., 2018). In the case of downy mildew (*Peronospora destructor*), the *DMR1* marker was associated with an *A. roylei*-derived resistance fragment, enabling marker-assisted backcrossing (Kim *et al*., 2016). Similarly, for gray mold (*Botrytis* spp.), RAPD-derived *SCAR-OPAN1* and *SNP-3 HRM* markers were developed to screen resistant genotypes (Kim *et al.,* 2021). Transcriptomic analyses further revealed stronger defense-related gene expression in *A. cepa* genotypes resistant to *A. porri*, while *Allium hookeri* exhibited resistance to *Thrips tabaci* through elevated lectin activity, which reduced pest damage and survival (Khandagale *et al*., 2022).

Resistance to Fusarium basal rot was identified in short-day varieties ‘Saba’ and ‘Saba-HS,’ characterized by upregulation of *Lectin*, *LOX*, and *Osmotin* genes (Poursakhi *et al*., 2024). ITS sequencing and PCR-based diagnostics have also aided the detection of onion twister disease (Patil *et al*., 2016). Pathogenicity assays revealed that *Colletotrichum gloeosporioides* induces twisting symptoms, whereas *Fusarium acutatum* causes neck elongation; co-infection along with GA_3 application intensifies symptom expression, offering insights for molecular marker based resistance breeding (Dutta *et al*., 2024).

In addition, gut microbiome profiling of *Thrips tabaci* populations from various Indian regions revealed diverse bacterial communities showing strong geographic variation, indicating potential for identifying microbial or host-associated molecular markers relevant to pest resistance (Gawande *et al*., 2019). Despite onion’s large genome (~16 Gb) and biennial growth cycle, these studies collectively highlight the expanding role of molecular tools in developing cultivars with enhanced resistance to diseases and insect pests (Khar *et al.,* 2022a; Finkers *et al*., 2021).

Molecular markers in abiotic stress management

Abiotic stresses such as drought, salinity, waterlogging, and heat pose major limitations to onion productivity. Molecular research has led to the identification of key stress-responsive genes and tolerant germplasm, supporting the development of resilient cultivars through molecular breeding.

For drought tolerance, *NAC* transcription factors (*CepNAC-IV*, *CepNAC-V*) play central roles in regulating stress adaptation (Gokce *et al.*, 2022), while the *1-SST* gene involved in fructan biosynthesis contributes to enhanced drought resilience (Liu *et al.*, 2022). Genotypes U12, U47, U49, and cultivar K52 have shown combined tolerance to both drought and salinity (Mahajan & Gupta, 2023). Salinity tolerance has been linked to aquaporin genes *AcPIP1* and *AcTIP2*, which maintain water transport and osmotic balance under saline conditions (Sanwal *et al.*, 2022; Solouki *et al.*, 2023).

Waterlogging tolerance is mediated by genes associated with anaerobic respiration (*ADH*, *PDC*) and the stress-responsive transcription factor *WRKY22*, which together sustain energy metabolism under oxygen-deficient conditions (Gedam *et al.*, 2023). In response to high-temperature stress, upregulation of heat shock protein (HSP) families (*HSP40*, *HSP60*, *HSP70*, *HSP90*, *HSP101*) and transcriptional regulation by *HSFA1* provide protection against protein denaturation (Galsurker *et al.*, 2018). Collectively, the use of molecular markers for identifying stress-responsive genes is accelerating the breeding of climate-resilient onion varieties capable of withstanding multiple abiotic challenges.

Conclusion

Molecular markers represent a transformative advancement in onion breeding, offering precise, efficient, and environment-independent tools to unravel complex traits and accelerate genetic improvement. Their applications extend across varietal identification, genetic diversity analysis, trait linkage, QTL mapping, and the characterization of cytoplasmic types for hybrid development. They have proven invaluable in improving traits such as bulb color, shape, TSS, pungency, flowering, and stress tolerance, while also facilitating the identification of male-sterile, maintainer, and restorer lines for hybrid breeding. Unlike conventional morphological markers, DNA-based markers provide reproducible and high-throughput insights, enabling more effective selection strategies. Advances in genomics, bioinformatics, and molecular technologies promise the development of superior onion cultivars with enhanced yield, resilience, and quality. This article underscores the pivotal role of molecular markers in modern onion breeding and highlights their potential to address global challenges in food security, sustainability, and climate adaptation.

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Author's contributions

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Conflict of Interest

The authors have no conflict of interest.

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Data sharing

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Informed Consent

All the authors agree with the content of this article.

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Review Article

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Medicinal herbs cultivation to ensure a sustainable supply of quality raw herbal drugs: A review

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ABSTRACT

Geographical Indications (GIs) are closely associated with product quality, as the agroecological conditions of a region impart unique characteristics to agricultural commodities. These distinctive quality traits facilitate the filing and registration of GI products. India, being one of the world's mega-biodiversity regions, possesses a rich diversity of medicinal plants. However, the increasing demand for herbal medicines has led to destructive harvesting practices, threatening the natural populations of these valuable species. Therefore, the protection and promotion of GIs can play a significant role in branding and marketing region-specific products, thereby enhancing the economic prosperity of local producers and manufacturers. In addition to supporting regional economic development, GI registration also strengthens export potential. Among the GI-tagged horticultural commodities, spices, medicinal, and aromatic crops together constitute less than 25% of the total share. Although GI registration in the horticulture sector has progressed considerably over the years, medicinal and aromatic crops still possess very few GI tags, highlighting the urgent need for greater awareness, documentation, and registration efforts in this sector.

Introduction

Raw herbal drugs are typically associated with the region where they are produced and are influenced by distinct local geographical conditions. The product's attributes, traits or reputation should primarily stem from its origin or roots. The growing consumer awareness regarding drug quality has prompted extensive scholarly investigation into the multidimensional nature of quality (Ilbery & Kneafsey, 2000). Consequently, more stringent regulatory frameworks have been implemented to mitigate the risks associated with food consumption and to safeguard public health (De Rosa, 2015). To address this issue, the World Trade Organization (WTO) has established various forms of Intellectual Property Rights (IPRs) aimed at supporting local producers of agricultural commodities and ensuring the provision of high-quality products, and one of them is Geographical Indication (GI). A geographical indication is any indication that has a good originating in a region or locality, where a particular quality, reputation, or other characteristic of the good is primarily attributable to its geographical origin

(WIPO, 2024). Since the adoption of the Agreement on Trade-Related Aspects of Intellectual Property Rights (the TRIPS Agreement) in 1994, which contains a section on geographical indications, this form of intellectual property (IP) has attracted increasing attention from policymakers and trade negotiators, as well as producers, lawyers, and economists across the world (European Commission WIPO, 2024).

The focus of GI protections has historically been on agricultural or food products, rather than non-edible items such as medicinal plants. Intellectual Property Rights (IPR) related to traditional medicinal plants did not attract significant attention until 1994-1995, when the United States Patent and Trademark Office granted a patent for turmeric due to its wound-healing properties. Around the same time, in 1994, the European Patent Office awarded a patent for Neem (*Azadirachta indica*) for its antifungal applications. Both patents were later revoked following legal objections from India. In response to such instances of biopiracy, the Indian government launched the Traditional Knowledge Digital Library (TKDL) in 2001. This initiative aimed to document and digitize traditional medicinal formulations from Indian systems of medicine, making them globally accessible to safeguard against unauthorized intellectual property claims (James, 2016; Pandit *et al.,* 2024). Following this, the Geographical Indication (GI) tag system in India was officially launched with the enactment of the Geographical Indications of Goods (Registration and Protection) Act, 1999, which came into effect on 15th September 2003 (PIB, 2003). This legislation provides legal protection to products uniquely linked to specific geographical locations, recognizing their distinct qualities and reputation. According to the 2023 World Intellectual Property Indicators (WIPO) report, approximately 58,400 GIs were registered and in force across 91 countries and regions globally. China reported the highest number of active GIs (9,785), followed by Germany (7,586), Italy (6,330), and France (6,098). In 2023, goods and services related to wines and spirits constituted the largest category, comprising 48.1% of the total, while agricultural products and foodstuffs accounted for 44.8%, and handicrafts represented 4.2%.

As of 2024, Asia recorded 3,973 registered GIs, of which 658 originated from India. Indian GIs, 203 were classified under the category of agricultural products (WIPO, 2024; FAO, 2025). Darjeeling Tea was the first Indian product to receive a Geographical Indication (GI) tag in 2004-2005. Since then, India has registered over 658 GI-tagged products across diverse categories, including agricultural commodities, handicrafts, textiles, and food items. However, the number of GI registrations pertaining specifically to medicinal and aromatic crop products remains limited, with only approximately 11 entries. This number further decreases when considering exclusively botanically derived substances used for therapeutic applications (Sachin *et al.*, 2023; Kadasani, 2023). Several factors contribute to this, including limited documentation and scientific validation of traditional knowledge related to medicinal plants, which makes it challenging to establish a clear link between the plant and its geographic origin. Along with weak enforcement mechanisms, insufficient institutional support further contributes to the neglect of GI for medicinal plants in national and international discourse.

Cultivating medicinal plants in ecologically suitable regions is vital for optimizing their phytochemical composition, therapeutic efficacy and long-term sustainability. Environmental variables such as climate, soil type, altitude, and indigenous farming practices significantly influence the biosynthesis of active constituents in these plants. When grown in their native or ecologically optimal zones, medicinal plants tend to exhibit superior yields and higher concentrations of bioactive compounds, e.g., Nagori Ashwagandha, Maghai Paan, Sojat Heena, Kashmiri Kesar, Ganjam Kewda, etc. Additionally, region-specific cultivation plays a key role in conserving local biodiversity, safeguarding traditional ethnobotanical knowledge, and improving the socio-economic conditions of rural communities (Kala *et al.,* 2006; Sharma *et al.,* 2020). Therefore, GI of medicinal plants is essential for preserving traditional knowledge, protecting biodiversity, promoting sustainable livelihoods, and supplying quality raw herbal drugs.

Table 1. List of medicinal plants filed for GI application

Name and Application No.	Crop	Region	Unique traits	Status
Nagori Ashwagandha and 1143.	*Withania somnifera* (Linn.) Dunal	Arid track of central to western Rajasthan.	Pencil thickness, brittle, dry root without any cavity, and having ≥35% starch content in roots.	Approved

Tanner's Senna of Kutch (Kutchi Aval) and 1528.	*Senna auriculata* L. Roxb	Kutch district of Gujarat.	Tannin content and antioxidant activity in bark, leaf, and pod harvested from the Kutch region	Filed
Southeast West Bengal Brahmi and 1391.	*Bacopa monnieri (L.) Wettst.*	North 24 Parganas, Medinipur, Nadia, Hooghly, Kolkata, Howrah, Sagar Islands and the nearby wetland areas of West Bengal	Rich in Bacoside A content and maximum herbage yield	Filed
West Bengal Thankuni (Gotu Kola) and 2017	*Centella asiatica*	West Bengal districts such as Darjeeling, Dinajpur, Malda, Murshidabad, Burdwan, Hooghly and Kolkata	Small-leaved (5.42-7.16 cm^2) variant rich in bioactive secondary metabolites such as Asiaticoside (4.14%) and Madecassoside (2.32%).	Filed
'Kangan Saji of Ghaggar and 1617	*Haloxylon stocksii* (Boiss.)	Ghaggar belt of Rajasthan, especially in Vijaynagar, Anupgarh and Suratgarh Tehsils of Sri Ganganagar as well as some parts of Hanumangarh and Bikaner districts of Rajasthan	Choa, Rota, and Saji salts are rich in carbonate (350-355 g/ kg), bicarbonate (75-80 g/ kg), sodium (324-328 g/ kg), and chloride (220-228 g/ kg).	Filed

Nagori Ashwagandha

The cultivation of *Withania somnifera* (Ashwagandha) is concentrated extensively in the western and central regions of Rajasthan, notably in the former Nagaur district and adjoining areas, including Bikaner, Churu, Barmer, Sikar, and Jodhpur (Bikaner District Gazetteer, 1962). Among the region-specific 'Nagori Ashwagandha' is internationally recognized due to its adaptation to the unique phytogeographic conditions of this semi-arid zone. This landrace exhibits distinct root characteristics, such as root length, root thickness, root brittleness, and higher starch content in the root, even at physiological maturity, under sandy to loamy sandy soils (Saran *et al.,* 2025). The genotype-environment interaction in this region significantly influenced root yield and phytochemical composition. Notably, roots of the variety Vallabh Shahi Ashwagandha-1 (VSA-1) selected from Nagori Ashwagandha contains thicker and more brittle roots as compared to those of other varieties.

In addition, the Nagori Ashwagandha demonstrates superior root morphology, including fresh root girth (4.17 cm), dry root girth (1.63 cm), root yield (5 t/ ha), starch yield (2.70 t/ ha) and a brittleness score of 9.33 (on a 10-point scale) due to minimum crude fiber (7.55-8.31%) content (Fig. 1). The minimum qualitative benchmark for Nagori Ashwagandha includes pencil-thick, cavity-free, brittle dry roots with a starch content of ≥35% at crop maturity (Saran *et al.,* 2025). These traits underscore the importance of region-specific cultivation in enhancing both the quality and pharmacological potential of ashwagandha.

Southeast West Bengal Brahmi

The Southeast West Bengal Brahmi ecotype is predominantly distributed in the southeastern region of West Bengal, particularly within the North 24 Parganas district and adjacent wetland areas (Hooker, 1890; Chowdhury and Mukherjee, 2015; Pattanayak *et al*., 2016; Mukherjee, 2018; Naskar *et al*., 2022). This elite germplasm of *Bacopa monnieri* (Linn.) Dunal is characterized by the longest pedicels (Fig. 2), its high saponin content, especially Bacoside A (3.65%) and its major constituent bacoside-A3 (0.679%), which are the primary bioactive compounds contributing to its pharmacological efficacy (Saran *et al.,* 2022).

Among Brahmi accessions collected from various geographic regions across India, those from the wetlands of southeast West Bengal consistently exhibit the highest bacoside accumulation, with Bacoside A content reaching up to 7.55 µg, surpassing levels observed in accessions from other parts of the country (Dey *et al.,* 2020).

The DBM-4 genotype from wetlands of south-eastern West Bengal yielded the highest dry herbage at 134 q/ ha, representing a 9% increase over the check variety (124 q/ ha). In terms of morphological characteristics, along with superior pedicel length, averaging 2.96 cm, which is about 19% longer than the check variety's pedicel length of 2.49 cm (Saran *et al.,* 2022). Additionally, the Brahmi variety Vallabhi Ganga Brahmi-1, developed through selective breeding from germplasm collected in the wetlands of south-eastern West Bengal, has demonstrated significantly higher total bacoside yield, reaching 181 kg/ ha, approximately 58% greater than the standard check variety CIM-Jagriti, which yields 115 kg/ ha.

The genotype demonstrates exceptional performance in terms of both biomass production and phytochemical yield when cultivated in the wetland agro-ecology of this region. This variety is particularly suited for commercial cultivation due to its high dry herbage yield potential (up to 170 q/ ha) and elevated Bacoside A content, meeting and exceeding industrial quality standards (≥1.5%) (Fig. 3).

Fig. 1. Roots of Nagori Ashwagandha

Fig. 2. Brahmi germplasm DBM-4 vs CIM Jagriti (Check)

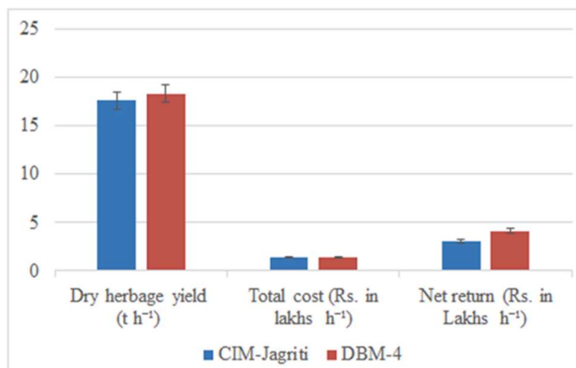

Fig. 3. Economic performance of Brahmi (DBM-4 and CIM Jagriti)

Tanner's Senna of Kutch (Kutchi Aval)

Tanner's Senna of Kutch (Kutchi Aval), scientifically known as *Senna auriculata,* is commonly found across the coastal and semi-arid regions of Kutch, Gujarat (Kutch District Gazetteers, 1971). It is a branched shrub with a 1.5-5 m height, trunk diameter of 20 cm, and brown lenticellate bark. The fruit of this Senna is visibly distinct in shape and size from other species of Senna (Fig. 4). Its distinctiveness and commercial importance stem from its high tannin content, primarily extracted from the bark. The flowers of *S. auriculata* (L.) Roxb. have been reported to contain a wide range of bioactive compounds, including alkaloids, glycosides, saponins, polyphenols, tannins, phlorotannins, terpenoids, triterpenes, carbohydrates, proteins, amino acids, anthraquinones, aloe-emodin, and sitosterols. The plant's leaves and stems are commonly used as alternatives to *S. angustifolia* (Tinnevelly Senna), especially for extracting sennosides and in hair dye formulations with henna. These phytoconstituents are linked to therapeutic effects, notably in managing diabetes mellitus and other health conditions (Prasathkumar *et al.,* 2021; Nille *et al.,* 2024).

Fig. 4. Plant parts of Tanner's Senna of Kutch (Kutchi Aval)

Kangan Saji of Ghaggar

The 'Kangan Saji of Ghaggar' (*Haloxylon stocksii)* is a widely distributed, cultivated plant in the Ghaggar belt of Rajasthan, especially in Vijaynagar, Anupgarh, and Suratgarh Tehsils of Sri Ganganagar (Sri Ganganagar district Gazetteer, 1972; Hedge, 1997), as well as some parts of Hanumangarh and Bikaner districts of Rajasthan—the 'Kangan Saji of Ghaggar' dwells differently from others in climate and edaphic characteristics. This species is characterized by its ability to accumulate high levels of sodium and chloride ions within leaf vacuoles. The plant features glabrous stems and ellipsoid, trigonous leaves that are obtuse or acute at the tip, measuring 0.3 to 0.8 cm in length (Rathore *et al*., 2012). Its primary use lies in the production of 'Saji'-also known as barilla or soda ash through the combustion of air-dried plant material. The Saji, derived from *H. stocksii* grown along the Ghaggar riverbed, is regarded as superior in quality. Three main grades of Saji salt are traditionally recognized: Choa, Intermediate (rota) and Saji. Choa salt, noted for its porous, ashy texture and white to purplish colour, is considered the highest quality, while the standard Saji is a denser, darker, stone-like material. The quality of Saji salt depends significantly on the skill of the local artisans during the burning and processing stages. The Bawaria community of Rajasthan continues to maintain this traditional practice. Additionally, factors such as soil type, plant variety, shrub age, and agronomic practices influence the final quality of the product. (Ahmed *et al.,* 2006).

West Bengal Thankuni (Gotu Kola)

The West Bengal Thankuni, commonly known as Mandukarpani in India, Gotu Kola worldwide, Golpatta, Paisa Jhar, Athane Jhar in the Darjeeling area and Dholamonia in the Dinajpur area of West Bengal (Subba *et al*., 2024), is an important medicinal herb that is used for its neuroprotective, immunomodulatory, anti-cancer, antimicrobial, antioxidative, and nerve-regenerative properties (Sabaragamuwa *et al*., 2018). In West Bengal, particularly, this plant often grows naturally as a weed in agricultural fields or on wastelands and is known as *Centella asiatica* (syn., *Hydrocotyle asiatica*) and *H. verticillata* (Hooker, 1886). Many people have cultivated it in their home gardens, either for its significant medicinal properties or for using its leaves as a leafy vegetable (Mondal and Khatua, 2015).

The bioactive compounds present in this plant are referred to as centellosides, consisting of two saponins (asiaticoside and madecassoside) and two sapogenins (asiatic acid and madecassic acid) (Nav *et al.*, 2021). These bioactive constituents are present in all plant parts, with leaves serving as the primary site of accumulation. Their quantitative profiles are strongly modulated by ecological factors and the geographical provenance of the plant material (Singh *et al*., 2022). Consequently, the selection and large-scale cultivation of elite, high-metabolite–yielding chemotypes are crucial for enhancing both agricultural productivity and industrial applications. The West Bengal Thankuni (Gotu Kola) small-leaved (5.42-7.16 cm^2) variant is rich in bioactive secondary metabolites such as Asiaticoside (4.14%) and Madecassoside (2.32%), which fit the industrial benchmarks. In contrast, the broad-leaved (21.92-33.15 cm^2) variant from Gujarat is rich in Madecassic acid and Asiatic acid. The Thankuni has been reported to contain the highest asiaticoside content (1.51–4.20%) among collections from different agro-ecological regions of India, with the maximum recorded in an accession from the Kolkata region (Rohini and Smitha, 2022).

Table 2. Secondary metabolite content in *Centella asiatica* from different phytogeographic regions

Location	Asiaticoside (%)	Madecassoside (%)	Madecassic acid (%)	Asiatic acid (%)	Total Triterpenes (%)
West Bengal	4.138	2.32	0.062	0.030	6.550
Nagaland (NEH)	1.511	0.86	0.169	0.068	2.608
Gujarat	0.543	1.987	0.260	0.194	2.984

The overall concentration of these secondary metabolites in West Bengal Thankuni (Gotu Kola) from West Bengal significantly exceeds that reported in the related species *Hydrocotyle verticillata*, underscoring its distinct phytochemical superiority and promising therapeutic potential for addressing emerging health disorders.

Conclusion

This review highlights the critical role of Geographical Indications (GIs) in recognizing, preserving, and promoting the unique identity and traditional knowledge associated with medicinal and aromatic plants (MAPs) in India. Despite historically limited attention to non-edible products under GI frameworks, this study showcases four distinct and region-specific medicinal plants: Nagori Ashwagandha, Southeast West Bengal Brahmi, West Bengal Thankuni (Gotu Kola) Tanner's, Senna of Kutch (Kutchi Aval), and Kangan Saji of Ghaggar, which demonstrate clear geographic and phytochemical distinctiveness. These landraces not only exhibit superior quality attributes and bioactive profiles but are also integral to local economies, sustainable livelihoods and biodiversity conservation. Documenting and validating the unique geographical and quality-linked characteristics of these resources strengthens the case for GI registration and supports the broader objectives of sustainable development, rural empowerment and global competitiveness of Indian herbal products. Promoting GIs in the MAPs sector can help ensure quality assurance, traceability and market differentiation, while also protecting indigenous knowledge systems. As consumer demand for authentic and region-specific herbal products continues to rise, establishing strong GI protection for medicinal plants becomes increasingly imperative for safeguarding India's rich phyto-pharmaceutical heritage.

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Author's Contribution

Parmeshwar Lal Saran, K.A. Kalariya, Rohan Sarkar, and Manish Kumar Mittal served as PI and Co-PIs of the project. They contributed to the GI filing process, conducted data collection and analysis, and drafted the review manuscript. Mrs. Manjari was also involved in sampling and data compilation as a Research Fellow under the GI Project.

Conflict of Interest

The author has no conflict of interest.

Data Sharing

This article did not generate any supplementary data.

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Research Article

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Influence of plant spacing on yield and yield components of improved onion varieties in Mersa District, North Wollo, Ethiopia

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ABSTRACT

This study evaluated the effects of plant spacing and variety on growth and yield of onion in Mersa District, North Wollo, Ethiopia. Treatments consisted of a factorial combination of three improved varieties and six planting spacing arranged in a randomized complete block design with three replications. Analysis of variance revealed that the interaction of spacing and variety significantly affected most growth and yield parameters. The highest marketable bulb yield (44.57 t/ ha) was obtained from the Nafis variety at 10 cm × 5 cm spacing. Maximum yields of Adama and Bombay Red were recorded at 20 cm × 3 cm spacing. The widest spacing produced the lowest yields across all varieties. Results indicate that optimal spacing is variety-specific; closer spacing favored higher productivity, particularly for Bombay Red and Adama, while Nafis performed well across moderate to close spacings. These findings provide practical recommendations to improve onion productivity under similar agro-ecological conditions.

Introduction

Onion (*Allium cepa* L.) is one of the most important vegetable and spice crops, belonging to the family Alliaceae (Gupta *et al.*, 2025). It is a widely cultivated herbaceous biennial crop believed to have originated in Central Asia, particularly in regions between Turkmenistan and Afghanistan, where its wild relatives still occur. Recent genomic evidence further supports its domestication in the mountainous regions of Iran and surrounding areas, where early selection focused on bulb size and storage quality (Han *et al.*, 2024). Onion was introduced into Ethiopia via Sudan and has since been widely distributed across different parts of the country. It is an indispensable component of Ethiopian cuisine, resulting in consistently high demand throughout the year.

Despite its importance, onion productivity in Ethiopia remains lower than the African and global averages (FAOSTAT, 2024). This low productivity is partly attributed to suboptimal agronomic practices, particularly the use of inappropriate plant spacing and poorly adapted varieties for specific agro-ecological conditions. Plant density is a critical factor influencing onion yield and bulb characteristics. For instance, Negash *et al.* (2022) reported that higher plant density (approximately 100 plants/ m², equivalent to 5 cm spacing) significantly increased total bulb yield, although it reduced individual bulb size. Similarly, Yemane *et al.* (2021) observed improved growth and yield of Adama Red and Bombay Red varieties at plant densities of 47.6 and 33 plants/ m², respectively, in the Aksum area.

The national recommendation for onion production in Ethiopia suggests a spacing of 40 cm between double rows, 20 cm between rows, and 10 cm between plants within rows (EIAR, 2017). However, such blanket recommendations often fail to consider variations in soil fertility and local growing conditions (Yemane *et al.*, 2021). As a result, farmers frequently adjust plant spacing to optimize yield, as increasing plant population density can enhance total yield per unit area (Assaye *et al.*, 2020). Nonetheless, achieving a balance between bulb size preferred by consumers and efficient land use remains a key challenge (Gebrekorkos *et al.*, 2017).

The use of suitable, well-adapted varieties in combination with appropriate agronomic practices is essential for maximizing onion productivity. In the Mersa district of North Wollo, Ethiopia, farmers employ varying planting spacings, often without clear guidance on optimal configurations for different varieties. The lack of location-specific recommendations contributes to reduced yields and inconsistent bulb quality. Therefore, this study was conducted to determine the optimum plant spacing for maximizing marketable bulb yield of three improved onion varieties under the agro-ecological conditions of Mersa district, North Wollo, Ethiopia.

Material and Methods

The experiment was conducted under irrigated conditions during the 2020–2021 cropping season at Mersa District, North Wollo, Ethiopia, located about 491 km northeast of Addis Ababa and 30 km south of Woldia town (11°39′00″–11°41′00″ N, 39°38′30″–39°40′30″ E; 1625 m amsl). The area receives an average annual rainfall of 875 mm with a bimodal distribution and a temperature range of 15–28.5°C. The soil is predominantly clay loam, classified as Vertisol, with slightly acidic to slightly alkaline reaction. The treatments comprised a factorial combination of three onion (*Allium cepa* L.) varieties (Adama Red, Bombay Red, and Nafis) and six planting spacings (20 × 7, 10 × 7, 20 × 5, 10 × 5, 20 × 3, and 10 × 3 cm), laid out in a randomized complete block design with three replications. The experiment included 54 plots (1.0 × 1.5 m each) with 1.0 m and 1.5 m spacing between plots and blocks, respectively, covering a total area of 270 m²; furrow spacing was maintained at 40 cm. Seeds were sown on 22 November 2020 on raised nursery beds (1 × 5 m) with 10 cm row spacing, and 45-day-old seedlings (3–4 leaves) were transplanted on 08 January 2021 after proper hardening.

The field was prepared by ploughing and harrowing, and uniform, healthy seedlings were transplanted as per treatment, with gap filling within one week. Fertilizer was applied at 200 kg ha^{-1} NPSB at transplanting and 100 kg ha^{-1} urea in two splits (half at transplanting and half six weeks later). The crop was irrigated using furrow irrigation at 4–5 day intervals during establishment and 5–7 day intervals thereafter, and irrigation was stopped 15 days before harvest. Harvesting was done at about 70% neck fall, followed by field curing for four days. Data were recorded on plant and plot bases; five plants per plot were sampled for plant-based parameters, while phenological data were recorded on a whole-plot basis, excluding border rows. Data were tested for normality and subjected to analysis of variance using SAS (version 9.3), and treatment means were compared using LSD at 5% probability level; Pearson's correlation analysis was also performed.

Results and Discussion

The interaction effects of plant spacing and variety on phenological, growth, and yield parameters of onion are presented in Table 1. Bolting percentage was significantly ($P<0.01$) influenced by both main effects and their interaction ($P<0.05$). The highest bolting (22.00%) was recorded in Adama Red at the closest spacing (10 × 3 cm), followed by Nafis (20.97%), whereas the lowest value (9.67%) was observed in Nafis at the widest spacing (20 × 7 cm). In general, bolting increased with closer spacing, likely due to increased competition for nutrients and moisture, leading to stress-induced reproductive development (Assaye *et al*., 2020).

Bulb diameter and bulb length were significantly ($P<0.01$) affected by spacing, variety, and their interaction. The largest bulb diameter (7.77 cm) and length (6.43 cm) were recorded in Nafis at 20 × 7 cm spacing, while the smallest diameter (3.83 cm) and shorter bulbs were observed in Adama Red under closer spacing (10 × 3 cm). Wider spacing favored bulb enlargement due to reduced competition and better resource availability, whereas closer spacing restricted bulb growth. Similarly, bulb neck thickness, bulb weight, and bulb dry weight showed significant variation among treatments. The highest neck thickness (1.60 cm), bulb weight (152.37 g), and dry weight (30.97 g) were obtained from Nafis at 20 × 7 cm spacing, while the lowest values were recorded under closer spacing, particularly in

Bombay Red and Adama Red. This trend indicates that lower plant density enhances vegetative growth and assimilate accumulation, resulting in larger bulbs (Muhammad *et al.*, 2017; Yemane *et al.*, 2021).

Table 1. The interaction effects of planting space and variety on different parameters of onion

Variety	Bolting percentage (%)					
Spacing	20 × 7 cm	10 ×7 cm	20 ×5 cm	10 ×5 cm	20 ×3 cm	10 ×3 cm
Adama Red	14.43^{fghi}	15.30^{efgh}	19.50^{abc}	19.17^{abc}	19.28^{abc}	22.00^{a}
Bombay Red	12.27^{hijk}	11.77^{ijk}	13.13^{ghij}	15.42efg	16.10^{defg}	16.83^{cdef}
Nafis	9.67^{k}	11.00^{jk}	13.50^{ghij}	18.23^{bcde}	18.83^{abc}	20.97^{ab}
LSD (5%) : 3.03; CV (%):11.12						
	Bulb diameter (cm)					
Adama Red	7.42^{ab}	6.83^{bcd}	6.66^{bcd}	5.77^{ef}	4.87^{g}	3.83^{h}
Bombay Red	7.65^{a}	6.87^{bcd}	6.45^{de}	6.43^{de}	5.87^{ef}	5.65^{f}
Nafis	7.77^{a}	7.20^{abc}	6.76^{bcd}	6.45^{de}	6.27^{def}	4.69^{g}
LSD (5%) : 0.73; CV (%):6.56						
	Bulb length (cm)					
Adama Red	5.72^{abc}	5.39^{cde}	5.67^{bcd}	4.97^{def}	5.07^{cdef}	4.57^{f}
Bombay Red	6.35^{ab}	5.10^{cdef}	4.93^{def}	5.17^{cdef}	4.72^{ef}	4.60^{f}
Nafis	6.43^{a}	6.36^{ab}	5.61^{cd}	5.11^{cdef}	5.22^{cdef}	4.73^{ef}
LSD (5%) : 0.74; CV (%) :7.19						
	Bulb neck thickness (cm)					
Adama Red	1.21^{bc}	1.13^{c}	0.92^{de}	0.72^{fg}	0.72^{fg}	0.65^{ghi}
Bombay Red	1.30^{b}	1.10^{c}	0.80^{ef}	0.68^{gh}	0.55^{ij}	0.48^{j}
Nafis	1.60^{a}	1.27^{b}	1.28^{b}	1.20^{bc}	0.97^{d}	0.59^{hij}
LSD (5%) : 0.12; CV (%):7.45						
	Bulb weight (g)					
Adama Red	127.80^{b}	112.00^{cde}	111.67^{de}	86.13^{gh}	77.47^{hi}	58.83^{k}
Bombay Red	149.33^{a}	124.07^{bc}	99.27^{f}	73.87^{ij}	67.60^{ijk}	63.67^{jk}
Nafis	152.37^{a}	128.33^{b}	123.80^{bcd}	102.33^{ef}	95.47^{fg}	72.13^{ij}
LSD (5%) :12.15 ; CV: 7.39						
	Bulb dry weight (g)					
Adama Red	23.75^{bc}	21.77^{cd}	19.64^{def}	16.39^{gh}	15.34^{hi}	12.42^{j}
Bombay Red	25.67^{b}	20.27^{de}	18.36^{efg}	17.84^{efgh}	12.78^{ij}	11.16^{j}
Nafis	30.97^{a}	20.49^{de}	19.70^{def}	17.26^{fgh}	13.34^{ij}	13.65^{ij}
LSD (5%) : 2.68; CV (%):8.97						
	Total bulb yield (t/ ha)					
Adama Red	28.73^{i}	33.99^{fgh}	32.62^{fgh}	34.80^{efg}	40.53^{abcd}	30.96^{gh}
Bombay Red	29.13^{i}	36.74^{cdef}	40.05^{bcde}	43.08^{ab}	35.46^{defg}	32.22^{fgh}
Nafis	30.40^{hi}	41.83^{abc}	45.84^{a}	42.90^{ab}	39.70^{bcde}	35.08^{defg}
LSD (5%) : 5.46; CV (%) : 9.05						

Means with same letter(s) are not significantly different at 5% level of significance

Total bulb yield was significantly (P<0.01) affected by both main and interaction effects. The highest yield (45.84 t/ ha) was obtained from Nafis at 20 × 5 cm spacing, followed by Bombay Red (43.08 t/ ha) at 10 × 5 cm, whereas the lowest yield (28.73 t/ ha) was recorded in Adama Red at the widest spacing (20 × 7 cm). Yield increased with closer to moderate spacing due to higher plant population per unit area, which compensated for reduced individual bulb size. Although wider spacing improved individual bulb traits, it resulted in lower yield per unit area. Thus, optimum productivity depends on balancing plant density and individual plant performance, in agreement with earlier findings (Rahel and Khasay, 2018).

Conclusion

Onion productivity in Mersa is significantly influenced by varietal differences and plant spacing, indicating that the blanket recommendation (40 × 20 × 10 cm) is not suitable for all varieties. The highest total bulb yield (45.84 t/ ha) was recorded from Nafis at 20 × 5 cm, while maximum marketable yield was obtained at 10 × 5 cm. Bombay Red also performed best at closer spacing (10 × 5 cm), whereas Adama Red showed improved yield under relatively narrow spacing. Wider spacing enhanced individual bulb size but reduced total yield. Therefore, spacing of 10 × 5 cm to 20 × 5 cm for Nafis and closer spacing (around 10 × 5 cm) for Bombay Red and Adama Red is recommended for higher productivity under similar agro-ecological conditions.

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Conflict of Interest

The authors have no conflict of interest to declare.

Data Sharing

All relevant data are included in the manuscript.

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Research Article

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Effect of exogenous application of plant growth regulators on vegetative growth and flowering of guava cv. Allahabad Safeda

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ABSTRACT

The experiment was conducted during 2024-25 at Rajasthan College of Agriculture, Udaipur, to evaluate the influence of exogenous application of plant growth regulators on vegetative growth and flowering of the guava cv. Allahabad Safeda. Foliar application of 200 ppm gibberellic acid proved most effective in enhancing the tree height (0.63±0.01 m), tree spread (1.57±0.04 m E-W and 1.25±0.06 m N-S) and canopy volume (10.36±0.34 m^3). The increase in stem diameter was greatest with 1500 ppm CCC (1.89±0.01 cm). The total chlorophyll content was greatest (2.32±0.12 mg/g) with 2 ppm brassinosteroids. Gibberellic acid (200 ppm) not only hastened the days to first flowering (27.20±0.32 days) but also accelerated the progression to 50% flowering (53.13±0.81 days). The highest fruit set (78.33±0.26%) and fruit retention (60.33±1.53%) and the highest number of flowers/ shoot (7.55±0.04) were recorded with 200 ppm gibberellic acid. The lowest fruit drop (39.67±1.53%) was also recorded in the treatment 200 ppm gibberellic acid. Overall, 200 ppm gibberellic acid was most effective in promoting vegetative growth and flowering in guava.

Introduction

Guava (*Psidium guajava* L.) a fruit revered for its nutritional richness and delectable flavour, is believed to have originated from the American tropics. *Psidium* is a large genus consisting of as many as 150 species listed in the Kew Index (Pommer and Murakami, 2009). Guava is cultivated in more than 50 countries across tropical and subtropical regions of the world, with major cultivation in India, Brazil, Mexico, Thailand, the USA (Hawaii and Florida), Australia, Philippines, China, Indonesia, Cuba, Java, Malaysia, Bangladesh, Sri Lanka, Myanmar and several African countries. Guava is the fourth most important fruit of India in terms of area and production. India is the world's top guava-producing country, with a growing area of 3.68 lakh ha and 5.48 million tons of production with a productivity of 14.9 MT per hectare (Anonymous, 2024). The top guava producing states in India are Madhya Pradesh, Uttar Pradesh, Andhra Pradesh, Tamil Nadu, Punjab, Bihar and Maharashtra. It can tolerate high temperatures up to 46.1ºC and low temperatures up to -1°C; however, extreme temperatures during flowering reduce the fruit set percentage and enhance the flower drop and fruit drop percentage.

PGRs have been reported to influence vegetative growth, flowering behaviour, fruit set and yield in guava. However, the response of guava to brassinosteroids, gibberellic acid, growth retardants (CCC) and ethrel with their concentrations varies under different agro-climatic conditions (Fischer and Melgarejo, 2021). Foliar application of PGRs improves flowering behaviour and vegetative growth, increases fruit set and enhances fruit retention, thereby reducing the proportion of flowers and young fruits that are shed prematurely (Lal and Das, 2017; Kumar *et al.*, 2025). Keeping in view, the study was undertaken to identify suitable plant growth regulators and concentrations for improving vegetative growth and flowering behaviour of guava.

Material and Methods

The experiment was conducted during 2024-25 on guava cv. Allahabad Safeda at the Horticulture Farm, Rajasthan College of Agriculture, Udaipur, Rajasthan (24° 34' N latitude and 73° 42' E longitude). Thirteen treatments *viz*., control (T_1), brassinosteroids @ 1 ppm (T_2), brassinosteroids @ 1.5 ppm (T_3), brassinosteroids @ 2 ppm (T_4), gibberellic acid @ 100 ppm (T_5), gibberellic acid @ 150 ppm (T_6), gibberellic acid @ 200 ppm (T_7), CCC @ 500 ppm (T_8), CCC @ 1000 ppm (T_9), CCC @ 1500 ppm (T_{10}), ethrel @ 250 ppm (T_{11}), ethrel @ 500 ppm (T_{12}) and ethrel @ 750 ppm (T_{13}) were applied by foliar spray, one month before flowering in a randomized block design and replicated thrice.

Observations for vegetative growth parameters like change in tree height, change in trunk diameter, change in tree spread (N-S & E-W), leaf chlorophyll content (a, b and total), change in canopy volume and light interception below canopy were observed and for flowering parameters like days taken to first flowering, 50% flowering, number of flowers per shoot, fruit set (%), fruit drop (%) and fruit retention (%) were taken. Trunk diameter was measured as described by Matthews and Mackie (2006). The tree spread was measured in all four directions, East-West and North-South tree spread was measured by metric tape at the time of application of treatment and after harvesting. The canopy volume was calculated by the formula of Westwood and Roberts (1963) and calculated as (m^3) = 4/3 $\pi a^2 b$ where, a = half of the spread and b = half of the height.

Light intensity was measured by 'Luxmeter' (electronic digital Luxmeter) at the crop surface and below the canopy. The reading was taken at the time of application of treatments and after harvesting. The chlorophyll content (chlorophyll 'a', chlorophyll 'b' and total chlorophyll) was estimated according to the method of Hiscox and Israelstom (1979). Recently matured leaf (4^{th} leaf) was collected from the individual trees, and chlorophyll was extracted in 5 ml DMSO from 25 mg fresh leaf tissue by DMSO. In the non-destructive DMSO method, tissues were incubated in a glass beaker for 24 hours at 60°C until the tissues became colourless. Absorbance at 663 and 645 nm was determined by a spectrophotometer. The contents of Chlorophyll 'a' and 'b' were calculated by the following equations:

$$Chlorophyll\ a = \frac{(12.7x\,A_{663}) - (2.69x\,A_{645})}{1000} \times \frac{\text{Volume of DMSO}}{\text{Weight of leaf sample}}$$

$$Chlorophyll\ b = \frac{(22.9x\,A_{645}) - (4.65x\,A_{663})}{1000} \times \frac{\text{Volume of DMSO}}{\text{Weight of leaf sample}}$$

Total chlorophyll was calculated by adding chlorophyll *'a'* to chlorophyll *'b'*.

The total number of flowers was counted on the five randomly selected shoots and the average number of flowers/ shoot was calculated. The per cent fruit set was calculated one month after anthesis from five tagged branches.

$$Fruit\ Set(\%) = \frac{\text{Number of fruits set (Initially)}}{Number\ of\ flowers} \times 100$$

$$Fruit\ Drop\ (\%) = \frac{(\text{Total no. of fruit set} - \text{Total no. of fruits at harvest})}{\text{Total no. of fruits set}} \times 100$$

Fruit retention was noted at the time of final harvesting, when the fruits were fully mature. The total number of fruits harvested was counted and the percentage fruit retention was calculated as follows:

$$Fruit\ Retention\ (\%) = \frac{\text{Total number of fruits retained}}{\text{Total number of fruits initially set}} \times 100$$

Data were subjected to analysis of variance (ANOVA) and treatment means were separated using Tukey's HSD test at 5% LOS.

Results and Discussion

Effect on growth parameters

The foliar application of plant growth regulators significantly influence the growth parameters of guava (Table 1). The application of gibberellic acid at 200 ppm (T_7) produced the greatest increase in tree height (0.63±0.01 m), marking a 133% improvement over the water-sprayed control. A dose-dependent response was evident within GA_3 treatments, as 150 ppm (T_6) also promoted substantial elongation (0.56±0.02 m), whereas lower concentrations were less effective. CCC @ 1500 ppm (0.19±0.01 m) suppressed vertical growth markedly.

Significant variation in the trunk diameter with the different levels of different plant growth regulators. The increase in trunk diameter varied markedly across PGR treatments, ranging from a minimal 0.46±0.00 cm under gibberellic acid at 200 ppm (T_7) to a maximal 1.89±0.01 cm with CCC at 1500 ppm (T_{10}). This inverse relationship between longitudinal and radial growth reflects the differential allocation of photo-assimilates and hormonal signals under each regulator. Conversely, exogenous gibberellic acid at increasing concentrations prioritized vertical extension. The lowest diameter gain in T_7 confirms that elevated GA_3 levels drive auxin-GA_3 cross-talk toward cell elongation rather than cambial proliferation. Mid-level GA_3 treatments (100-150 ppm) produced moderate trunk thickening, indicating a dose-dependent trade-off between height and girth.

Gibberellic acid at 200 ppm (T_7) produced the widest change in tree spread (1.57±0.04 m in the east-west axis and 1.25±0.06 m north-south), whereas CCC at 1,500 ppm (T_{10}) yielded the narrowest change in tree spread, with just 0.47±0.03 m (E-W) and 0.38±0.04 m (N-S), respectively. Gibberellic acid's ability to break apical dominance and stimulate axillary bud outgrowth explains its dramatic effect on lateral branch extension. GA_3 at 200 ppm in our study more than doubled lateral expansion relative to the control, reflecting a dose-dependent promotion of internodal elongation and branch proliferation.

Gibberellic acid at 200 ppm (T_7) maximized the change in canopy volume (10.36±0.34 m^3), in stark contrast to Ethrel at 750 ppm (T_{13}), which produced the smallest change in canopy volume (3.89±0.12 m^3). These structural changes dramatically altered light penetration. Under T_7, below-canopy light intercepted averaged just 23885 lux, reflecting dense foliage, whereas CCC @ 1500 ppm treated trees (T_{10}) admitted up to 30227 lux beneath the canopy (above-canopy irradiance was 73820 lux for all plots). These results are in conformity with Jain and Dashora (2007), Sharma and Tiwari (2015), Carpenter *et al.* (2019) and Sarolia *et al.* (2019).

Table 1. Effect of exogenous plant growth regulators on growth parameters of guava

Treatment	Change in tree height (m)	Change in trunk diameter (cm)	Change in tree spread (E-W) (m)	Change in tree spread (N-S) (m)	Change in canopy volume (m^3)
Control (T_1)	0.27±0.01^{def}	0.67±0.00^{e}	0.67±0.02^{fgh}	0.53±0.03^{def}	4.53±0.21^{efg}
Brassinosteroids @ 1 ppm (T_2)	0.30±0.02^{de}	0.69±0.01^{e}	0.74±0.01^{f}	0.59±0.03^{de}	5.30±0.09^{de}
Brassinosteroids @ 1.5 ppm (T_3)	0.35±0.01^{cd}	0.93±0.02^{d}	0.87±0.03^{e}	0.70±0.03^{cd}	6.12±0.09^{d}
Brassinosteroids @ 2 ppm (T_4)	0.42±0.01^{bc}	1.21±0.03^{c}	1.04±0.03^{d}	0.83±0.03^{bc}	7.07±0.06^{c}
Gibberellic acid @ 100 ppm (T_5)	0.47±0.03^{b}	0.92±0.00^{d}	1.18±0.02^{c}	0.95±0.03^{b}	8.29±0.10^{b}
Gibberellic acid @ 150 ppm (T_6)	0.56±0.02^{a}	0.54±0.01^{f}	1.41±0.01^{b}	1.13±0.03^{a}	9.52±0.37^{a}
Gibberellic acid @ 200 ppm (T_7)	0.63±0.01^{a}	0.46±0.00^{f}	1.57±0.04^{a}	1.25±0.06^{a}	10.36±0.34^{a}
CCC @ 500 ppm (T_8)	0.26±0.01^{ef}	1.00±0.02^{d}	0.64±0.01^{fgh}	0.52±0.04^{ef}	5.14±0.12^{ef}
CCC @ 1000 ppm (T_9)	0.23±0.02^{ef}	1.37±0.00^{b}	0.59±0.01^{ghi}	0.46±0.03^{ef}	4.45±0.09^{efg}
CCC @ 1500 ppm (T_{10})	0.19±0.01^{f}	1.89±0.01^{a}	0.47±0.03^{i}	0.38±0.04^{f}	4.02±0.10^{g}
Ethrel @ 250 ppm (T_{11})	0.28±0.02^{de}	0.66±0.01^{e}	0.71±0.04^{fg}	0.57±0.03^{de}	5.19±0.07^{ef}
Ethrel @ 500 ppm (T_{12})	0.26±0.01^{ef}	0.92±0.01^{d}	0.64±0.02^{fgh}	0.51±0.03^{ef}	4.35±0.05^{fg}
Ethrel @ 750 ppm (T_{13})	0.23±0.01^{ef}	1.24±0.02^{c}	0.58±0.03^{hi}	0.47±0.04^{ef}	3.89±0.12^{g}

Mean ± SE followed by the same letter is not significantly different at p=0.05 as determined by Tukey's HSD test.

Effect on light interception and chlorophyll content

The data presented in Table 2 indicate that foliar application of plant growth regulators significantly influences light interception and chlorophyll content in guava. Under T_7, below-canopy light intercepted averaged just 23885 lux, reflecting dense foliage, whereas CCC @ 1500 ppm treated trees (T_{10}) admitted up to 30227 lux beneath the canopy (above-canopy irradiance was 73820 lux for all plots). Brar *et al.* (2012) similarly demonstrated that denser guava canopies sharply reduce solar radiation reaching inner foliage, undermining sub-canopy photosynthesis and fruit quality.

The foliar application of brassinosteroids at 2 ppm (T_4) resulted in the highest leaf pigment levels with chlorophyll 'a' (1.50±0.10 mg/ g), chlorophyll 'b' (0.82±0.02 mg/ g) and total chlorophyll (2.32±0.12 mg/ g), whereas ethrel at 750 ppm (T_{13}) produced the lowest values (1.12±0.01, 0.58±0.02 and 1.70±0.01 mg/ g, respectively).

Table 2. Effect of exogenous plant growth regulators on light interception and chlorophyll content

Treatment	Light interception below canopy (Lux)	Chlorophyll 'a' (mg/ g)	Chlorophyll 'b' (mg/ g)	Total chlorophyll (mg/ g)
Control (T_1)	29715^{abc}	1.27±0.04^{bcdef}	0.67±0.01^{bcde}	1.94±0.03^{cdefg}
Brassinosteroids @ 1 ppm (T_2)	28948^{cd}	1.45±0.01^{abc}	0.75±0.05^{abc}	2.20±0.06^{abc}
Brassinosteroids @ 1.5 ppm (T_3)	28124^{d}	1.47±0.02^{ab}	0.79±0.05^{ab}	2.26±0.07^{ab}
Brassinosteroids @ 2 ppm (T_4)	27175^{e}	1.50±0.10^{a}	0.82±0.02^{a}	2.32±0.12^{a}
Gibberellic acid @ 100 ppm (T_5)	25955^{f}	1.37±0.04^{abcde}	0.73±0.01^{abcd}	2.10±0.03^{abcde}
Gibberellic acid @ 150 ppm (T_6)	24722^{g}	1.41±0.07^{abcd}	0.75±0.01^{abcd}	2.16±0.06^{abcd}
Gibberellic acid @ 200 ppm (T_7)	23885^{g}	1.43±0.03^{abcd}	0.78±0.01^{ab}	2.20±0.04^{abc}
CCC @ 500 ppm (T_8)	29110^{bc}	1.32±0.04^{abcdef}	0.72±0.02^{abcd}	2.03±0.06^{bcdef}
CCC @ 1000 ppm (T_9)	29794^{abc}	1.28±0.04^{bcdef}	0.69±0.01^{abcde}	1.98±0.03^{cdef}
CCC @ 1500 ppm (T_{10})	30227^{a}	1.26±0.04^{cdef}	0.67±0.01^{bcde}	1.93±0.03^{defg}
Ethrel @ 250 ppm (T_{11})	29052^{bc}	1.23±0.02^{def}	0.64±0.01^{cde}	1.87±0.02^{efg}
Ethrel @ 500 ppm (T_{12})	29894^{ab}	1.17±0.01^{ef}	0.62±0.03^{de}	1.79±0.02^{fg}
Ethrel @ 750 ppm (T_{13})	30360^{a}	1.12±0.01^{f}	0.58±0.02^{e}	1.70±0.01^{g}

*Light interception above canopy: 73820 lux

Effect on flowering parameters

The data presented in Table 3 demonstrate that foliar application of plant growth regulators significantly affects the flowering parameters of guava. Gibberellic acid at 200 ppm (T_7) not only hastened the days to first flowering (27.20±0.32) but also accelerated its progression, bringing 50% of the flowers into bloom in just 53.13±0.81 days, compared with 42.27±0.86 and 69.17±1.57 days in the untreated control; a reduction of nearly 15 days was seen. In contrast, untreated control (T_1) required the longest duration to reach 50% flowering (69.17±1.57 days), reflecting the natural pace of floral development in the absence of hormonal stimulation. Similar results were found by Jain and Dashora (2007) in guava cv. Sardar.

Gibberellic acid at 200 ppm (T_7) produced the highest flower count, 7.55±0.04 flowers per shoot, whereas the untreated control (T_1) had the fewest, at just 4.84±0.40 flowers per shoot. The foliar gibberellic acid at 200 ppm (T_7) produced the highest fruit set (78.33±0.26%) versus just 59.13±1.44% in the untreated control.

Fruit drop closely mirrored initial set rates. Under GA_3 treatment (T_7), post-set abscission was only 39.67±1.53%, while in the control it was 58.47±1.72%. Gibberellins exert an anti-abscission effect by down-regulating ethylene biosynthesis in the abscission zone and maintaining cell-wall integrity around the pedicel. By contrast, ethrel treatment @ 750 ppm amplifies ethylene release, up-regulating abscission enzymes and provoking 50-55% fruit drop, a phenomenon broadly observed across pome, stone and tropical fruit studies summarized by Rademacher (2000).

The foliar spray of gibberellic acid at 200 ppm (T_7) showed the highest fruit retention (60.33±1.53%) versus just 41.53±1.72% in the untreated control. Gibberellins enhance sink strength in young ovaries by up-regulating cell-cycle genes and stimulating assimilate partitioning toward developing floral buds. Similar advances have been reported by Sharma and Tiwari (2015) and Agnihotri *et al*. (2016).

Table 3. Effect of exogenous plant growth regulators on flowering parameters of guava

Treatment	Days taken to first flowering (DAS)	Days taken to 50% flowering (DAS)	Number of flowers per shoot	Fruit set (%)	Fruit drop (%)	Fruit retention (%)
Control (T_1)	42.27±0.86^{a}	69.17±1.57^{a}	4.84±0.40^{d}	59.13±1.44^{e}	58.47±1.72^{a}	41.53±1.72^{e}
Brassinosteroids @ 1 ppm (T_2)	39.10±0.62^{ab}	61.43±1.63^{b}	6.36±0.18^{bc}	70.43±1.92^{bc}	47.13±2.10^{de}	52.87±2.11^{ab}
Brassinosteroids @ 1.5 ppm (T_3)	36.63±0.07^{bc}	59.13±0.83^{bc}	6.64±0.09^{abc}	72.23±1.24^{ab}	46.37±1.70^{de}	53.63±1.70^{ab}
Brassinosteroids @ 2 ppm (T_4)	35.13±0.27^{bcd}	57.60±0.40^{bcd}	6.75±0.28^{ab}	73.87±0.83^{ab}	45.80±1.97^{de}	54.20±1.97^{ab}
Gibberellic acid @ 100 ppm (T_5)	32.70±1.68^{cde}	57.27±0.50^{bcd}	6.61±0.27^{abc}	72.07±0.90^{ab}	46.77±2.24^{de}	53.23±2.24^{ab}
Gibberellic acid @ 150 ppm (T_6)	31.10±0.00^{def}	57.03±0.73^{bcd}	6.83±0.12^{ab}	74.10±2.23^{ab}	45.33±1.16^{de}	54.67±2.00^{ab}
Gibberellic acid @ 200 ppm (T_7)	27.20±0.32^{f}	53.13±0.81^{d}	7.55±0.04^{a}	78.33±0.26^{a}	39.67±1.53^{e}	60.33±1.53^{a}
CCC @ 500 ppm (T_8)	37.10±0.79^{b}	61.00±1.88^{b}	5.28±0.19^{d}	67.97±2.28^{bcd}	48.03±0.85^{cd}	51.97±0.85^{bc}
CCC @ 1000 ppm (T_9)	35.13±0.98^{bcd}	58.63±1.18^{bc}	5.58±0.15^{cd}	64.70±2.51^{cde}	51.30±1.32^{abcd}	48.70±1.32^{bcde}
CCC @ 1500 ppm (T_{10})	30.63±0.94^{ef}	56.70±0.26^{bcd}	6.39±0.12^{bc}	63.23±0.50^{de}	56.27±1.05^{ab}	43.73±1.05^{de}
Ethrel @ 250 ppm (T_{11})	35.27±0.70^{bcd}	59.03±0.58^{bc}	6.56±0.07^{abc}	64.00±0.93^{cde}	50.63±1.19^{bcd}	49.37±1.19^{bcd}
Ethrel @ 500 ppm (T_{12})	32.83±0.96^{cde}	57.57±0.53^{bcd}	6.78±0.34^{ab}	63.77±0.67^{cde}	55.27±1.40^{abc}	44.73±1.40^{cde}
Ethrel @ 750 ppm (T_{13})	28.80±0.84^{ef}	55.20±0.38^{cd}	7.38±0.19^{ab}	61.83±1.16^{de}	57.60±1.17^{ab}	42.40±1.18^{de}

Mean ± SE followed by the same letter is not significantly different at p=0.05

Conclusion

The present study concluded that exogenous application of plant growth regulators markedly affects vegetative growth and flowering behaviour in guava. Gibberellic acid at 200 ppm proved most effective, enhancing plant height, canopy development, early flowering, fruit set, and retention while reducing fruit drop. CCC at 1500 ppm restricted vertical growth but promoted stem thickening, contributing to a compact canopy structure. Brassinosteroids at 2 ppm improved chlorophyll content and photosynthetic efficiency, whereas ethrel at 750 ppm negatively influenced growth by increasing fruit drop and chlorophyll degradation. Overall, gibberellic acid at 200 ppm is recommended for improving growth, productivity, and orchard management in guava.

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Conflict of Interest

The authors declare 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 foliar application of elicitors on growth and yield of turmeric (*Curcuma longa* L.)

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ABSTRACT

A field experiment was conducted during 2024-25 at farmer's field to study the effect of foliar spray of elicitors on growth and yield of turmeric. The experiment was laid out in RBD with seven treatments replicated thrice. Application of salicylic acid (100 ppm) at 150, 180 and 210 days after planting (DAP), consistently produced maximum plant height (103.39 cm), number of leaves (15.82), leaf length (60.48 cm) and leaf width (16.24 cm). Yield attributes viz., total rhizomes (26.15), number and weight of mother rhizomes (2.95; 186.12 g), number and weight of primary rhizomes (10.12; 199.53 g), number and weight of secondary rhizomes (11.40; 58.07 g), yield per plant (443.72 g) and dry recovery (26.17%) were recorded highest with salicylic acid (100 ppm) sprayed at 150, 180 and 210 DAP. Therefore, salicylic acid at 100 ppm is recommended to improve growth and yield of turmeric.

Introduction

Turmeric (*Curcuma longa* L.) is an important spice crop cultivated widely in tropical and subtropical regions. India is the leading producer of turmeric, contributing nearly 80% of the global output, with an area of 2.97 lakh ha and production of 10.42 lakh tonnes during 2023-24, with an average productivity of 3.5 t/ ha (Turmeric Outlook, 2025). Major turmeric producing states in India include Maharashtra, Telangana, Tamil Nadu, Karnataka, Madhya Pradesh, Odisha and West Bengal. The presence of curcuminoids, particularly curcumin, imparts important medicinal properties (Anusuya and Sathiyabama, 2015). Despite its value, turmeric faces poor sprouting, weak growth, low yield and quality due to lack of effective crop management practices.

In recent years, foliar application of elicitors has emerged as an eco-friendly and sustainable approach to improve crop growth, yield and quality (Brugger *et al.*, 2006). Jasmonic acid plays a vital role in regulating plant growth, rhizome development, senescence and stress responses (Ahmad and Murali, 2015). Salicylic acid improves ion regulation, stress tolerance and phenolic compound synthesis (Ghasemzadeh and Jaafar, 2012). Thus, the use of elicitors represents a promising approach to enhance vegetative growth and yield attributes. In view of this, the present study was undertaken to assess the effectiveness of foliar application of different elicitors on the growth and yield of turmeric.

Material and Methods

A field experiment was conducted at farmer's field in Sindhol village near Manhalli, Bidar, Karnataka during 2024-25. The experiment was laid out in a Randomized Block Design (RBD) with seven treatments and three replications. Each plot measured 6 m x 6 m. Rhizomes of cultivar IISR Prathibha were planted at a spacing of 40 cm between rows and 15 cm between plants. The treatments consisted of control (water spray), salicylic acid (100 and 150 ppm), jasmonic acid (200 ppm), chitosan (100 and 150 ppm) and potassium silicate (1500 ppm). Foliar sprays were applied at 120, 150, 180 and 210 days after planting (DAP).

Growth parameters viz., plant height, number of leaves, leaf length, leaf width and number of tillers per plant were recorded on ten plants. Observations on growth parameters were recorded at 120 DAP (before spray application), 150 DAP (30 days after the first spray), 180 DAP (30 days after the second spray) and 210 DAP (30 days after the third spray).

Rhizome yield was determined at the time of harvest. Yield parameters viz., number of rhizomes/ plant, number of mother rhizomes/ plant, weight of mother rhizomes per plant, number of primary rhizomes/ plant, weight of primary rhizome/ plant, number of secondary rhizomes/ plant, weight of secondary rhizomes/ plant, fresh rhizome yield/ plant, fresh rhizome yield/ plot and dry rhizome recovery (%) were recorded. The observations were recorded following the standard procedures (DASD, 2020) and data were subjected to statistical analysis (Fisher, 1950).

For dry rhizome recovery (%), fingers were separated from the mother rhizomes. Fresh samples were boiled in clean water at 50–60°C for 60 minutes until soft. The boiled rhizomes were then sun-dried for 15 days, ground into powder and sieved. The dry rhizome recovery (%) was calculated using the following formula:

$$\text{Dry recovery (\%)} = \frac{\text{Dry weight of powder (g)}}{\text{Fresh weight of the rhizome (g)}} \times 100$$

Results and Discussion

Effect of elicitors on growth attributes

The results revealed that foliar application of elicitors significantly influenced all plant growth parameters of turmeric at 150, 180 and 210 DAS. Foliar spray of salicylic acid @ 100 ppm produced the highest plant height (98.14-103.39 cm), which is on par with chitosan @ 100 ppm, both significantly exceeding the control (Table 1). Salicylic acid @ 150 ppm and chitosan @ 150 ppm showed moderate improvements over the control, but they were less effective. Jasmonic acid @ 200 ppm and potassium silicate @ 1500 ppm recorded only marginal gains.

Salicylic acid @ 100 ppm producing the highest leaf numbers (13.50–15.82), which is on par with chitosan @ 100 ppm (12.92–15.23), both surpassing the control (Table 1). Higher concentrations (150 ppm) were less effective, indicating a dose-dependent response.

Salicylic acid @ 100 ppm produced the longest leaves (58.20-60.48 cm), which is on par with chitosan @ 100 ppm, Treatments with salicylic acid @ 150 ppm and chitosan @ 150 ppm also increased leaf length compared to the control but were less effective than their lower-dose counterparts. Jasmonic acid @ 200 ppm and potassium silicate @ 1500 ppm recorded moderate improvements, whereas the untreated control maintained the shortest leaves (Table 2).

Salicylic acid @ 100 ppm produced the widest leaves (15.90-16.75 cm), which is on par with chitosan @ 100 ppm (14.86-15.41 cm), whereas higher doses showed reduced effectiveness. Jasmonic acid and potassium silicate provided moderate increases, while the control recorded the narrowest leaves (11.92–12.49 cm) (Table 2).

Salicylic acid @ 100 ppm recorded the highest tiller numbers (1.65-1.90), which is on par with chitosan @ 100 ppm (1.59-1.86). Treatments with salicylic acid @ 150 ppm and chitosan @ 150 ppm also improved tiller production compared to the control but were less effective than their lower dose counterparts. Jasmonic acid @ 200 ppm and potassium silicate @ 1500 ppm recorded relatively lower tiller counts while, the control had the lowest tiller numbers (1.20–1.36) (Table 3). These findings are consistent with the finding of Harish *et al*. (2021), El-Sherif *et al*. (2022), Aduguba *et al*. (2024), Sivaranjani *et al.* (2022) and Chintakovid *et al*. (2025).

Table 1. Effect of foliar spray of elicitors on plant height and number of leaves on turmeric

Treatments	Plant height (cm)				Number of leaves per plant			
	120 DAP	150 DAP	180 DAP	210 DAP	120 DAP	150 DAP	180 DAP	210 DAP
Control	76.61	81.58	85.84	86.46	7.93	9.88	10.70	11.20
Chitosan @ 100 ppm	77.38	95.41	99.08	100.27	8.00	12.92	14.40	15.23
Chitosan @ 150 ppm	78.48	89.48	93.13	95.98	7.80	10.95	12.70	12.95
Jasmonic acid @ 200 ppm	77.62	84.06	88.84	89.89	7.93	10.50	11.79	12.65
Potassium silicate @ 1500 ppm	76.07	83.28	87.80	88.12	8.13	9.81	10.77	11.55
Salicylic acid @ 100 ppm	78.10	98.14	102.30	103.39	8.06	13.50	14.97	15.82
Salicylic acid @ 150 ppm	77.73	91.62	95.62	96.33	7.80	11.02	13.06	13.19
Mean	77.43	89.08	93.23	94.35	7.95	11.23	12.63	13.23
SEm±	2.64	2.83	2.85	3.10	0.27	0.42	0.47	0.63
CD (5%)	NS	8.72	8.79	9.57	NS	1.28	1.46	1.95

Table 2. Effect of foliar spray of elicitors on leaf length and leaf width of turmeric

Treatments	Leaf length (cm)				Leaf width (cm)			
	120 DAP	150 DAP	180 DAP	210 DAP	120 DAP	150 DAP	180 DAP	210 DAP
Control	47.43	49.07	50.30	51.40	10.54	12.02	12.49	11.92
Chitosan @ 100 ppm	49.23	57.10	58.10	59.30	11.21	14.96	15.41	14.86
Chitosan @ 150 ppm	48.65	55.97	56.98	58.10	11.08	13.55	14.24	13.91
Jasmonic acid @ 200 ppm	46.42	51.63	52.63	53.80	10.40	12.35	12.95	12.58
Potassium silicate @ 1500 ppm	46.17	50.74	51.74	52.94	10.31	12.94	13.04	12.73
Salicylic acid @ 100 ppm	49.83	58.20	59.50	60.48	11.41	15.90	16.75	16.24
Salicylic acid @ 150 ppm	48.62	56.12	57.12	58.40	11.17	13.97	14.37	14.07
Mean	50.05	54.12	55.19	56.34	10.87	13.67	14.18	13.76
SEm±	1.64	1.67	1.80	1.82	0.37	0.49	0.53	0.49
CD (5%)	NS	5.15	5.55	5.46	NS	1.50	1.62	1.51

Table 3. Effect of foliar spray of elicitors on number of tillers per plant in turmeric

Treatments	120 DAP	150 DAP	180 DAP	210 DAP
Control	1.10	1.20	1.27	1.36
Chitosan @ 100 ppm	1.00	1.59	1.76	1.86
Chitosan @ 150 ppm	1.10	1.45	1.66	1.78
Jasmonic acid @ 200 ppm	1.00	1.31	1.46	1.49
Potassium silicate @ 1500 ppm	1.00	1.37	1.49	1.51
Salicylic acid @ 100 ppm	1.10	1.65	1.80	1.90
Salicylic acid @ 150 ppm	1.10	1.51	1.70	1.75
Mean	1.06	1.44	1.59	1.66
SEm±	0.04	0.07	0.10	0.11
CD (5%)	NS	0.21	0.31	0.34

Effect of elicitors on yield attributes

Foliar application of elicitors significantly enhanced rhizome production in turmeric (Table 4). Salicylic acid at 100 ppm recorded the highest total number of rhizomes per plant (24.47), mother rhizomes (2.95), primary rhizomes (10.12) and secondary rhizomes (11.40), which is on par with chitosan at 100 ppm, which recorded high values for total number of rhizomes per plant (23.39), mother rhizomes (2.90), primary rhizomes (9.42) and secondary rhizomes (11.05). Treatments with salicylic acid at 150 ppm and chitosan at 150 ppm also showed moderate increases. Jasmonic acid @ 200 ppm and potassium silicate @ 1500 ppm recorded only slight improvements over the control.

Foliar application of elicitors significantly influenced rhizomes weight in turmeric (Table 5). The maximum weight of mother rhizomes (186.12 g), primary rhizomes (199.53 g) and secondary rhizomes (58.07 g) were recorded with salicylic acid at 100 ppm, which is on par with chitosan at 100 ppm. Moderate increases were observed with higher concentrations of salicylic acid at 150 ppm and chitosan at 150 ppm, indicating a dose-dependent response where moderate doses were more effective than higher levels. Jasmonic acid at 200 ppm and potassium silicate at 1500 ppm showed only slight improvements over the control.

The fresh rhizome yield of turmeric was significantly influenced by foliar application of elicitors (Table 6). The salicylic acid at 100 ppm produced the highest values (443.72 g/ plant and 88.75 kg/ plot), which is on par with chitosan at 100 ppm (431.89 g/ plant and 86.38 kg/ plot). Dry recovery (Table 6) was highest with salicylic acid @ 100 ppm (26.17%) and chitosan @ 100 ppm (25.06%) showing a substantial increase over the control (18.20%). These findings are in agreement with the results reported by Velayutham and Parthiban (2013), Harish *et al.* (2021), El-Sherif *et al*. (2022), El-Sherif *et al*. (2022), Sivaranjani *et al*. (2022), Aduguba *et al*. (2024) and Tantawy *et al*. (2021).

Table 4. Effect of foliar spray of elicitors on number of rhizomes/ plant in turmeric

Treatments	Number of rhizomes	Number of mother rhizomes	Number of primary rhizomes	Number of secondary rhizomes
Control	16.97	2.53	6.70	7.70
Chitosan @ 100 ppm	23.39	2.90	9.42	11.05
Chitosan @ 150 ppm	21.39	2.77	8.69	9.93
Jasmonic acid @ 200 ppm	19.07	2.68	7.89	8.50
Potassium silicate @ 1500 ppm	18.10	2.59	7.35	8.16
Salicylic acid @ 100 ppm	24.47	2.95	10.12	11.40
Salicylic acid @ 150 ppm	21.89	2.86	8.77	10.26
Mean	20.75	2.76	8.42	9.57
SEm±	0.64	0.09	0.27	0.36
CD (5%)	1.98	0.28	0.82	1.11

Table 5. Effect of foliar spray of elicitors on weight of rhizomes per plant (g)

Treatments	Weight of mother rhizomes	Weight of primary rhizomes	Weight of secondary rhizomes
Control	155.64	165.99	48.52
Chitosan @ 100 ppm	179.08	195.17	57.64
Chitosan @ 150 ppm	167.79	179.31	53.30
Jasmonic acid @ 200 ppm	160.85	169.06	49.55
Potassium silicate @ 1500 ppm	158.97	174.86	50.50
Salicylic acid @ 100 ppm	186.12	199.53	58.07
Salicylic acid @ 150 ppm	169.40	181.69	54.20
Mean	168.27	180.80	53.11
SEm±	5.29	5.37	1.63
CD (5%)	16.29	16.56	5.01

Table 6. Effect of foliar spray of elicitors on fresh rhizome yield and dry recovery

Treatments	Fresh rhizome yield/ plant (g)	Fresh rhizome yield/ plot (kg)	Dry recovery (%)
Control	370.15	74.03	18.20
Chitosan @ 100 ppm	431.89	86.38	25.06
Chitosan @ 150 ppm	400.40	80.08	21.50
Jasmonic acid @ 200 ppm	379.46	75.90	19.93
Potassium silicate @ 1500 ppm	384.33	76.87	18.53
Salicylic acid @ 100 ppm	443.72	88.75	26.17
Salicylic acid @ 150 ppm	405.29	81.06	22.87
Mean	402.18	80.44	21.75
SEm±	5.29	2.37	0.89
CD (5%)	16.29	7.30	2.75

Conclusion

In conclusion, foliar application of elicitors had a significant positive effect on the growth and yield of turmeric. Among the treatments, salicylic acid at 100 ppm emerged as the most effective, recording the highest plant height (103.39 cm), number of leaves (15.23), leaf length (60.8 cm), leaf width (16.24 cm) and tillers/ plant (1.90). Its performance was found to be statistically on par with chitosan at 100 ppm when applied at 150, 180, and 210 DAP. Furthermore, salicylic acid at 100 ppm significantly enhanced yield attributes, including the number of rhizomes (24.47), number of mother rhizomes (2.95) and weight (186.12 g), number of primary rhizomes (10.12) and weight (199.53 g), number of secondary rhizomes (11.40) and weight (58.07), yield per plant (443.72 g), yield per plot (88.75 kg) and dry recovery (26.17).

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Conflict of Interest

The authors declare no conflict of interest.

Data Sharing

All relevant data are within the manuscript.

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Research Article

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Jodhpur Jeera-1: High seed yielding variety of cumin for Rajasthan

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ABSTRACT

Jodhpur Jeera-1 (MCU-105) is a newly developed high-yielding cumin (*Cuminum cyminum* L.) variety developed through pure line selection from a local germplasm collection (GP-205) at Dr. B.R. Choudhary Agricultural Research Station, Mandor, Agriculture University Jodhpur, Rajasthan. The variety was evaluated under the AICRP on Spices at five locations across Rajasthan and Gujarat for three consecutive years (2021-22 to 2023-24). In multi-location coordinated trials, Jodhpur Jeera-1 recorded a weighted mean seed yield of 566 kg/ ha, registering a 22.8% yield advantage over the national check variety GC-4 (469 kg/ ha). The variety possesses a superior essential oil content of 4.34%, which is 3.6% higher than that of GC-4 and is characterized by a tall, erect plant type with abundant branching and compact umbels. It matures in 118–138 days, placing it in the medium maturity group. Under natural field conditions, Jodhpur Jeera-1 exhibits moderate resistance to both Fusarium wilt and Alternaria blight, recording a mean blight disease incidence of 26.0% compared to 34.4% in GC-4, along with lower infestation by aphids and thrips. The variety was recommended for commercial cultivation in irrigated, timely sown *rabi* conditions of Rajasthan in 2025.

Introduction

Cumin (*Cuminum cyminum* L.) is an important annual herbaceous spice crop belonging to the family Apiaceae. It originated in the Irano-Turanian region, including the Eastern Mediterranean to South Asia, and has been cultivated since ancient times. Cumin is cultivated in many parts of the world, particularly in India, Iran, Syria, Turkey and other regions (Singh *et al.*, 2023). India is the world's largest producer and exporter of cumin, contributing about 70% of global production (Ramesh *et al.,* 2025). In India, cumin is cultivated over approximately 10.94 lakh hectares, with a production of about 7.24 lakh tonnes during

2024–25. The average productivity is around 662 kg/ha (Anonymous, 2025). Rajasthan and Gujarat states together accounted for over 80–99% of the national area and production of cumin.

It is a medicinal and aromatic plant that has been used since ancient times for its distinctive flavor, primarily derived from its essential oil (mainly cuminaldehyde, 2–5%). It is widely used in food preparations, including curries, pickles, soups, sauces, bakery products and liquors. Additionally, cumin is valued in traditional medicine for its therapeutic properties, particularly in the treatment of digestive disorders, cough, and flatulence (Merah *et al.*, 2020).

Cumin is a cool-season, *rabi* crop mainly grown during the winter in northern and western India, thriving in arid and semi-arid regions. High humidity during flowering and fruiting promotes fungal diseases, while excessive heat or frost can reduce yield. The crop needs 120 frost-free days and is vulnerable to hot, dry winds or excess moisture at maturity (Allaq *et al.,* 2020).

Biotic and abiotic stresses, along with management practices, significantly affect yield and quality. Like other seed spices, cumin is attacked by several fungal diseases. Alternaria blight (*Alternaria burnsii* or *A. alternata*) is one of the most important and devastating diseases, yield losses up to 50–80% under favorable conditions (humid, cloudy weather). Other major diseases include wilt (*Fusarium oxysporum* f. sp. *cumini*) and powdery mildew (*Erysiphe* spp.), leading to substantial economic damage in major growing areas like Rajasthan and Gujarat (Dharajiya *et al.,* 2023).

In light of these circumstances, a breeding program was undertaken to develop high-yielding cumin varieties with resistance to Alternaria blight and Fusarium wilt through systematic evaluation of available germplasm.

Material and Methods

Evaluation of advance cumin breeding lines

Cumin germplasm and breeding lines were evaluated at the Dr. B. R. Choudhary Agricultural Research Station, Mandor (Rajasthan) during *rabi* season for two successive years 2019-20 for seed yield potential and disease resistance in augmented block design. Further, in initial varietal trials (IVT) in the year 2020-21 at three locations *viz.,* Mandor, Jalore and Samdari, laid out in randomized block division (RBD) with three replications. The row to row and plant to plant distance was maintained at 30 cm and 5-7 cm, respectively. The crop was raised following the recommended agronomical and plant protection methods.

Among the germplasm and breeding lines, MCU-73 and MCU-105 were found superior and selected for further evaluation. Both the entries were further evaluated under coordinated varietal trials (CVT) at five different locations *viz.,* Ajmer (Rajasthan), Mandor (Rajasthan), Jobner (Rajasthan), Sanand (Gujarat) and Jagudan (Gujarat) for three consecutive years 2021-22, 2022-23 and 2023-24. Three years pooled data of all the five locations were analysed and presented in All India Coordinated Research Project on Spices (AICRPs) for superiority against national check variety GC-4. The data were subjected to analysis of variance (Panse and Sukhatme, 1986) and a pooled analysis of variance, where genotypes were assumed as fixed and test environments as random factors (Peterson, 1938).

Pedigree

Cumin variety Jodhpur Jeera-1 (JJ-1) has been developed from the germplasm material collected from Merta area of Nagaur district, Rajasthan. Pure line selection of breeding method was employed to select superior plant type (GP-205). Further the genotype was evaluated under AICRP on Spices using codes MCU-105. Based on its performance over the years and locations, the advanced entry MCU-105 (code CUM-47) has been recommended for release in the Annual Group Meet XXXVth AICRP on Spices workshop held during 15-17th October, 2024 CCS HAU, Hisar. Subsequently, advanced entry MCU-105 was released and notified as variety in the name of Jodhpur Jeera-1 (Gazette Notification S.O.No./4000 (E); dated 01.09.2025).

Results and Discussion

Yield performance under initial evaluation trials

Total 6 genotypes along with one check variety GC 4, were evaluated at ARS, Mandor, Jalore and ARSS, Samdari. Two test entries, MCU-73 (542 kg/ ha; 19.6%) and MCU-105 (522 kg/ ha; 15.2%) were found superior over check variety GC-4 (453 kg/ ha). Based on these yield performances, the genotypes MCU-73 and MCU-105 were contributed to AICRP trials.

Table 1. Seed yield performance of cumin entries against check GC-4 over location and years

S.No.	Entry	Seed yield (kg/ ha)					% increase over check
		Mandor 2019-20	Mandor 2020-21	Jalore 2020-21	Samdari 2020-21	Mean	
1.	MCU-73	567	817	611	175	542	19.6
2.	**MCU-105**	**581**	**728**	**634**	**146**	**522**	**15.2**
3.	MCU-2	532	578	603	97	452	-0.2
4.	**GC-4 (C)**	**444**	**639**	**592**	**139**	**453**	
	G. Mean	323	559	528	114	381	
	SEm±	23	43	27	9		
	CD at 5%	67	133	84	28		
	CV (%)	12.4	13.4	8.9	13.6		

Table 2. Seed yield of cumin varieties in coordinated trials at different locations

Particulars	Mean seed yield (kg/ ha)			Weighted mean	Percent increase over national check GC-4
Year of testing	2021	2022	2023		
Entries/ No. of locations	5	3	2	10	
Check (GC-4)	**513**	**335**	**560**	**469**	**-**
CZC-94	299	147	534	300	-38.7
CZC-135	591	393	545	522	12.3
MCU 73	487	359	595	470	0.8
MCU 105	**584**	**470**	**664**	**566**	**22.8**
JC 18-10	414	200	275	322	-31.9
JC 18-09	372	143	331	295	-39.2
UC 350	405	262	515	384	-18.6
UC 257	187	95	636	249	-50.6
UC 250	227	162	692	301	-38.6
SPS/166/2-3	256	168	562	291	-39.9
BC 13	348	257	457	343	-26.7

Yield performance under multi-location testing

Under AICRP on Spices, the advanced entry MCU-105 (code CUM-47) was performed better as compared to other entries along with national check. Three years pooled data of seed yield for each evaluating centre are presented in Table 2, revealed that the seed yield of proposed entry MCU-105 (584 kg/ ha, 470 kg/ ha, 664 kg/ ha) was found to be superior over national check GC-4 (513 kg/ ha, 335 kg/ ha, 560 kg/ ha) over the years and gained 22.8% higher pooled yield. The consistent performance of MCU-105 over three consecutive years (2021-22 to 2023-24) and across five diverse locations in Rajasthan and Gujarat demonstrates that its yield advantage is stable and not a location- or season-specific phenomenon. Such genotype × environment consistency is a critical criterion for the release of any new variety (Ramesh *et al.*, 2025).

Quality performance

The essential oil content in the variety was considered as quality parameter for the release, henceforth, essential oil was extracted from the dried seed of cumin every year using hydro-distillation method. Three years cumulative data of essential oil are presented in Table 3, revealed that the proposed advanced entry MCU-105 (JJ-1) contained 4.34% essential oil, which was 18.5 per cent higher than national check GC-4 (4.19%). JJ-1 consistently outperformed or matched GC-4 across most AICRP centers in essential oil accumulation, with particularly high values at Jagudan (4.80%) and Sanand (4.94%), suggesting that irrigated, warm-humid agroclimatic zones of Gujarat may further enhance oil biosynthesis in this variety.

Table 3. Essential oil (%) content in seed of the cumin variety tested at various AICRP centres (pooled data 2021-22 to 2022-23)

Location-season	MCU-105	GC 4	Mean	F (Prob)	CD (p=5%)	CV (%)	SE (m)
Ajmer-2022	3.55	3.51	3.31	0.026	0.412	7.354	0.141
Ajmer-2023	3.96	3.87	4.06	0	0.101	1.467	0.034
Jagudan-2021	4.8	4.4	4.66	0	0.348	4.41	0.119
Jagudan-2022	4.73	4.27	4.66	0	0.348	4.41	0.119
Jagudan-2023	4.13	3.4	3.63	0.016	0.593	9.661	0.202
Jobner-2021	4.54	5.18	4.79	0	0.264	3.249	0.09
Jobner-2022	4.43	4.4	4.34	0	0.394	5.352	0.134
Jobner-2023	3.96	4	4.17	0.003	0.649	9.189	0.221
Sanand-2021	4.94	4.68	5.3	-	-	-	-
Grand Mean	4.34	4.19	4.33	-	-	-	-
% higher than GC-4					18.5%		

Resistance to Alternaria blight and Fusarium wilt

The new variety Jodhpur Jeera-1 (MCU-105) possess moderately resistance to Alternaria blight and Fusarium wilt, which causes severe yield loss in cumin. Percent disease index showed that MCU-105 received lower disease incidence as compared to National checks GC-4 (Table 4). Based on three years pooled data, 26.0% PDI blight disease recorded in proposed variety against 34.4% in national checks GC-4. Dharajiya *et al.* (2023) demonstrated that resistance to *F. oxysporum* f. sp. *cumini* in GC-4 involves up-regulation of specific metabolic pathways, and it is likely that JJ-1 has evolved partial resistance through different or overlapping biochemical mechanisms that merit further investigation.

Table 4. Performance of MCU-105 genotypes against Alternaria blight and Fusarium wilt disease in CVT (pooled data 2021-22 to 2023-24)

Disease	Item	JJ-1 (MCU-105)	National Check GC-4
Wilt	1st year	12.0	11.0
	2nd year	29.8	23.7
	3rd year	24.4	28.5
	Mean	22.1	21.1
Blight	1st year	18.4	33.2
	2nd year	31.2	38.8
	3rd year	26.8	29.8
	Mean	26.0	34.4

Plate 1. Plant of Jodhpur Jeera-1 at seed formation stage


Plate 2. Seeds of Jodhpur Jeera-1

Adaptability and stability

JJ-1 was evaluated across five geographically diverse locations *i.e.* Ajmer, Mandor and Jobner in Rajasthan that collectively represent the major cumin-growing agroclimatic zones of Rajasthan. The variety demonstrated above-average performance across all three years of testing, with particularly strong yield expression in the third year (664 kg/ ha at pooled level), suggesting good adaptation to evolving crop management conditions and climatic variability. This broad adaptability is a key requirement for commercial release under the AICRP system (Ramesh *et al.,* 2025).

The variety's maturity duration of 118–138 days places it in the medium maturity group, which is well-suited to the *rabi* cropping schedule of Rajasthan, allowing farmers flexibility in sowing time while avoiding terminal heat and moisture stress during grain filling. Its upright growth habit and compact canopy also facilitate mechanized harvesting and reduce lodging risk, which are increasingly important traits as the farming workforce in Rajasthan transitions toward mechanization.

Conclusion

Jodhpur Jeera-1 (MCU-105), developed through pure line selection from local germplasm represents a significant advancement. JJ-1 recorded a weighted mean seed yield of 566 kg/ ha. It was 22.8% higher than the national check GC-4. It showed higher essential oil content (4.34%). The variety exhibited moderate resistance to Alternaria blight and Fusarium wilt. It also showed broad adaptability across major cumin-growing regions of Rajasthan and Gujarat. Overall, JJ-1 offers a comprehensive agronomic advantage under irrigated *rabi* conditions.

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Conflict of Interest

The authors have no conflict of interest to declare.

Data Sharing

All relevant data are included in the manuscript.

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Research Article

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Survey on the prevalence of fungal diseases and it's impact on the cultivation of fig (*Ficus carica* L.) in Purandar region of Maharashtra

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ABSTRACT

The study evaluated the frequency of fungal infections in fig and their economic implications across 100 orchards in the Purandar region, Maharashtra. Rust-related pathogens (*Cerotelium fici or Phakopsora sp.*) were the most common disease, appearing in 75% of assessed farms, with 25% reporting concomitant infections such as leaf blight and mite infestations. Economic analysis indicated that 50% of farms suffered substantial income loss, with an average market disruption score of 3.25 on a 5-point scale. According to regression analysis, yield was more strongly predicted by farm size ($\beta = 2.517$) than by years of expertise ($\beta = 0.228$). The research underscores the pressing necessity for rust-resistant cultivars, comprehensive disease management, and focused extension initiatives to guarantee the enduring viability of fig farming in the Purandar region.

Introduction

Fig (*Ficus carica* L.), a deciduous tree in the Moraceae family, is one of the oldest cultivated fruit crops, originally from Southwest Asia and currently grown in tropical and subtropical climates around the world. This crop is particularly well-suited to dry and semi-arid agro-climatic zones due to its ability to thrive in water-scarce environments and poor soils. It provides both nutritional value and commercial potential with minimal resource inputs (Singh *et al.*, 2023; Tripathi *et al.*, 2025). As of 2021, India's fig cultivation area was 5,912 hectares, with an annual production of 14,695.56 tonnes. Maharashtra is India's biggest producer, accounting for around 2,705 MT from 947 hectares, surpassing Karnataka and Uttar Pradesh. Purandar tehsil in Pune district is the state's most notable fig-growing zone, spanning 1,332 hectares throughout Purandar and Bhor tehsils, with Purandar taluka accounting for 45.92% of the cultivated area (Daundkar *et al.*, 2016). The Adriatic Common type, which bears fruit without pollination, was granted Geographic Indication (GI) status in 2016, highlighting its agricultural and economic importance for smallholder farmers' livelihoods in semi-arid Maharashtra.

Despite the economic significance, fig farming faces serious and escalating risks from pathogenic fungal infections. According to reports, 94% of fig farmers in Purandar suffer from extensive fungal infections, which result in considerable crop losses. Fig rust, caused by *Cerotelium fici or Phakopsora sp.*, is the most common and devastating fungal disease in *F. carica* worldwide, causing red-orange pustules on leaf undersides, necrosis and defoliation, and a significant reduction in fruit yield and marketability (Avasthi *et al.*, 2023; Khot *et al.*, 2024). Beyond rust, phytopathogenic fungi collectively account for approximately 20% of global crop production loss (Davies *et al.*, 2021), whereas fungal infections in horticulture cause 40-60% of seasonal crop losses, including 15-20% in the field and another 15-20% during packing, storage, and transport (Tripathi *et al.*, 2024). These losses incur major economic consequences, including increased fungicide spending, lower fruit quality and lower market pricing, with smallholder farmers being especially vulnerable.

Despite the increasing economic importance of fig production in India, rigorous field-level assessments of disease prevalence, economic effect, and farmer management strategies are limited. This study fills that gap by quantifying the prevalence and severity of fungal diseases in fig orchards in the Purandar region, assessing the associated economic losses, documenting farmers' existing management practices, and identifying knowledge gaps to inform targeted, sustainable disease management strategies applicable to semi-arid fig-producing regions across India.

Material and Methods

Study area and population

The survey was executed in prominent fig-producing villages within the Purandar, Daund, and Khedshivapur regions from August 2022 to August 2025, utilizing a standardized questionnaire in both English and Marathi. The regions constitute a significant fig cultivation zone distinguished by varied agricultural methods and various farmer populations. A survey of 100 orchards growing figs was conducted to evaluate the economic implications, yield, and prevalence of the disease.

Survey Design and Data Collection

A standardized questionnaire was conducted via direct interviews with farmers, focusing on cultivation traits, disease prevalence, yield efficacy, and market performance. The data variables encompass years of agricultural experience, farm size (in acres), total yield (in tonnes), types of diseases observed, frequency of field inspections, and dependence on expert consultation. Additionally, farmers used a five-point Likert scale to score the perceived price effect (1 being no effect and 5 being a severe impact). Disease presence was classified as rust-related, multiple (rust combined with blight or mite infestations) or absent. Management practices were documented based on the principal disease-control strategy employed: chemical pesticide application, traditional remedies, or no treatment.

Classification of groups

Farms were classified into three categories according to the stated economic impact of disease. Such as severely impacted, slightly impacted, and unaffected. These groups were utilized to evaluate yield characteristics, price effect scores and management techniques.

Statistical analysis

Descriptive statistics were calculated to summarize yield, area, and price-related variables. The variability in farmer experience and farm size was evaluated by standard deviation metrics. Correlation analysis (Pearson's r) was utilized to investigate the correlations among experience, yield, price effect and farm area. A one-way ANOVA assessed yield disparities across farms impacted by single versus multiple disease outbreaks, with a significance threshold established at $p < 0.05$. A multiple linear regression analysis was performed to assess the impact of farming experience and cultivated area on total production, utilizing the following model:

$$\text{Yield} = -10.91 + 0.228(\text{Years}) + 2.517(\text{Area})$$

Regression coefficients were analyzed to ascertain the relative impacts of farm size and experience on productivity.

Evaluation of spatial productivity

Productivity at the village level was assessed by compiling farm-specific data to ascertain total and per-acre yields for each site. A comparative analysis revealed high-efficiency and low-efficiency clusters to emphasize geographical diversity in cultivation results.

Results and Discussion

The survey of 100 fig orchards in the Purandar region indicated significant variability in farm features as well as widespread disease-induced economic disruption. Rust-related infections were the most common disease issue, impacting 75% of examined farms. This significant prevalence supports previous epidemiological evaluations that showed extensive fig rust occurrence in Maharashtra (Parthasarathy *et al.*, 2020; Avasthi *et al.*, 2023). An additional 25% of farms showed co-occurring diseases such as leaf blight and red spider mite infestations, demonstrating the complicated disease dynamics found in semi-arid orchard systems (Habib *et al.*, 2025). The average market disruption score was 3.25 on a 5-point scale, showing significant economic pressure due to disease prevalence (Table 2, Fig. 1).

Inspection frequency data revealed primarily proactive management behaviour: 50% of farmers monitored weekly, 25% monitored daily, and the remaining 25% followed alternative schedules (Fig. 1). Expert consultation was evenly distributed across four categories: agricultural extension agencies, pesticide dealers, undefined guidance, and no consultation, with each accounting for 25% of respondents. This homogeneous distribution reveals a substantial need for structured, science-based extension support for fig producers in the region.

According to the economic impact assessment, 87% of farms saw a major income reduction, 8% were mildly impacted, and 5% indicated no obvious effect (Table 3). Quantitative investigation revealed that severely afflicted farms produced an average output of 1.19 tonnes/ acre, compared to 0.97 tonnes/ acre on unaffected farms. A prevalent local market price of ₹ 50,000 per tonne results in an estimated yearly loss of ₹ 11,000 per acre for badly impacted orchards. The projected yearly income loss for seriously afflicted farms was ₹ 47,300 per farm (95% CI: ₹ 42,100-₹ 52,500), based on an average farm size of 4.29 acres. This represents a gross income decrease of nearly 23% from disease-free baseline yields. Disease severity had a significant negative connection with yield per acre ($r = -0.847$, $p < 0.001$; Table 3), indicating that fungal pathogenicity is the principal cause of productivity reduction. These findings are consistent with global evidence that major fungal infections cause 10-30% crop losses each year, with considerable downstream market implications (Savary *et al.*, 2019; Davies *et al.*, 2021).

The correlation study indicated significant correlations between significant farm variables. A negative association ($r = -0.881$) between farmer experience and perceived price effect suggests that experienced farmers are better equipped to handle market shocks. The negative association between experience and farm area ($r = -0.971$) indicates that newer entrants tend to manage larger holdings, presumably reflecting recent agricultural investments in the region. The positive correlation between yield and price effect ($r = 0.951$) is most likely due to premium or volume-based market dynamics, in which high-yielding farms use pricing arrangements to enhance the economic impact of disease-related production changes.

A one-way ANOVA revealed non-significant difference in yield between farms with single or multiple disease infections ($F = 0.807$, $p = 0.534$). This non-significant finding implies that active chemical control may somewhat mitigate the compounding impact of co-occurring pathogens, lowering the additive yield penalty that would otherwise be expected due to concurrent disease loads.

Multiple linear regression confirmed the predicted model: Yield = -10.91 + 0.228 (Years) + 2.517 (Area). The area coefficient (2.517) outperformed the experience coefficient (0.228), indicating that farm size is a far better predictor of total yield than farming experience. This conclusion suggests that economies of scale play a significant role in the current production system, which has practical consequences for extension programme design. Land consolidation techniques may produce bigger productivity gains than experience-based farmer training programmes alone.

The majority of farmers (61%) used chemical pesticides as their primary disease management method, with only a small percentage using traditional medicines or no treatment (Fig. 2). While chemical pesticides provide effective short-term management, over-reliance creates long-term hazards, such as pesticide residual issues in fruits destined for premium and export markets. Sustainably addressing rust disease necessitates a concerted move toward creating rust-tolerant cultivars and deploying regionally customized Integrated Pest Management (IPM) packages.

A village-level productivity analysis found significant spatial variability across the Purandar region. Sonori and Dive had the highest total production (126.2 tonnes from 22 farms; 0.89 tonnes/ acre), while Singapur (Lawande Vasti) had the highest per-acre efficiency (3.0 tonnes/ acre). Khor achieved a volume-efficiency balance (1.95 tonnes/ acre), indicating that the local conditions were optimal. High-efficiency villages like Singapur and Khor can serve as model sites for farmer-to-farmer knowledge transfer, resulting in targeted and replicable productivity gains across the region.

Table 1. Production statistics of fig worldwide from 2017-2021

S.No.	Year	Area harvested (ha)	Production (tonnes)	Yield (t/ ha)
1.	2017	5667	14462.78	2.55
2.	2018	5791	14882.73	2.57
3.	2019	5822	14566.58	2.50
4.	2020	5867	14637.37	2.49
5.	2021	5912	14695.56	2.49

Table 2. Economic impact of fungal diseases on fig

Economic Impact	Farms	Avg. yield (tonnes)	Avg. area (acres)	Yield per Acre (tonnes)	Avg. price effect
Severe impact	87	4.5	4.29	1.19	4.14
Slightly impacted	8	7.09	8.88	0.76	2.13
No effect	5	3.33	3	0.97	1

Table 3. Economic impact on average market price and average yield

Economic Impact	Farms	Avg. price effect	Avg. yield (tonnes)	Avg. area (acres)	Avg. yield/ acre
Decreased significantly	87	4.14	4.5	4.29	1.19
Slightly impacted	8	2.13	7.09	8.88	0.76
No noticeable effect	5	1	3.33	3	0.97

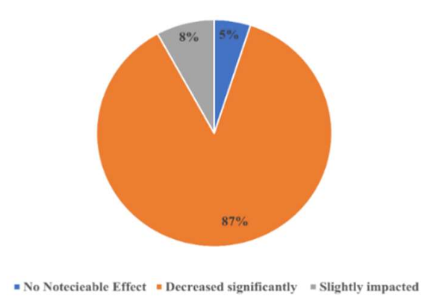

Fig. 1. Economic impact of fungal diseases on fig farms

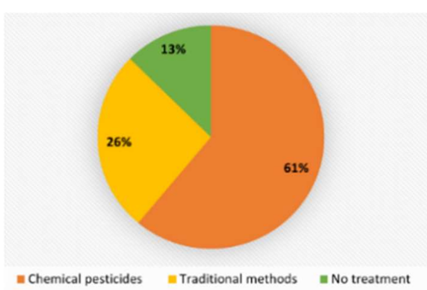

Fig. 2. Management practices and effectiveness at farmer's field

Conclusion

This research provides critical evidence-based insights for developing targeted interventions to improve the sustainability and profitability of fig cultivation. The findings back up urgent priorities such as developing rust-resistant varieties, expanding targeted extension services, implementing location-specific management strategies, and launching land consolidation projects. The long-term viability of fig cultivation as an economic option for smallholder farmers is crucial for addressing the complex issues identified by this thorough survey study, which requires coordinated efforts across research, extension, and policy domains.

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Conflict of Interest

The authors have no conflict of interest to declare.

Data Sharing

All relevant data are included in the manuscript.

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Research Article

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Symptomatology and evaluation of fungicides for management of black spot disease in Apple Ber

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ABSTRACT

The experiment was carried out during three consecutive years (2023–24, 2024–25 and 2025–26) on Apple ber. Among the various foliar pathogens affecting ber, the black spot caused by a fungus *Isariopsis personata* var. *zizyphi* producing regular black coloured fringed spots on the lower leaf surface. Spraying of Tebuconazole 50% + Trifloxystrobin 25% (WG) (0.1%) immediately after the appearance of the symptoms found most effective by reducing the leaf spot disease from 46.94 PDI to 17.20 PDI with yield of 64.39 kg/ tree.

Introduction

Ber (*Zizyphus mauritiana* Lamk.) commonly referred as Indian Jujube, Indian date and Chinese apple throughout the world play a major role in attaining the food security. Ber and apple ber are the highly recommended crops for the rainfed, arid and semi-arid regions which support the regular income of the small and marginal farmers all over India. Ber crop could be cultivated well in the marginal land with poor soil fertility and give good yield with less input (Mareeswari *et al*., 2012). This fruit crop required minimum inputs and highly recommended for Horti–Agri–Pastoral System of cultivation.

Though it is a highly remunerative crop, it's production is interrupted by many foliar diseases like, powdery mildew, rust, Alternaria, Cercospora, Phoma and black leaf spot (Kumar *et al.*, 2017). Among these, black spot fungus caused by the fungus *Isariopsis personata* var. *zizyphi* produces some black powdery growth on the lower surface of leaves. It reduced the photosynthetic area which resulted in yield reduction. In advance cases the pathogen causes necrotic patches and rotting on fruits. In severe cases the leaf spot incidence ranged up to 55% disease index. It is the most devastating disease in ber cultivation during monsoon season and causing economic losses.

Material and Methods

The experiment was carried out in the B2 block located of RRS, Aruppukottai, Tamil Nadu (Latitude 9.945º and Longitude 77.92º) over three consecutive years (2023–24, 2024–25 and 2025–26). Experiment was carried out on eight year old plants of Apple ber in randomized block design with seven treatments replicated thrice. Two trees are being maintained for each replication planted at 8 m x 8 m spacing. The treatments included Tebuconazole 50% + Trifloxystrobin 25% (WG) at 0.1% (T_1), Hexaconazole 5% EC at 0.1% (T_2), *Bacillus subtilis* (Bbv 57) at 0.5% (T_3), Azadirachtin (Neem oil, 1500 ppm) at 0.15% (T_4), garlic extract at 10% (T_5), Chlorothalonil at 0.2% (T_6) and control (T_7).

Normally the disease occurred during the second week of December every year. First spray was made immediately after the occurrence of the disease and second spray at 15 days intervals. Per cent Disease Index (PDI) is calculated on 15th day after the second spray. Fruits were harvested at physiological maturity. Average fruit yield per tree was recorded. In each year the spray was scheduled during the month of December during which the disease occurred. The pooled mean was derived and statistically analyzed.

Four branches, one from each side of the tree were selected. Ten leaves from each branch were examined. The disease severity was assessed using a rating scale ranging from 0 to 5 based on the extent of leaf area affected. A grade of 0 indicated no symptoms on the leaf. Grade 1 corresponded to 1–10% of the leaf area covered, while grade 2 represented 11–25% coverage. Grade 3 was assigned when 26–50% of the leaf area was affected, and grade 4 indicated 51–75% coverage. A grade of 5 denoted severe infection, with 76–100% of the leaf area covered. PDI was calculated as the formula given by Mckinney (1923).

$$PDI = \frac{\text{Sum of all numerical ratings}}{\text{Total number of leaves x Maximum grade in the scale}} \times 100$$

$$PROC = \frac{\text{PDI in control plot} - \text{PDI in treatment plot}}{\text{PDI in control plot}} \times 100$$

Results and Discussion

Symptoms and morphological confirmation of the pathogen

The pathogen produces round and blackish, fringed spots on the lower surface of the leaves (Fig. 1). These blackish growth merge with each other and lead to the reduction of chlorophyll content and finally affects the photosynthetic efficiency of the crop. The occurrence of the disease has been noticed in the ber orchards of RRS Aruppukottai during the past 15 years. It belongs to the Phylum: Ascomycota, Class: Dothidiomycetes and Order: Capnodiales Taxonomically it is highly related with sooty mould fungus (*Capnodium* spp.). While observing under microscope under the 40 X magnification, the mycelia are dark brown colored. Conidia are dark brown, septate (3-4 septations) and spindle shaped, broad in the middle portion and tapering towards the end (Fig. 2).

The disease was observed during the late December month of every monsoon season. Gupta *et. al.* (1977) recorded the occurrence of *Isariospsis* leaf spot for the first time from Haryana. Verma and Kumar (1992) discovered that cloudy weather with medium temperature during October-November is favourable for the disease development. Black spots which are sooty, tuft, circular to irregular in shape developed on the under surface of the leaves. In advance cases, the blackish mycelia covered the entire leaf surface and showed bleaching of chlorophyll and discolouration in turn resulted in yield loss.

Saha *et al.* (2022) reported that black spot disease affected approximately 70–80% of the leaf area. While observing under 40 X, magnification the conidiophores were long, multiseptate and dark brown. Conidia were light brown, multicellular, broader at the middle, tapering towards the end, bent and measuring 17-42 x 8.5-10.2 µm in size. This morphological observation is in conformity with our findings and assured that the pathogen is *Isariopsis personata* var *zizyphi* irrespective of the locations under study.

Fig. 1. Black, round and fringed spot of black spot disease

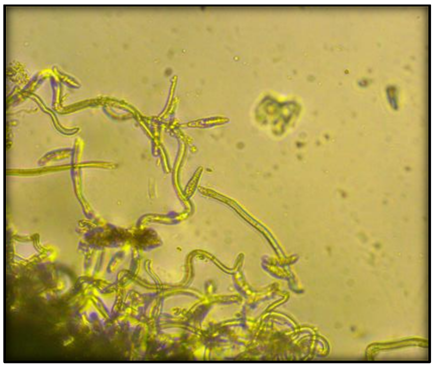

Fig. 2. Brownish, septate mycelia of *I. personata var. zizyph*i under 40 X magnification

Evaluation of fungicides, botanicals and bio control agents against black spot

The results presented in Table 1 indicated significant differences among treatments in reducing disease intensity (PDI), increasing percent disease control (PDC) and improving yield over the three consecutive years under field conditions. Among all treatments, Tebuconazole 50% + Trifloxystrobin 25% WG @ 0.1% consistently recorded the lowest PDI (26.11, 17.00 and 8.50) and the highest disease control (51.04%, 69.10% and 73.84%) during 2023–24, 2024–25 and 2025–26, respectively. It also produced the highest pooled yield (64.43 kg/ tree), making it the most effective treatment.

Chlorothalonil @ 0.2% was the next best treatment, showing comparatively low PDI and high disease control across all years, with a pooled yield of 62.16 kg/ tree. Neem oil @ 0.15% and Hexaconazole 5% EC showed moderate effectiveness, with intermediate PDI reduction and yield improvement.

Biological and botanical treatments such as *Bacillus subtilis* @ 0.5%, Garlic extract @ 10% and Neem oil @ 0.15% were less effective compared to chemical fungicides, recording higher PDI and lower yields.

Table 1. Effect of different fungicide treatments on the incidence of black spot

Treatments	2023-24			2024-25			2025-26			Pooled mean		
	PDI	PROC	Yield (kg/ tree)	PDI	PROC	Yield (kg/ tree)	PDI	PROC	Yield (kg/ tree)	PDI	PROC	Yield (kg/ tree)
T_1: Tebuconazole 50% + Trifloxystrobin 25% (WG) @ 0.1%	26.11^{a}	51.04	63.28	17.00^{a}	69.1	65.50^{a}	8.50^{a}	73.84	64.5^{a}	17.20^{a}	64.66	64.43^{a}
T_2: Hexaconazole 5% EC @ 0.1%	29.85^{bc}	44.03	58.38	27.50^{c}	50.91	60.70^{b}	13.75^{b}	57.69	59.31^{c}	23.70^{ab}	50.88	59.46^{ab}
T_3: *Bacillus subtilis* (Bbv 57) @ 0.5%	33.39^{d}	37.39	47.43	44.00^{e}	20.00	51.25^{d}	20.15^{c}	38.00	46.25^{e}	32.51^{c}	31.80	48.31^{cd}
T_4: Neem oil (Azadirachtin) @ 5%	28.17^{ab}	47.18	55.35	36.00^{d}	34.55	57.50^{c}	18.25^{c}	43.85	49.48^{d}	27.47^{bc}	41.86	54.11^{bc}
T_5: Garlic extract @ 10%	32.23^{cd}	39.40	47.77	45.50^{e}	18.18	49.50^{d}	24.35^{d}	25.08	38.34^{f}	34.03^{c}	27.55	45.20^{d}
T_6: Chlorothalonil @ 0.2%	27.97^{ab}	47.55	60.32^{ab}	24.00^{b}	56.36	63.90^{a}	10.00^{a}	69.23	62.25^{b}	20.66^{ab}	57.71	62.16^{a}
T_7: Control	53.33^{e}	-	42.43^{d}	55.00^{f}	-	40.00^{e}	32.50^{e}	-	27.57^{g}	46.94^{d}	-	36.00^{e}
CD (0.05)	2.54		5.38	1.63		1.77	1.55		0.55	7.99		5.99
SEd	1.16		2.47	0.75		0.89	3.38		1.21	3.67		2.75
CV (%)	4.32		5.64	2.53		1.79	10.44		1.37	15.53		6.375

Overall, the pooled data clearly demonstrated spraying of Tebuconazole 50 % + Trifloxystrobin 25 % (WG) performed well in the management of black spot disease of ber reducing the leaf spot from 46.94 to 17.20 percentage (pooled data) which was followed by Chlorothalonil (0.2%) (20.66 PDI) and Hexaconazole (0.1%) (23.70 PDI) compared to control (46.94 PDI). The above best performing fungicide recorded the highest yield (64.43 kg/ tree) compared to control (36 kg/ tree).

Kumar *et al*. (2003) also attempted to manage the black spot disease in the ber cultivar Gola. Spraying of Carbendazim 50 WP (0.1%) @ 10 litre solution per plant during the early November was found to be most effective in the management of the disease in turn showing 77.21% reduction over control (PDC) followed by Mancozeb (0.2%) (54.78 PDC) and neem powder, Nimuri (54.37 PDC). Kumar *et al.* (2017) concluded that spraying of Propiconazole @ (0.1%) immediately after the occurrence of the disease and at 15 days interval recorded 73% reduction over control and increased the yield from 40 kg/ tree to 88.80 kg/ tree of cv. Gola. Zhang *et al.* (2021) investigated the antifungal activity of the fungicide Propiconazole against the fungus *Penicillium digitatum*. They reported that it inhibits the fungal enzyme 14-α-demethylase (CYP51), a key component of the cytochrome P450–dependent pathway responsible for ergosterol synthesis, an essential sterol in the fungal cell membrane. Singh *et al*. (2024) recorded minimum disease intensity (14%) in the plots sprayed with Propiconazole 25 EC@ (0.1%) followed by Carbendazim @ (0.1%) and Copper oxychloride @ (0.3%) (26%) respectively. Likely, the trees sprayed with Propiconazole recorded the maximum yield (63.83 kg/ tree) followed by Carbendazim (60.65 kg/ tree) compared to control (49.41 kg/ tree).

Conclusion

Ber black spot is a major disease causing significant yield losses in ber-growing regions of India, primarily by reducing the effective photosynthetic leaf area due to characteristic black fungal growth on the leaf surface. The present study clearly demonstrated that fungicidal treatments effectively manage the disease. Among the treatments, Tebuconazole 50% + Trifloxystrobin 25% (WG) proved most effective, recording a 64.66% reduction over control and the highest yield (64.43 kg/ tree). This was followed by Chlorothalonil (57.71% disease control) and Hexaconazole (50.88% disease control). Overall, the results highlight the superiority of combination fungicide in controlling ber black spot and improving productivity compared to untreated plants.

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Conflict of Interest

The authors of this paper do not have any conflict of interest.

Data Sharing

All relevant data are within the manuscript.

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Research Article

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Field evaluation of fungicides against leaf and fruit spot diseases of pomegranate (*Punica granatum* L.)

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ABSTRACT

A two-year field experiment was conducted during Mrig Bahar to evaluate the efficacy of different fungicides against Anthracnose and Alternaria leaf and fruit spot diseases of pomegranate (*Punica granatum* L.). Three sprays were applied at 15-day intervals and disease observations on leaves and fruits were recorded 15 days after the final spray. Results revealed that the ready-mix fungicide Tebuconazole 50% + Trifloxystrobin 25% WG was found significantly effective treatment, recording the lowest PDI of 14.03% on leaves and 13.96% on fruits. Plants treated with Tebuconazole 50% + Trifloxystrobin 25% WG showed minimum yield loss.

Introduction

Pomegranate (*Punica granatum* L.) is one of India's fastest-growing commercially important fruit crops, valued for its export potential, drought tolerance and high nutritional quality. Globally, India ranks first in pomegranate cultivation with an area of 0.224 million hectares and production of 2.842 million metric tonnes during 2023-24 (Anonymous, 2024a). During the financial year 2023-24, India exported 72,011 metric tonnes of pomegranate valued at USD 69.08 million to countries such as the United Arab Emirates, Bangladesh, Nepal, Netherlands, Saudi Arabia, Sri Lanka, Thailand, Bahrain, and Oman (Anonymous, 2024b). The crop is cultivated widely in Maharashtra, Karnataka, Andhra Pradesh, Gujarat, Rajasthan and Tamil Nadu. Maharashtra is considered the pomegranate basket of India, contributing nearly two-thirds of the country's cultivated area (Anonymous, 2009). In Gujarat, pomegranate occupies an area of 43.53 thousand hectares with an annual production of 0.629 million tonnes and productivity of 14.45 tonnes per hectare (Anonymous, 2024b). It is intensively grown in northern districts such as Kutch, Banaskantha, Morbi, and Mehsana, and is now expanding rapidly in the Saurashtra region.

The increasing popularity of pomegranate cultivation in Gujarat is mainly due to the high economic returns from the crop. Despite its expanding cultivation, productivity in Gujarat has not increased proportionately because of severe damage caused by insect pests and diseases. Among fungal diseases, anthracnose caused by *Colletotrichum gloeosporioides* and leaf spot caused by *Alternaria alternata* are the most destructive (Berbegal *et al.*, 2014). These diseases affect leaves and fruits, reducing yield, market quality and causing considerable economic losses to growers (Dev *et al.*, 2017). Therefore, systematic studies on the in vivo effectiveness of suitable chemical fungicides are urgently needed to develop efficient management strategies for controlling Anthracnose and Alternaria diseases in pomegranate under Gujarat conditions.

Material and Methods

The study was conducted at the Department of Plant Pathology, College of Agriculture, Junagadh Agricultural University (JAU), Junagadh, Gujarat (latitude: 21.49519ºN and longitude: 70.42207ºE) during 2018-19 and 2019-20. Field experiments were undertaken during Mrig Bahar (November-December) in a seven-year-old pomegranate cv. Bhagwa. Seven fungicidal treatments along with an untreated control were evaluated against anthracnose and Alternaria diseases. The trial was laid out in a Randomized Block Design (RBD) with three replications, keeping one plant per replication. Sprays were initiated at the onset of disease symptoms. A total of three sprays were applied at an interval of 15 days using a manually operated high-volume knapsack sprayer. Disease intensity was recorded on 40 leaves per plant (10 leaves from each direction) and 20 fruits per plant (5 fruits from each direction) using a 0-5 disease rating scale. The per cent disease index (PDI) was calculated according to Wheeler (1969) and the data obtained was transformed into angular transformations and analyzed statistically.

$$\text{PDI} = \frac{\sum \text{Numerical ratings observed}}{\text{No. of units observed} \times \text{maximum grade or rating}} \text{x100}$$

Where, Units = leaves/ fruits

Per cent disease control (PDI) was also computed based on reduction in PDI over untreated control. The recorded data were subjected to angular transformation before statistical analysis. Recoded the rating/ per cent incidence reaction (Table 1) as described by Archana, 2012 and Jagtap *et al.,* 2015.

Table 1. Disease rating/ per cent incidence reaction

Disease rating (Grade)	Per cent area of infection	
	On fruits	On leaf
0	No infection	No infection
1	1-10	1-5% leaf area covered
2	11-25	6-10% leaf areas covered
3	26-50	11-25% leaf areas covered
4	51-75	26-50% leaf areas covered
5	> 75	> 50% leaf areas covered

Yield losses due to Anthracnose and Alternaria were estimated simultaneously with fungicide evaluation. The treatment found most effective in disease suppression was considered as disease-free check. Fruit yield of each plant under all treatments was recorded and compared with the check treatment. Per cent yield loss was calculated using the formula suggested by Colpauzos *et al*. (1976):

$$\text{Yield loss (\%)} = \frac{\text{Yield of check} - \text{Yield of treatment}}{\text{Yield of check}} \times 100$$

Table 2. List of treatments used for field evaluation

S.No.	Treatments	Concentration (%)	Qty. per 10 liter water
1.	Propineb 70% WP	0.2	28.57 g
2.	Mancozeb 75% WP	0.2	26.66 g
3.	Iprobenfos 48% EC	0.02	4.16 ml
4.	Picoxystrobin 22.52% SC	0.02	8.88 ml
5.	Difenoconazole 250 EC	0.02	8 ml
6.	Epoxiconazole 50 g/ l + Pyraclostrobin 133 g/ l	0.01	5.81 ml
7.	Tebuconazole 50% + Trifloxystrobin 25% WG	0.025	3.33 g
8.	Control	-	-

Results and Discussion

A two-year field study was conducted during Mrig Bahar season to evaluate the comparative efficacy of different fungicides against Anthracnose and Alternaria diseases of pomegranate. Seven fungicidal treatments along with an untreated control were tested.

Effect of fungicides on leaf spot disease

The analyzed data indicated that all fungicidal treatments significantly reduced leaf spot severity over untreated control during both years of experimentation. Among the treatments, the ready-mix fungicide Tebuconazole 50%+ Trifloxystrobin 25% WG at 0.025 per cent proved most effective, recording the lowest pooled PDI of 14.03 per cent. This treatment was statistically at par with Epoxiconazole 50 g/ l + Pyraclostrobin 133 g/ l at 0.01 per cent, which recorded 16.01 per cent. PDI.

These were followed by Picoxystrobin 22.52% SC at 0.02 per cent with 19.06 per cent PDI (Table 3).

Moderate disease reduction was observed with Difenoconazole 25 EC (21.50%), Iprobenfos 48 EC (24.06%) and Propineb 70 WP (27.50%). Mancozeb 75 WP at 0.2 per cent was found least effective among the tested fungicides, though still superior to the untreated control. In terms of disease control over untreated control, Tebuconazole + Trifloxystrobin registered the highest reduction (56.62%), followed by Epoxiconazole + Pyraclostrobin (50.49%). Mancozeb recorded the lowest disease reduction (7.47%).

These findings are in agreement with earlier reports of Jayalakshmi (2015) and Archana and Jamadar (2014), who also observed lower efficacy of Mancozeb against leaf and fruit spot diseases of pomegranate. However, Shreeshail *et al.* (2016) reported Mancozeb (0.2%) as effective against *Colletotrichum gloeosporioides*, indicating possible variation due to environmental conditions, pathogen virulence, or spray schedules. effect of fungicides on fruit spot disease

Table 3. Effect of different fungicides on leaf spots of pomegranate

S.No.	Treatment	Conc. (%)	2018-19	2019-20	Pooled	PDI (%)
1.	Propineb 70% WP	0.2	32.66 (29.17)*	32.57 (25.83)	32.61 (27.50)	14.95
2.	Mancozeb 75% WP	0.2	34.23 (31.67)	34.55 (28.17)	34.39 (29.92)	7.47
3.	Iprobenfos 48% EC	0.02	31.32 (27.08)	29.45 (21.04)	30.39 (24.06)	25.58
4.	Picoxystrobin 22.52% SC	0.02	28.55 (22.92)	26.20 (15.21)	27.38 (19.06)	41.04
5.	Difenoconazole 250 EC	0.02	29.72 (24.67)	25.61 (18.33)	27.67 (21.50)	33.51
6.	Epoxiconazole 50 g/ l + Pyraclostrobin 133 g/ l	0.01	26.37 (19.79)	22.88 (12.22)	24.62 (16.01)	50.49
7.	Tebuconazole 50% + Trifloxystrobin 25% WG	0.025	24.39 (17.22)	23.63 (10.83)	24.01 (14.03)	56.62
8.	Control	-	35.83 (34.33)	34.86 (30.33)	35.34 (32.33)	0.00
	SEm±		1.36	1.04	0.86	
	CD at 5%		4.14	3.16	2.49	
	CV (%)		7.78	6.28	7.11	
	Y					
	SEm±				0.43	
	CD at 5 %				1.24	
	Y x T					
	SEm±				1.21	
	CD at 5 %				NS	

*Data outside the parentheses are arcsine transformed, whereas inside are re-transformed values

Effect of fungicides on fruit spot disease

The data revealed (Table 4) that all fungicidal treatments significantly reduced fruit spot severity compared with untreated control. The lowest pooled PDI of 13.96 per cent was recorded with Tebuconazole 50% + Trifloxystrobin 25% WG at 0.025 per cent, followed by Epoxiconazole 50 g/ l + Pyraclostrobin 133 g/ l at 0.01 per cent (17.22%) and Picoxystrobin 22.52% SC at 0.02 per cent (21.11%). Difenoconazole 25 EC, Iprobenfos 48 EC and Mancozeb 75 WP recorded 24.69, 27.61 and 30.75 per cent PDI, respectively, Propineb 70 WP was found least effective in reducing fruit spot severity among all tested fungicides. The highest fruit spot incidence was observed in untreated control plants with 33.17 per cent PDI. Maximum disease control over untreated control was obtained with Tebuconazole + Trifloxystrobin (57.91%), followed by Epoxiconazole + Pyraclostrobin (48.08%), Propineb showed minimum disease control.

The superior performance of Tebuconazole + Trifloxystrobin may be attributed to the combined systemic and protective action of triazole and strobilurin groups, which inhibit fungal growth and sporulation effectively. However, under the present field conditions, these fungicides were comparatively less effective than the newer combination fungicides. Among all treatments, Tebuconazole 50% + Trifloxystrobin 25% WG consistently recorded the lowest disease intensity on leaves and fruits and provided maximum disease control, followed by Epoxiconazole + Pyraclostrobin. Therefore, these fungicides can be recommended as effective options for integrated disease management of pomegranate under field conditions.

Similar results were earlier reported by Ekabote and Narayanswarmy (2019) against anthracnose of pomegranate, Ann *et al.* (2017) against anthracnose of black pepper caused by *C. gloeosporioides*, and Ginoya and Gohel (2015) against *Alternaria alternata* causing chilli fruit rot. Tziros *et al.* (2017) also reported Iprobenfos (Kitazin) as effective against pomegranate anthracnose, while Jamadar and Patil (2007) found Difenoconazole 25 EC highly effective.

Table 4. Effect of different fungicides on fruit spots of pomegranate

S.No.	Treatment	Conc. (%)	2018-19	2019-20	Pooled	PDI (%)
1.	Propineb 70% WP	0.2	35.15 (33.17)	32.36 (29.83)	33.75 (31.50)	5.03
2.	Mancozeb 75% WP	0.2	33.41 (30.33)	32.46 (31.17)	32.93 (30.75)	7.29
3.	Iprobenfos 48% EC	0.02	31.39 (27.17)	33.57 (28.13)	32.48 (27.61)	16.76
4.	Picoxystrobin 22.52% SC	0.02	26.54 (20.00)	30.02 (22.22)	28.28 (21.11)	36.35
5.	Difenoconazole 250 EC	0.02	29.42 (24.17)	32.31 (25.21)	30.86 (24.69)	25.55
6.	Epoxiconazole 50 g/ l + Pyraclostrobin 133 g/ l	0.01	24.42 (17.22)	27.38 (17.22)	25.90 (17.22)	48.08
7.	Tebuconazole 50% + Trifloxystrobin 25% WG	0.025	22.04 (14.17)	23.53 (13.75)	22.79 (13.96)	57.91
8.	Control	-	35.76 (34.17)	34.59 (32.17)	35.18 (33.17)	0.00
		SEm±	1.28	1.11	0.85	
		CD at 5%	3.88	3.36	2.45	
		CV (%)	7.44	6.22	6.84	
		Y				
			SEm±		0.42	
			CD at 5 %		NS	
		Y x T				
			SEm±		1.20	
			CD at 5 %		NS	

*Data outside the parentheses are arcsine transformed, whereas inside are re-transformed values

Assessment of yield losses in pomegranate due to Anthracnose and Alternaria

Amongst fungal diseases, fruit spot caused by *Colletotrichum gloeosporioides* results in rotting of pomegranate fruits within a week of infection, causes drastic reduction in the yield as well as marketability of the produce. This study was undertaken to evaluate the relative efficacy of different fungicides for management of anthracnose of pomegranate and affect fruit yield. The fungicide which found effective in suppression of diseases effectively in field (Tebuconazole 50% + Trifloxystrobin 25% WG) was used to maintain disease free condition (control). The fruit yield of each plant was recorded in each treatment and compare with the control. Per plant production recorded for different treatment and yield loss was calculated as described in materials and methods. Among all tested fungicides highest fruit yield was recorded in Tebuconazole 50% + Trifloxystrobin 25% WG (15.75 kg/ plant) followed by Epoxiconazole 50 g/ l + Pyraclostrobin 133 g/ l with 13.12 kg/ plant which was at par with Picoxystrobin 22.52% SC with 12.25 kg/ plant fruit yield (Table 5). The next on par yield levels were obtained in Difenoconazole 250 EC, Iprobenfos 48 % EC and Propineb 70% WP with 11.22, 10.36 and 9.65 kg/plant fruit yield.

Maximum yield loss was recorded in untreated control (47.74%) which was followed by Mancozeb 75% WP (44.55%) followed by Propineb 70% WP (38.72). yield loss recorded in iprobenfos 48 % EC, Difenoconazole 250 EC and Picoxystrobin 22.52% SC were 34.26%, 8.80% and 22.24%, respectively. Minimum yield loss was recorded in Epoxiconazole 50 g/ l + Pyraclostrobin 133 g /l (16.74%). Jayalakshmi (2015) recorded the effect of fungicides against *Colletotrichum gloeosporioides* causing anthracnose of pomegranate and found iprobenfos (0.2%), carbendazim (0.2%), Difenconazole (0.1%), Captan (0.3%) and Mancozeb (0.3%) were at par in yield of pomegranate. Effectiveness of Difenconazole was recorded by Jamadar and Patil (2007).

Table 5. Effect of different fungicides on fruit yield of pomegranate

S.No.	Treatment	Conc. (%)	2018-19 (kg/ plant)#	2019-20 (kg/ plant)#	Pooled	Yield loss (%)
1.	Propineb 70% WP	0.2	17.34 (8.99)*	18.72 (10.32)*	18.03 (9.65)	38.72
2.	Mancozeb 75% WP	0.2	16.66 (8.29)	17.60 (9.18)	17.13 (8.74)	44.55
3.	Iprobenfos 48 % EC	0.02	18.06 (9.69)	19.34 (11.02)	18.70 (10.36)	34.26
4.	Picoxystrobin 22.52% SC	0.02	20.08 (11.79)	20.85 (12.71)	20.46 (12.25)	22.24
5.	Difenoconazole 250 EC	0.02	18.94 (10.60)	20.07 (11.83)	19.50 (11.22)	28.80
6.	Epoxiconazole 50 g/ l + Pyraclostrobin 133 g/ l	0.01	20.82 (12.67)	21.60 (13.57)	21.21 (13.12)	16.74
7.	Tebuconazole 50% + Trifloxystrobin 25% WG	0.025	23.16 (15.49)	23.59 (16.02)	23.37 (15.75)	0.00
8.	Control	-	16.24 (7.89)	16.99 (8.58)	16.62 (8.23)	47.74
	SEm±		0.93	0.91	0.65	
	CD at 5%		2.82	2.76	1.88	
	CV (%)		8.51	7.95	8.23	
	Y					
	SEm±				0.33	
	CD at 5%				NS	
	Y x T					
	SEm±				0.92	
	CD at 5%				NS	

*Data outside the parentheses are arcsine transformed, whereas inside are re-transformed values
#Mean of three replications

Conclusion

In conclusion, the two-year study showed that fungicidal sprays effectively managed Anthracnose and Alternaria diseases in pomegranate during Mrig Bahar and improved yield. Tebuconazole 50% + Trifloxystrobin 25% WG at 0.025% was the most effective, reducing disease intensity and increasing fruit yield (15.75 kg/ plant). Its superior performance highlights the benefit of combining systemic and protective fungicides. Therefore, Tebuconazole + Trifloxystrobin followed by Epoxiconazole + Pyraclostrobin is recommended for effective disease management under field conditions.

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Conflict of Interest

The authors declare no conflict of interest.

Data Sharing

All relevant data are within the manuscript.

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Research Article

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Compatibility evaluation of *Trichoderma* species with plant extracts under *In-vitro* conditions

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ABSTRACT

The present study evaluated the compatibility of botanical insecticides with three *Trichoderma* species, *T. hamatum*, *T. harzianum* and *T. asperellum* to assess their suitability for integrated disease management. Seven botanicals marigold, karanj, lantana, parthenium, datura, periwinkle and neem were tested at 5% and 10% concentrations. All botanicals exhibited high compatibility with *T. hamatum* and *T. asperellum*, with mycelial growth ranging from 64.17 mm to 88.50 mm and growth inhibition below 30%. For *T. harzianum*, most botanicals at 5% concentration were compatible, while karanj, datura, neem and lantana at 10% showed moderate compatibility, with growth inhibition ranging from 33.15% to 44.26%. These findings suggest that botanical insecticides, particularly at lower concentrations, can be safely integrated with *Trichoderma* spp. without compromising their growth, offering a promising approach for sustainable crop protection.

Introduction

In recent years, growing concerns over the excessive and indiscriminate use of synthetic pesticides have encouraged the search for safer alternatives in plant disease management. Although chemical control has been effective to some extent, its continuous application often leads to resistance development in pathogens, a decline in beneficial soil microflora, and negative impacts on human health and the environment (Meena *et al.*, 2020; Hobbelen *et al.*, 2014). As a result, botanicals derived from medicinal and aromatic plants such as neem (*Azadirachta indica*), garlic (*Allium sativum*), tulsi (*Ocimum sanctum*) and onion (*Allium cepa*) have emerged as eco-friendly options due to their antifungal and antibacterial properties.

Trichoderma species are widely recognized as effective biocontrol agents that suppress a broad range of soil-borne plant pathogens through mechanisms such as competition for nutrients and space, mycoparasitism, secretion of antifungal metabolites and induction of plant defense responses (Sood *et al.*, 2020; Vinale *et al.*, 2013). Beyond pathogen suppression, they also promote nutrient uptake, plant growth and yield improvement (Campos *et al.*, 2020). Because of these multifaceted roles, Trichoderma has been widely adopted in seed treatment, soil application and bioformulations marketed as biopesticides and biofertilizers (Kumar *et al.*, 2014). However, the combined use of Trichoderma with botanicals requires careful evaluation. Some plant extracts can be compatible and enhance its efficacy, while others may inhibit its growth and colonization, reducing its effectiveness as a biocontrol agent (Bagwan, 2010).

Since plant extracts are increasingly used in sustainable agriculture, understanding their compatibility with Trichoderma is essential for integrated disease management. The present study was therefore undertaken to evaluate the *In-vitro* compatibility of *Trichoderma hamatum*, *T. harzianum* and *T. asperellum* with selected plant extracts commonly used as botanicals in plant protection practices.

Material and Methods

The study was conducted in the Trichoderma Laboratory, Agricultural Research Station, Ummedganj, Kota, Rajasthan, India during 2025. Compatibility tests were performed using the poisoned food technique (Nene and Thapliyal, 1993) in a Completely Randomized Design (CRD) with three replications. The treatments comprised T_1 = Marigold (*Tagetes erecta*), T_2 = Karanj (*Pongamia glabra*), T_3 = Lantana, (*Lantana camera*), T_4 = Parthenium (*Parthenium hysetrophorus*), T_5 = Datura (*Datura stramonium*), T_6 = Periwinkle (*Vinca rosea*) and T_7 = Neem (*Azadirachta indica*) each at 5% and 10% along with T_8 = Control.

Potato Dextrose Agar (PDA) medium was sterilized and cooled to approximately 40°C before amending with the required concentrations of plant extracts. About 20 ml of amended PDA was poured into 9 cm sterilized Petri plates. Plates without botanicals served as controls. Nine insecticides were tested at recommended concentrations. A 5 mm mycelial disc from a 7-day-old culture of *Trichoderma hamatum*, *T. harzianum*, or T. *asperellum* was aseptically transferred to the center of each Petri plate. Plates were incubated at 25 ± 2°C and radial mycelial growth was measured after 7 days.

Mycelial growth inhibition was calculated using the formula advocated by Vincent (1947) as given below.

$$\text{Percent growth inhibition (PGI)} = \frac{C-T}{C} \times 100$$

Where, I = percent inhibition, C = colony diameter in control, and T = colony diameter in treatment.

The nature of compatibility is classified based on the percentage of inhibition: treatments showing 0–30% inhibition are considered highly compatible, those with 30.1–60% inhibition are moderately compatible, 60.1–90% inhibition indicates slight compatibility and values exceeding 90% are regarded as non-compatible (Saha *et al*., 2023). Data on mycelial growth and inhibition percentage were subjected to arcsine transformation before analysis of variance (ANOVA). Treatment means were compared at a 5% significance level.

Results and Discussion

Effect of plant extracts on Trichoderma hamatum

The *In-vitro* study revealed that all tested plant extracts exhibited high compatibility with *T. hamatum*, as indicated by low levels of mycelial growth inhibition (<30%). The radial growth of the fungus ranged from 64.17 mm to 85.50 mm, confirming that none of the botanicals significantly suppressed fungal growth. Among the treatments, periwinkle (*Catharanthus roseus*) and marigold (*Togetes erecto*) extracts showed the least inhibitory effect, recording minimal inhibition of 5.00-12.22%, followed by parthenium (*Parthenium hysterophorus*) with inhibition below 10% even at higher concentration. This suggests excellent compatibility with *T. hamatum*, moderate inhibition was observed with extracts of lantana (*Lantana camara*), datura (*Datura stramonium*) and neem (*Azadirachta indica*), where inhibition ranged between 16-25%. The highest inhibition was recorded in karanj (*Porigamia pinnata*) extract (28.70% at 10%), but it still remained within the highly compatible category (Table 1, Fig.1). Dubey *et al.* (2007) observed that botanicals like neem and lantana showed minimal inhibition on Trichoderma while effectively suppressing pathogens. Singh (2012) also emphasized that botanical extracts can be combined with biocontrol agents due to their selective toxicity.

Table 1. Effect of plant extracts on *T. hamatum*

Plant extracts	Radial growth* (mm)		Per cent inhibition (%)**		Nature of compatibility
	5%	10%	5%	10%	
T_1 : Marigold (*Tagetes erecta*)	85.00	79.00	5.56 (13.62)	12.22 (20.45)	H H
T_2 : Karanj (*Pongamia glabra*)	71.67	64.17	20.37	28.70	H

			(26.82)	(32.39)	H
T_3 : Lantana (*Lantana camera*)	81.17	72.83	9.81	19.07	H
			(18.24)	(25.89)	H
T_4: Parthenium (*Parthenium hysetrophorus*)	84.00	82.00	6.67	8.89	H
			(14.93)	(17.32)	H
T_5 : Datura (*Datura stramonium*)	74.50	67.50	17.22	25.00	H
			(24.51)	(29.99)	H
T_6 : Periwinkle (*Vinca rosea*)	85.50	83.17	5.00	7.59	H
			(12.88)	(15.98)	H
T_7 : Neem (*Azadirachta indica*)	74.83	68.67	16.85	23.70	H
			(24.23)	(29.13)	H
T_8 : Control	90.00	90.00	0.00	0.00	-
			(0.00)	(0.00)	
SEm±	0.37	0.33	0.55	0.45	-
CD (5%)	1.11	0.99	1.66	1.34	-

*Average of three replications; **Figures in parentheses are Arc sine transformed
H: Highly compatible, M: Moderately compatible, S: Slightly compatible N: Non-Compatible

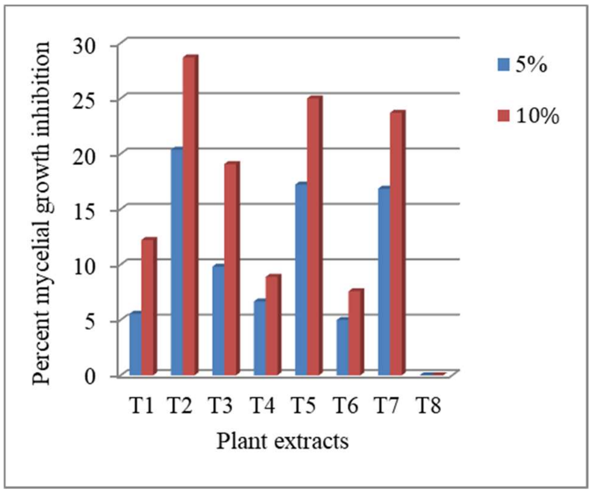

Fig. 1. *In-vitro* evaluation of per cent mycelial growth inhibition of *T. hamatum* against plant extracts

Effect of plant extracts on Trichoderma harzianum

The *In-vitro* evaluation demonstrated that *T. harzianum* exhibited high compatibility with most plant extracts at lower concentration (5%), where marigold, karanj, lantana, parthenium, datura, periwinkle and neem supported substantial mycelial growth (81.17-69.83 mm) with less than 30% inhibition, indicating their suitability for combined use (Table 2, Fig. 2). However, at higher concentration (10%), certain extracts such as karanj, lantana, datura and neem exhibited moderate compatibility, reducing fungal growth (60.17-50.17 mm) and increasing inhibition (33.15-44.26%), suggesting a dose-dependent effect of plant metabolites on fungal growth.

Among all treatments, periwinkle and marigold extracts showed minimal inhibitory effect even at higher concentrations, whereas neem (10%) recorded the highest inhibition (44.26%), though still within a tolerable range. Therefore, the findings indicate that most botanicals, particularly at lower concentrations, are compatible with *T. horzionum* and can be effectively integrated into eco-friendly disease management strategies. Similar observations have been reported by Dubey *et al.* (2007) and Sharma *et al.* (2012), who documented that botanicals such as neem and lantana exert minimal inhibitory effects on Trichoderma, supporting their combined application in integrated disease management programs.

Table 2. Compatibility of plant extracts with *T. harzianum*

Plant extracts	Radial growth* (mm)		Per cent inhibition (%)**		Nature of compatibility
	5%	10%	5%	10%	
T_1 : Marigold (*Tagetes erecta*)	79.67	73.33	11.48 (19.77)	18.52 (25.48)	H
					H
T_2 : Karanj (*Pongamia glabra*)	71.00	59.83	21.11 (27.34)	33.52 (35.37)	H
					M
T_3 : Lantana (*Lantana camera*)	73.00	60.17	18.89 (25.74)	33.15 (35.15)	H

					M
T_4: Parthenium (*Parthenium hysetrophorus*)	77.33	71.50	14.07 (22.02)	20.56 (26.94)	H
					H
T_5: Datura (*Datura stramonium*)	71.50	59.83	20.56 (26.94)	33.52 (35.37)	H
					M
T_6 : Periwinkle (*Vinca rosea*)	81.17	75.33	9.81 (18.16)	16.30 (23.79)	H
					H
T_7 : Neem (*Azadirachta indica*)	69.83	50.17	22.41 (28.25)	44.26 (41.70)	H
					M
T_8 : Control	90.00	90.00	-	-	-
SEm±	0.58	0.46	0.79	0.55	-
CD (5%)	1.75	1.37	2.38	1.66	-

*Average of three replications; **Figures in parentheses are Arc sine transformed
H: Highly compatible, M: Moderately compatible, S: Slightly compatible N: Non compatible

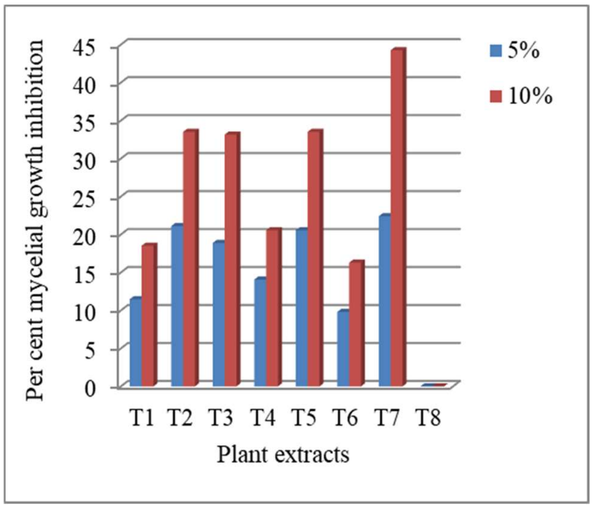

Fig. 2. In-vitro evaluation of per cent mycelial growth inhibition of *T. harzianum* against plant extracts

Effect of plant extracts on Trichoderma asperellum

The *In-vitro* evaluation indicated that *T. asperellum* exhibited high compatibility with all tested plant extracts, as reflected by substantial mycelial growth (67.33-88.50 mm) and low per cent inhibition (<30%) across both 5% and 10% concentrations (Table 3, Fig. 3). Among the treatments, periwinkle and marigold extracts showed the least inhibitory effect, recording minimal inhibition of 1.67-10.74% and 3.15-10.37%, respectively, followed by lantana and parthenium. Even at higher concentration (10%), extracts of karanj, datura and neem caused only mild inhibition (18.70-25.19%), confirming their compatibility. Comparative evaluation among three species revealed that *T. asperellum* was the most tolerant, followed by *T. hamatum*, while *T. harzianum* showed relatively higher sensitivity at increased concentrations. The results clearly suggest that these botanicals can be safely integrated with *T. asperellum* in eco-friendly disease management programs. Similar findings were reported by Dubey *et al.* (2007) and Sharma *et al.* (2012). They also observed that various botanicals extracts has minimal inhibitory effects on Trichoderma, supporting their combined use in integrated disease management strategies.

These findings indicate that plant extracts, particularly at lower doses, can be integrated with *Trichoderma* spp. without affecting their growth, thus holding promise for eco-friendly disease management strategies. Marigold and periwinkle are known to release root exudates that are generally supportive of microbial growth, including *Trichoderma* spp. These exudates are non-toxic and do not interfere with the fungal metabolism, unlike certain other plant species that may release inhibitory substances. Although direct studies on these specific plants are limited, existing research indicates that their chemical profiles create a favorable environment for mutual interaction and colonization by Trichoderma (Bharti *et al.*, 2024). Neem extract showed mycelial growth inhibition may be because of it contain azadirachtin that disrupts cellular integrity and enzyme pathways, leading to halted spore germination and hyphal growth (Almeida *et al.*, 2017). These findings are in confirmatory with the earlier reports of several workers (Bhagwan, 2010; Waware, 2015; Maheshwari, 2014; Tapwal *et al.*, 2012). In an integrated disease management framework, *T. asperellum* can work synergistically with compatible fungicides to reduce pathogen load while minimizing chemical fungicide dependence (Kumari *et al.,* 2025).

Table 3. Effect of plant extracts on *T. asperellum*

Plant extracts	Radial growth* (mm)		Per cent inhibition (%)**		Nature of compatibility
	5%	10%	5%	10%	
T_1 : Marigold (*Tagetes erecta*)	87.17	80.67	3.15 (10.16)	10.37 (18.76)	H H
T_2 : Karanj (*Pongamia glabra*)	74.83	67.33	16.85 (24.23)	25.19 (30.12)	H H
T_3 : Lantana (*Lantana camera*)	84.67	78.50	5.93 (14.03)	12.78 (20.94)	H H
T_4: Parthenium (*Parthenium hysetrophorus*)	83.67	76.17	7.04 (15.37)	15.37 (23.07)	H H
T_5: Datura (*Datura stramonium*)	77.33	70.33	14.07 (21.99)	21.85 (27.87)	H H
T_6 : Periwinkle (*Vinca rosea*)	88.50	80.33	1.67 (6.95)	10.74 (19.11)	H H
T_7 : Neem (*Azadirachta indica*)	79.00	73.17	12.22 (20.45)	18.70 (25.62)	H H
T_8 : Control	90.00	90.00	0.00 (0.00)	0.00 (0.00)	-
SEm±	0.49	0.36	0.93	0.49	-
CD (5%)	1.48	1.09	2.79	1.48	-

*Average of three replications; **Figures in parentheses are Arc sine transformed
H: Highly compatible, M: Moderately compatible, S: Slightly compatible N: Non compatible

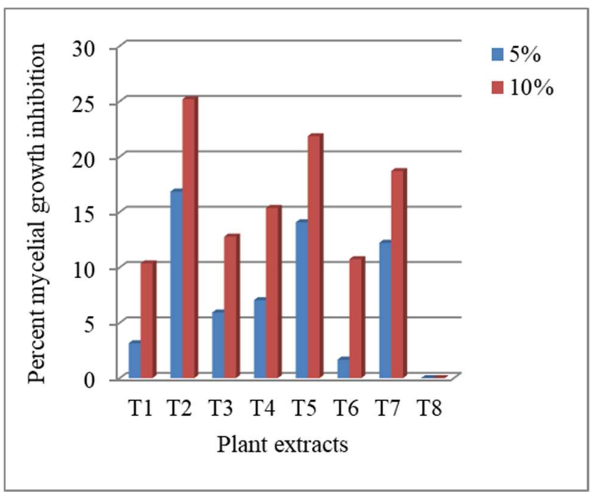

Fig. 3. In-vitro evaluation of per cent mycelial growth inhibition of *T. asperellum* against plant extracts

Conclusion

The present study demonstrated that most plant extracts were compatible with *T. hamatum*, *T. harzianum* and *T. asperellum*, particularly at lower concentration (5%). Marigold, parthenium and periwinkle showed high compatibility at both concentrations, while karanj, lantana, datura and neem were highly compatible mainly at 5% concentration. The radial growth and low inhibition (<30%) indicated that these botanicals did not adversely affect the growth of *Trichoderma* spp. Among the three species, *T. asperellum* exhibited maximum tolerance, followed by *T. hamatum*, whereas *T. harzianum* was comparatively more sensitive at higher concentration (10%). The study highlights that plant extracts, when used at appropriate concentrations, can be safely combined with *Trichoderma* spp. This compatibility supports their integration in eco-friendly and sustainable disease management strategies, reducing dependency on chemical fungicides while enhancing biological control efficiency.

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Conflict of Interest

The authors have no conflict of interest to declare.

Data Sharing

All relevant data are included in the manuscript.

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Research Article

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Economic status of agriculture labour: A case study of Rajasthan

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ABSTRACT

The study was conducted to examine the socio-economic status of agricultural labours, analyze wage and employment patterns, assess income and expenditure patterns, and investigate gender discrimination among agricultural labour in Rajasthan. The collected data revealed that the socio-economic status of agricultural labour was below the average compared to workers in other sectors. The highest level of employment was generated through crop harvesting activities. Annual employment in agriculture averaged 214 days. The average wage rate was ₹ 332.64 in the agricultural sector and ₹ 406.73 in the non-agricultural sector. The total annual family income and total family expenditure were ₹ 194,957 and ₹ 157,857, respectively.

Introduction

India is the world's second-most populous country, and it covers the 17.70% population comparison to the world. India has 1,210,193,422 population were reported of India (Census, 2011). Agriculture is primary sector of the Indian economy. The data provided by the census (2001) of India reveals that about 69 per cent of the total workers were engaged in agriculture and allied activities in the year 1981; while in the year 1991 the share of agriculture in total employment slightly declined to 68 per cent and nearly 70 per cent of the rural and 8 per cent of the urban households still depend on it employment and livelihood (Bhakar *et al*., 2007). Agriculture is the major sector of this state economy which plays an important role in the national income, providing large employment opportunities, supplying adequate food grains to the growing population and giving a scope for earning foreign exchange through export (Kekani, 2013). It still remains the most acceptable fact that agriculture continues to play greater role in the socio-economic development of the country (Raju, 2017). Out of 28 states and 8 Union Territory of India, Rajasthan covers 3,42,239 km^2 or 10.4 per cent of India's total geographical area. Rajasthan has total population 68,548,437 were recorded (Census, 2011). Labour is one of the most important components out of four factors of production (Land, labour, capital and organization). In growth of any country, human labour play important role. Human labour constitutes the most neglected category in India's rural structure. Their income is low and employment is irregular. Since, they have no skill or training and no alternative employment opportunities (Vetrivel and Manigandan, 2013). The majority of the Indian rural workers are dependent on agriculture, and among the workers the number of agricultural labours is quite high (Singh, 2017). As per the survey carried out by the National Sample Survey Organization in the year

2009-10, the total employment, in both organized and unorganized sectors in the country was out of the order of 46.5 crore comprising around 2.8 crore in the organized sector and the balance 43.7 crore workers in the unorganized sector. Out of 43.7 crore workers in the unorganized sector, there are 24.6 crore workers employed in the agricultural sector, about 4.4 crore in construction work and remaining in manufacturing and service. Agricultural labours are mainly economically and socially backward people. They are working on various farms for wages. The productivity in agriculture depends upon the efficiency of agricultural labours, which in turn depends upon their socio-economic conditions (Singh, 2019). Most of the agricultural labours in the village are landless and so they depend on wage paid employment. Employment and wages are the important factors which influence the livelihood status of agriculture labour.

The problem of agricultural labours is not only a critical problem but also a puzzling problem for India. When one has to primarily think of millions of people involved in agricultural operations. The present study is focused on the socio-economic conditions, employment, income and expenditure pattern of agricultural labours in Rajasthan.

Methodology

On the basis of population of agricultural labour, Bikaner (Hyper Arid Partial Irrigated Zone- IC) and Jaipur (Semi-Arid Eastern Plain Zone-IIIA) districts Rajasthan state were selected. 120 agriculture labours were selected randomly for the present study. The primary data (July 2020 to June 2021) was collected from the selected agriculture labour through personal interview method. The secondary data were collected from various published sources.

Analysis of data

(i) Analysis of socio-economic status of agricultural labour in Rajasthan

Tabular analysis was used for the study of socio-economic status of agriculture labour. The statistical tools like average, percentage etc. were employed to summarize and compare various socio-economic characteristics in the present study.

(ii) Employment pattern and wages of agricultural labour in Rajsathan

To find out employment patterns wages of agriculture labour chi-square test was used.

$$X^2 = \sum \frac{(O-E)^2}{E}$$

Where:

X^2 is the chi-square test statistic

Σ is the summation operator (it means "take the sum of")

O is the observed frequency

E is the expected frequency

(iii) Regression analysis of income and expenditure pattern of agriculture labour in Rajasthan

Estimate the income and expenditure functions with the help of multiple linear regression analysis. The following functional forms were used with different variables.

Income function

$Y = a + b_1 X_1 + b_2 X_2 + b_3 X_3 + u$

Where,

Y = Annual total family income (₹)

a = Intercept

X_1 = Total employment (man days)

X_2 = Earners (Number per family)

X_3 = Livestock (Number per family)

bi's = Regression coefficients of respective explanatory variables

u = Error term in sufficient independence variables

Expenditure function

$Y = a + b_1 X_1 + b_2 X_2 + b_3 X_3 + u$

Where,

Y = Expenditure (₹)

a = Intercept

X_1 = Total income per family (₹)

X_2 = Family size ((Number per family)

X_3 = Value of fixed capital assets (₹)

bi's = Regression coefficients of respective explanatory variables

u = Error term in sufficient independence variables

Results and Discussion

The data presented in Table 1 shows that female labour were more than male labours in agriculture sector because there are social restrictions on female in villages, due to which they do not work away from village and female labour work around the village. Almost similar results were reported by Swamikannan and Jeyalakshmi (2015). In the present study, it was found that out of the total 120 respondents 59 respondents were belong to young age, 39 respondents were belong to middle age and 22 respondents were belong to old age. The result shows that young age agricultural labours were more than old age and middle age agricultural labours because the young agricultural labours were more efficient so first people prefer young people to work in the field. During this study it was found that the majority of the agricultural labours were belong to schedule caste and constituted 58.30 per cent of the sample respondent, 23.30 per cent were belong to other backward caste, 16.70 per cent were belong to schedule tribe caste and 1.70 per cent were belong to general caste in Jaipur district. In Bikaner district 56.70 per cent sample respondents were belong to schedule caste, 33.30 per cent were other backward caste, 1.70 per cent were schedule tribe caste and 8.30 per cent were general caste.

Most of the agricultural labours were belong to high school education group. 46 respondents were belong to high school education group, 29 respondents were belong to illiterate group, 24 respondents were belong to middle education group, 14 respondents were belong to primary education group and 7 respondents were belong to graduated education group out of 120. The result shows that the a few labours were achieved higher level education due to poor family condition and they belong from rural background and the peoples of the villages give less importance to the education. During the analysis of the data, it was observed that the majority of the agricultural labours were belonging to joint family, because in villages the family does not consist only of husband, wife and their children but also of uncle, aunts and cousin and grandsons. Living in a joint family has many advantages such as sharing responsibility; managing financial burdens, teamwork, and strong support system. The analysis of the data collected from the agricultural labours shows that majority of the agricultural labours have 4 to 6 members in family in both the districts.

Table 1. Social status of agricultural labours in Jaipur and Bikaner districts of Rajasthan

Particulars	Jaipur (n = 60)	Bikaner (n = 60)	Overall (n = 120)
Gender			
Male	17 (28.20)	36 (60.00)	53 (44.20)
Female	43 (71.80)	24 (40.00)	67 (55.80)
Age			
Young (< 35year)	29 (48.30)	30 (50.00)	59 (49.20)
Middle (35 to 50 year)	21 (35.00)	18 (30.00)	39 (32.50)
Old (> 50 year)	10 (16.70)	12 (20.00)	22 (18.30)
Social caste			
ST	10 (16.70)	1 (1.70)	11 (9.20)
SC	35 (58.30)	34 (56.70)	69 (57.50)
OBC	14 (23.30)	20 (33.30)	34 (28.30)
General	1 (1.70)	5 (8.30)	6 (5.00)
Education			
Illiterate	16 (26.70)	13 (21.70)	29 (24.20)
Primary	11 (18.30)	3 (5.00)	14 (11.70)
Middle	11 (18.30)	13 (21.70)	24 (20.00)
High school	19 (31.70)	27 (45.00)	46 (38.30)
Graduate	3 (5.00)	4 (6.60.)	7 (5.80)
Marital status			
Married	47 (78.30)	53 (88.30)	100 (83.30)
Unmarried	13 (21.70)	7 (11.70)	20 (16.70)
Type of family			
Nuclear	14 (23.30)	17 (28.30)	31 (25.80)
Joint	46 (76.70)	43 (71.70)	89 (74.20)
Family size			
Below 4	1 (1.70)	3 (5.00)	4 (3.30)
4 to 6	32 (53.30)	33 (55.00)	65 (54.20)
Above 6	27 (45.00)	24 (40.00)	51 (42.50)
Social participation			
No participation	43 (71.70)	48 (80.00)	91 (75.80)
Cooperative society/NGO	17 (28.30)	12 (20.00)	29 (24.20)

Source: Primary survey (2020-21)
*Figures in parameters show in per cent of total labour

Economic status of agricultural labour

In this study, the data collected from the respondents, majority of the respondents were lived in *pakka* house in both the districts (Table 2). The possible region of this was several government schemes for houses like PM *Awas Yojna*, for weaker section. The data showed that the majority of agricultural labours in both districts were casual labours, as employment in agriculture is seasonal in nature. On an average 81 respondents were casual labours and 39 respondents were permanent labours. The result shows that the majority of the agricultural labours were have 2 to 4 earners in family in both the districts. In the present study, the data shows that the income of 66 agricultural labours were ₹ 100000 to ₹ 200000, 21 respondent's income ₹ 50000 to ₹ 100000 and 33 respondent's income were more than ₹ 200000 as a whole. The result shows that the most of agricultural labour family income was low due to seasonal nature of employment in agriculture sector and wage rate was also low compared to another sector.

The results show that most of the agricultural labours have no land and some agricultural labours have small piece of land because people sell out their land due to poor irrigation facility, money requirement for family and urbanization. The results also show that the most of agricultural labour borrowed money from non-institutional sources due to lack of knowledge.

Table 2. Economic status of agricultural labour in Jaipur and Bikaner districts of Rajasthan

Particulars	Jaipur (n=60)	Bikaner (n=60)	Overall (n=120)
	Type of house		
Kachha house	4 (6.70)	17 (28.30)	21 (17.50)
Pakka house	56 (93.30)	43 (71.70)	99 (82.50)
	Occupation		
Permanent labour	26 (43.30)	13 (21.70)	39 (32.50)
Causal labour	34 (56.70)	47 (78.30)	81 (67.50)
	Number of earners		
Only 1	5 (8.30)	16 (26.70)	21 (17.50)
2 to 4	55 (91.70)	44 (73.30)	99 (82.50)
	Annual income		
₹ 50,000 to ₹ 100000	10 (16.70)	11 (18.30)	21 (17.50)
> ₹ 1,00,000 to ₹ 200000	32 (53.30)	34 (56.70)	66 (55.00)
> ₹ 200000	18 (30.00)	15 (25.00)	33 (27.50)
	Land holding		
0 to 0.5 ha	45 (75.00)	51 (85.00)	96 (80.00)
> 0.5 to 1 ha	15 (25.00)	09 (15.00)	24 (20.00)
	Source of borrowing money		
Bank	05 (8.34)	05 (8.34)	10 (8.34)
Cooperative society	05 (8.33)	04 (6.66)	09 (7.50)
Relative and friend	39 (65.00)	42 (70.00)	81 (67.50)
No borrowing money	11 (18.33)	09 (15.00)	20 (16.66)

Source: Primary survey (2020-21), *Figures in parameters show in per cent of total labour

Employment pattern and wages of agricultural labour in Jaipur and Bikaner districts

Employment was one of the most important aspects for livelihood. As the employment patterns were strong, they have good income which makes their standard of living better. Overall situation shows that non agriculture sector provided 241.74 days of employment. The significant Chi-square value 5.78 indicates that there is sufficient evidence to conclude that the observed distribution is not the same as the expected distribution. The result shows that in agriculture sector, employment days less than the other sector because the employment in agriculture sector depends on seasons. The highest employment received for agricultural labour when agricultural labour work in both the sectors. Similar results were reported by Swamikannan and Jeyalakshmi (2015).

Table 3. Season wise distribution of employment days in Jaipur and Bikaner districts of Rajasthan

Season	Jaipur (n = 60)			Bikaner (n = 60)			Overall (n = 120)		
	Agriculture	Non-agriculture	Both Sector	Agriculture	Non-agriculture	Both Sector	Agriculture	Non-agriculture	Both Sector
				Kharif					
July	15.03	19.41	21.25	11.00	14.18	22.50	14.40	16.91	21.66
August	18.92	15.66	17.20	15.12	19.00	21.94	17.65	18.23	20.86
September	21.03	15.00	19.16	21.57	23.33	21.37	21.30	19.16	20.42
October	22.35	15.00	19.00	24.03	26.00	22.00	23.19	20.50	20.50
Total	77.33	65.07	76.61	71.72	82.51	87.81	76.54	74.8	83.44
				Rabi					
November	22.03	17.00	22.66	21.98	24.00	24.14	22.01	20.50	23.46
December	17.37	16.67	21.56	12.59	22.33	23.40	15.60	21.52	22.21
January	18.96	20.00	20.00	13.09	23.00	24.00	17.11	22.44	22.00
February	19.96	21.75	21.00	21.23	23.08	21.00	20.49	22.76	21.00
Total	78.32	75.42	85.22	68.89	92.41	92.54	75.21	87.22	88.67
				Zaid					
March	21.52	17.50	22.5	25.07	25	22.5	23.08	23.75	22.5
April	14.66	23.28	24.75	11.12	20.33	20.00	12.96	21.42	23.8
May	13.44	21.47	21.50	2.00	14.00	0.00	12.30	17.73	21.5
June	9.14	16.46	23.33	2.00	15.00	0.00	8.25	16.40	23.33
Total	58.76	78.71	92.08	40.19	74.33	42.5	56.59	79.30	91.13
Grand total	214.41	219.92	253.91	180.80	249.25	222.85	208.34	241.74	263.24
		Chi-square value					5.78*		

Source: Primary survey (2020-21), *Significant at 1 per cent level of significance

Table 4. Distribution of mode of payment of wages of agricultural labour In Jaipur and Bikaner districts of Rajasthan

Mode of payment	Jaipur (n=60)	Bikaner (n=60)	Overall (n=120)
Cash	45 (75.00)	43 (71.70)	88 (73.40)
Kind	4 (6.70)	3 (5.00)	7 (5.80)
Both (cash and kind)	11 (18.30)	14 (23.30)	25 (20.80)

Source: Primary survey (2020-21), *Figures in parameter show in per cent of total labours

Mode of payment

Result shows that most of the agricultural labour received wages in cash because in kind they only found grain and vegetables to eat from the field while most of family's requirement goes to non-food items such as cloth, fuel and light, education, health, social function, narcotics, etc.

Table 5 shows that the average wage rate in Jaipur district were ₹ 329.64 in *kharif* season, ₹ 325.79 in *rabi* season and ₹ 329.15 in *zaid* season. In case of Bikaner district average wage rate were ₹ 352.48 in *kharif* season, ₹ 335.42 in *rabi* season and ₹ 351.48 in *zaid* season. Annual average wage rate in Jaipur district was ₹ 328.19 and ₹ 346.46 in Bikaner district in agriculture sector. In non-agricultural sector, annual average wage rate was ₹ 391.67 in Jaipur and ₹ 421.83 in Bikaner district. The results shows that the wage rate was low in compared to last year in agriculture sector. The possible reason may be COVID- labour migration. In the study period many labours return to their village and engaged in farming work. The migration of labour effect the demand-supply of labour and another reason was that were no need working experience and education in agriculture so most unemployed labour work in agriculture sector. The chi square value was non-significant. The chi square value indicates that the difference between expected and actual value is likely just due to chance. Thus, it is concluded that the collected sample does not support the hypothesis of a difference. Two variables are equally likely.

Table 5. Season wise wage rate of agricultural labour in Jaipur and Bikaner districts of Rajasthan

Season	Jaipur (n = 60)		Bikaner (n = 60)		Overall (n = 120)	
	Agriculture	Non-agriculture	Agriculture	Non-agriculture	Agriculture	Non-agriculture
Kharif						
July	328.92	352.33	349.14	357.52	339.03	354.92
August	324.66	381.25	337.21	366.38	330.93	373.81
September	325.23	390.50	360.49	379.09	342.86	384.79
October	339.77	392.00	363.09	458.06	351.43	425.03
Avg. wage	329.64	379.02	352.48	390.26	341.06	384.64
Rabi						
November	338.03	370.00	370.25	430.94	354.14	400.47
December	315.93	415.08	310.59	455.83	313.26	435.45
January	318.71	410.69	309.19	467.82	313.95	439.25
February	330.49	438.59	351.68	467.80	341.08	453.19
Avg. wage	325.79	408.59	335.42	455.59	330.60	432.09
Zaid						
March	341.91	370.93	358.83	471.04	350.37	420.98
April	322.40	408.41	367.10	452.84	344.75	430.62
May	330.70	409.65	350.00	389.28	340.35	399.46
June	321.62	360.71	330.00	365.00	325.81	362.85
Avg. wage	329.15	387.42	351.48	419.64	340.32	403.48
Annual Avg. wage	328.19	391.67	346.46	421.83	337.32	406.73
Chi square value					2.07^{NS}	

Source: Primary survey (2020-21), NS-non significant

Table 6. Average annual family income of agricultural labour in Jaipur and Bikaner districts of Rajasthan

Particulars	Jaipur (n = 60)	Bikaner (n = 60)	Overall (n = 120)
Wage earning	70838 (33.38)	65513 (36.85)	68175 (34.96)
Crop production	14250 (6.71)	13633 (7.67)	13941.50 (7.15)
Livestock production	10841 (5.10)	15483 (8.71)	13162 (6.75)
Petty business	600 (0.28)	791 (0.45)	695.50 (0.35)
Business and services	14766 (6.95)	6000 (3.37)	10383 (5.32)
Other earners	100883 (47.54)	76316 (42.93)	88599 (45.44)
Total	212178 (100)	177736 (100)	194957 (100)

Source: Primary survey (2020-21), *Figures in parameters show in per cent of total income

Table 6 shows that the total income from wage earning was ₹ 68175 and it contributes 34.96 per cent of total family income. Crop production was ₹ 13941.50 and share 7.15 per cent of total family income. The income from livestock contributed ₹ 13162 and share 6.75 per cent of total family income. Petty business contributed ₹ 695.5 and share 0.35 per cent of total family income. The income from business and services contributed ₹ 10383 and share 5.32 per cent of total family income. The total family income was ₹ 194957, in which 45.44 per cent income contribute by other family earners in Rajasthan as a whole.

Table 7. Income functions of agricultural labour in Jaipur and Bikaner districts of Rajasthan

Particulars	Jaipur		Bikaner	
	Coefficients	Standard error	Coefficients	Standard error
Intercept	-29353.89	6262.52	6244.59	8234.94
Total employment days (X_1)	511.30*	42.68	414.90*	35.83

Number of earners (X_2)	3697.32*	3735.55	7939.23*	2139.61
Number of livestock (X_3)	572.07	769.71	306.66	467.59
R^2	0.83		0.72	

*Significant at 1 percent

The study revealed that, all the three variables have jointly explained 83 per cent variation in Jaipur district and 72 per cent variation in Bikaner district of total income received from various sources (Table 7). In Jaipur district the coefficients of independent variables *viz.,* total employment days positive, number of earners was negative and number of livestock positive. X_1 and X_2 was significant at 1 per cent level of significant and same pattern follow in Bikaner district by all the independent variable. Table 8 revealed that the expenditure on food items were ₹ 52741 and non-food items was ₹ 113506 and annual expenditure of agricultural labour family were ₹ 166247 in Jaipur district. The maximum share in expenditure on food (31.72%) followed by fuel and light (16.56%), social function (9.62%) health (9.91%), narcotics (8.44) education (6.70%), transportation (4.96%) and narcotics (4.53%). In Bikaner district the total expenditure on food items were ₹ 45233 and total non-food items were ₹ 104223 and total expenditure of agricultural labour family were ₹ 1494677. The maximum share of expenditure on food (31.03%) followed by fuel and light (16.02%), narcotics (10.72%), health (8.72%), social function (8.21%), transportation (8.04%), education (7.52%), and cloth (7.39%). Result shows that the maximum share of expenditure on food, fuel and light.

Table 8. Annual family expenditure of agricultural labour in Jaipur and Bikaner districts of Rajasthan

Particulars	Jaipur (n = 60)	Bikaner (n = 60)	Overall (n = 120)
Food	52741 (31.72)	45233 (30.26)	48987 (31.03)
Cloth	13068 (7.86)	11055 (7.39)	12051 (7.63)
Fuel and light	27540 (16.56)	23063 (15.43)	25301 (16.02)
Health	14820 (8.91)	12950 (8.66)	13885 (8.79)
Education	11143 (6.70)	11241 (7.52)	11192 (7.09)
Transportation	8255 (4.96)	12025 (8.04)	10140 (6.42)
Social function	15996 (9.62)	12271 (8.21)	14133 (8.95)
Narcotics	14044 (8.44)	16025 (10.72)	15035 (9.52)
Other	8640 (5.19)	5604 (3.74)	7122 (4.46)
Total	166247 (100)	149467 (100)	157857 (100)

Source: Primary survey (2020-21), *Figures in parameters show in per cent of total expenditure

Table 9 shows that the monthly average family expenditure of agricultural labour of the both of district. Highest expenditure ₹ 16245.44 in month of March followed by ₹ 15237.27 in month of October and ₹ 14578.44 in month of April in Jaipur district. Whereas in Bikaner district, highest expenditure ₹ 15474.10 in month of October followed by ₹ 14575.33 inmonth of March and ₹ 13577.79 in month of April. Total expenditure was ₹ 166247.00 in Jaipur district and ₹ 149466.90 in Bikaner district. Overall total expenditure were ₹ 157857.00. Result shows that the expenditure of the family were different every month because ofthe requirement of family was different at different months and depends on food, lightand fuel, education, health, social function, narcotics, transportation etc.

Table 9. Monthly average family expenditure of agricultural labour in Jaipur and Bikaner districts of Rajasthan

Month	Jaipur	Bikaner	Overall
July	12827.42	11664.04	12245.73
August	13265.65	12670.80	12968.23
September	14586.54	12534.20	13560.37
October	15237.27	15474.10	15355.69

November	14410.78	12854.48	13632.63
December	12325.32	11398.64	11861.98
January	12520.78	11458.45	11989.62
February	14257.90	11245.27	12751.59
March	16245.44	14575.33	15410.39
April	14578.20	13577.79	14078.00
May	13512.47	12723.35	13117.91
June	12479.22	9290.44	10884.83
Total	166247.00	149466.90	157856.90

Source: Primary survey (2020-21)

The Table 10 shows that, all the three independent variables have jointly explained 65 per cent variation in Jaipur district and 61 per cent variation in Bikaner district of total income received from various sources. In Jaipur, the family size was significant at 5 per cent level of significance. Whereas in Bikaner, the family size and fixed capital assets were significant at 1 per cent level of significance.

Table 10. Expenditure functions of agricultural labour in Jaipur and Bikaner districts of Rajasthan

Particulars	Jaipur		Bikaner	
	Coefficients	Standard error	Coefficients	Standard error
Intercept	-16084.30	19038.19	24664.57	16752.04
Total income X_1	-0.07	0.15	-0.08	0.14
Family size X_2	23091.79**	2476.43	14490.70*	2255.99
Fixed capital assets X_3	0.01	0.01	0.03*	0.03
R^2	0.65		0.61	

*Significant at 1 per cent level of significance, **Significant at 5 per cent level of significance

Conclusion

The present study shows that the socio-economic status of agricultural labour in Rajasthan state. Agricultural labour comes from a poor background and low social-economic background. Majority of agricultural labour belong to female category. Majority of agricultural labour have 2 to 4 earners in family and total family income were between to 1-2 lakh rupees. Wage rates are low in the agriculture sector compared to other sectors. The highest income comes from other then agricultural earners in the family. The total family incomes were ₹ 194957 and total family expenditure were ₹ 157857.

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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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Antifungal susceptibility and characterization of *Alternaria alternata* causing leaf spot disease on *Aloe vera* (L.) Burm. f. in India

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ABSTRACT

Leaf spot is destructive disease of Aloe vera and the disease incidence during 2021-22 was varying 50-100%. The symptoms initially appeared as minute circular, water-soaked lesions on the adaxial surface of leaves, which later developed into depressed dark brown to black necrotic spots. The associated pathogen was isolated in pure culture on PDA exhibited rapid growth, initially light grey to olive green, which gradually turned dark brown to black with abundant sporulation. The colony surface typically showed concentric zonation, while the reverse side of the plate appears dark brown to black pigmentation. Further, ITS region was sequenced, and based on morphological and molecular sequences the pathogen was identified as Alternaria alternata. The pathogen found highly sensitive to the fungicides Difenoconazole under In-vitro conditions, and was found to be the most effective in inhibiting mycelial growth of the test pathogen Alternaria alternata.

Introduction

Aloe (*Aloe barbadensis* Miller) is a perennial succulent plant belonging to the family *Asphodelaceae* (formerly placed under *Liliaceae*), renowned for its medicinal, pharmaceutical and economic importance. Aloe gel is extensively utilized in the cosmetic industry for the manufacture of soaps, shampoos, hair-care products, toothpaste, body creams and several other personal care products. The plant is well adapted to hot and dry tropical climates and is widely distributed across Asia, Africa and other tropical and subtropical regions of the world. In India, it is commercially cultivated in Gujarat, Rajasthan, Maharashtra, Andhra Pradesh and Tamil Nadu (Surjushe *et al*., 2008; Sutaliya *et al*., 2025). More than 250 species of aloe are known worldwide and have very much important role in traditional medicine (Baby and Justin, 2010). Aloe have more than 200 compounds, among which 75 are biologically active compounds. The leaf of aloe made up of three layers, the outer most layer is protective layer, 15-20 cm cells thick in size and synthesize carbohydrates and proteins. The active compounds of aloe are anthraquinones, chromones, polysaccharides, and enzymes. (Sahu *et al*., 2013). It has laxative, antihelminthic, uterine stimulant properties and also effective for the treatment of sores and wounds, skin disease, colds and coughs, constipation, piles, asthma, ulcer, diabetes and various fungal infections. In some part of the country, it is also used to prepare curry (Kumar and Yadav, 2014).

Despite of immense antimicrobial properties, the aloe plants reported susceptible to various fungal and bacterial phytopathogens. Leaf spots diseases are the most frequently and widely reported, wherever aloe crop grown (Kamalakannan *et al.*, 2008; Bajwa *et al*., 2010; Chavan and Korekar, 2011; Silva and Singh, 2012), whereas, bacterial soft rot disease reported predominantly during hot season in high humid areas (Meena *et al*., 2023). The fungal as well as bacterial pathogens are responsible for qualitative and quantitative degradation. The genus "*Alternaria*" is ubiquitous fungus found from temperate to tropical ecology in different habitats such as soil, plants and atmospheres (Woudenberg *et al*., 2015). Species of the genus *Alternaria* affect almost all types of the agriculture produces and leads substantially economic loss (Meena, 2012; Meena *et al*., 2020; Sun *et al*., 2023). The genus consisting of opportunist pathogenic species and causes disease on more than 380 host plant species (Chavan and Korekar, 2011).

The present study focusing on the identification of the associated phytopathogenic fungi, pathogenesis and study on the fungicidal susceptibility using commercial fungicides. During the farmers' field visits, symptoms such as water-soaked lesion, small brown to necrotic spots were observed in different part of Gujarat. The similar symptoms were also observed at different stages on the leaf of aloe in the experimental field at ICAR- DMAPR, Anand, Gujarat during 2021-22. Therefore, keeping all above in view a systematic study was carried out for better understanding the leaf spot disease of aloe.

Material and Methods

Sample collection and isolation

The symptom exhibited leaf samples were collected in aluminum foil from experimental field of ICAR-DMAPR, Anand, and farmers' fields of Gujarat. Further, to isolate the associated causal agent of disease, colonized tissues of leaves were cut into small pieces and surface sterilized with 70% alcohol followed by 2 min immersion in 2% sodium hypochlorite (NaOCl) and subsequently three washing with sterile distilled water. Then 2-3 small sections of leaf placed on solidified and sterilized potato dextrose agar (PDA) Petri plates, supplemented with 250 ppm chloramphenicol to prevent bacterial contamination. The inoculated cultures were incubated at 25 $\pm$ 2 ◦C in BOD incubator. Observation was recorded after 6-7 days of inoculation. After 7-8 days sub culture was carried out by transferring hyphal tip on new PDA plate. Microscopic slides were prepared by mounting in stain to examine morphological characteristics of pathogen for the 7 days old culture.

DNA extraction and PCR amplification

The genomic DNA was extracted from 3 days old fungal mycelium grown on PDA using the commercial DNA isolation kit (Qaigen). Further for characterization at molecular level the internal transcribed spacer (ITS) *ITS* region was amplified using ITS1 and ITS4 (White *et al*., 1990). Genomic PCR reaction was carried out using PCR master mix (Thermo scientific), 10mM forward and reverse primers of *ITS* region, DNA template and nuclease free water in the 25 µl reaction. The PCR cycle was performed at initial denaturation 94°C for 5 min followed by 35 cycles of PCR reaction (94°C for 30 second, 55°C annealing for 40 sec and extension at 72°C for 1 min) and final extension at 72°C for 10 mins as prescribed earlier (Meena *et al.*, 2019). Amplified products were separated on 1% low melting agarose gel. Expected size bands were excised from gel and purified using GeneJET Gel Extraction Kit (Thermo scientific). Purified PCR products were sequenced bi-directionally.

Bioinformatics analysis and identification of pathogen

Sequence similarity search was carried out using Blast*N* tool of NCBI for putative identification of pathogen. Multiple sequence alignment was performed using clustal W algorithm of Mega X with the *ITS* region homologues nucleotide sequences of genus *Alternaria* and one out group member *Stemphylium vesicarium.* Aligned sequences of *ITS* region were used to construct phylogenetic tree with the maximum likelihood tree algorithm using 1000 bootstrap replicates in Mega X software (Kumar *et al.*, 2018).

Pathogenicity test

To confirm pathogenicity of the isolated fungus from infected leaves of *Aloe vera*, a suspension culture of conidia (5×10^5 conidia/mL), collected from colonies on the purified fungal isolate AALS-1 were used to inoculate on aloe leaves. Three plants were used and three leaves per plant were inoculated, whereas the control group was treated with the sterile distilled water. After inoculation, the plants were covered with the clean transparent polythene bags to maintain the higher humidity and plants were transferred to green house for incubation and observations on disease development were recorded regularly. The experiment was repeated twice; the causative agent was reisolated from symptoms developed on inoculated plant and was compared morphologically with the original strain.

Antifungal susceptibility assay

Eight commercially available fungicides were procured (Table 1) and their efficiency was analyzed against the isolated AALS-1 under *In-vitro* conditions. The experiment was based on mycelia growth inhibition using fungicide sensitivity assay. Optimized concentration of fungicides mixed in sterilized PDA medium and poured into petri plates. A fungal mycelium disk of 6 mm from the 7 days old pure culture was cut with cork-borer and placed at the center on fungicides amended PDA plates. The percent inhibition of the mycelium growth over the control was calculated by the formula (Vincent, 1947) described as per cent inhibition (PI) is equal to mycelium growth of the fungus in control treatment minus mycelium growth of the fungus with treatment multiplied by 100 and divided by mycelium growth of the fungus in control treatment (measured in mm or cm).

Statistical analysis

The data recorded from the experiments were subjected to Dunnett's statistical test for statistical analysis appropriate to the CRD by using the Microsoft Excel package. The results were presented at 5% level of significance (P = 0.05). The critical difference (CD) values were calculated to compare the various treatment means.

Results and Discussion

Symptoms and pathogenesis

Alternaria is a ubiquitous fungus found from temperate to tropical ecology in different habitats such as soil, plants and atmospheres. Diverse species of the genus *Alternaria* affect almost all types of the agriculture produces and leads substantially economic loss (Meena *et al*., 2012). Several studies showed the different causal agent associated with the leaf spot diseases on aloe. Avasthi *et al.* (2015 & 2018) reported the leaf spot disease of aloe caused by *Fusarium proliferatum* and *Curvularia* species. Similarly Ghosh *et al*. (2018) also reported *Alternaria brassicae* causing leaf spot disease on aloe, but the symptoms reported on the leaves were not consistent with the symptoms observed in the present study. In another study conducted by Ghosh *et al.* (2016), the results were in corroborating with the present study where identical symptoms and pathogen (*A. alternata*) were reported.

To resolve this, a systemic data was recorded on pathogenesis and *Alternaria alternata* was identified as causal agent of leaf spot disease on aloe based on the morphological as well as molecular keys. The survey results and field observations revealed that all the aloe plants were infected with leaf spot disease and the disease incidence was recorded up to 100% during mild winter season. The disease symptoms were initiated with minute, circular, transparent water-soaked lesions on adaxial surfaces of aloe leaves and within 3 days the lesions changed to light brown. The lesions were then in later stage after 7-8 days, became enlarged and transformed into dark brown to black spot. As disease progressed the spots became necrotic and the leaf surface finally developed into a saucer shaped depression on adaxial and the corresponding abaxial surfaces within one month (Fig.1, A-E). Under the sever condition the leaf surface covered with numerous such spots and each spot make the depression, which lead substantial yield reduction. Further, disease was initiated under control condition to establish the Koch's postulates and similar symptoms were recorded (Fig.1, G-I).

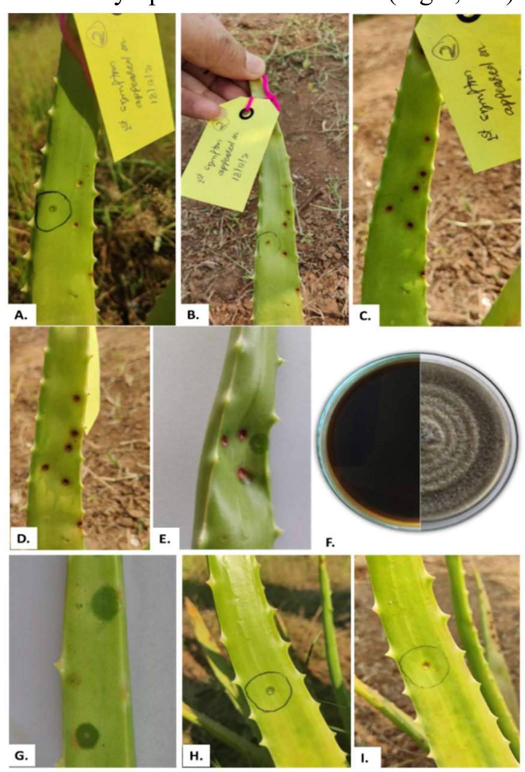

Fig.1. Pathogenesis of *Alternaria alternata* on *Aloe vera* where the progress was recorded at 3 days interval from (A-D) and depression form after 30 days of infection (E), typical colony morphology of *A. alternata* on PDA and Koch's postulates were also established (F-H).

Morphological and molecular characterization

The fungus produced olive green to dark brown color mycelium growth on PDA and the reverse pigmentation varied from dark brown to blackish color. The mycelium was grey-brownish, multi celled, distinctly septate and produced irregular branches. The typical concentric zonation of fungal growth was distinctly visible in ten days old culture (Fig.1, F). The microscopic observation revealed the dark muriform type conidia, brown to black olivaceous color with the typical 18-36×8-16 µm size and the arises from the long-branched chains of conidiophores. The conidiophores were of 16-45×3-7 µm in size. Thus, based on the morphological features the fungus primarily identified as *Alternaria spp.* pathogen associated with leaf spot of aloe. The morphological features including the mycelium growth, conidial chain and conidia characteristics described here are consistent with those of *A. alternata* described by Simmons (Simmons, 1995; Sun *et al*., 2023). Further, approximately 550 bp *ITS* region of isolated pathogen sequenced. The retrieved sequence contigs of the *ITS* region assembled to generate the consensus sequences and submitted to NCBI GenBank (Accession number PZ374808). The BLAST*n* analysis showed highest 99.83% similarity with *Alternaria alternata* (Accession number PX068376.1). Further phylogenetic tree was constructed using maximum likelihood method using sequences of *ITS* region of *Alternaria* spp. including other closely related genera and it clustered with the *Alternaria alternata* (Fig. 2).

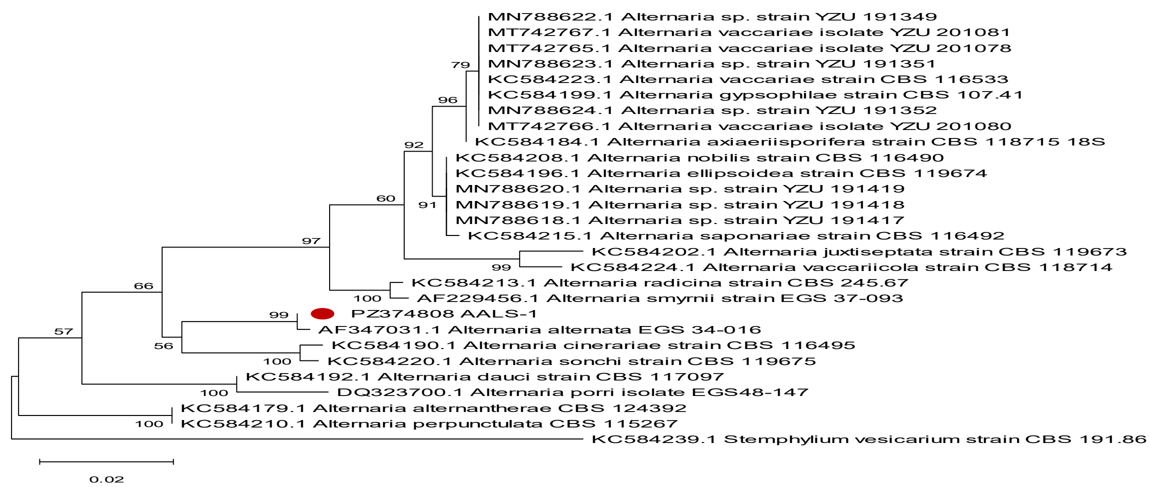

Fig. 2. The maximum likelihood (ML) tree was generated with 1000 bootstrap replications using the Tamura-3-parameter model. The ML tree is inferred from *ITS* sequences of genus *Alternaria* and one out group member *Stemphylium vesicarium.* Bootstrap support values greater than 50% are pointed out at the nodes. Isolate AALS-1 with red circle sequences in the present study. The bar indicates the substitutions number per position

In-vitro antifungal susceptibility

The results of *in vitro* efficacy of fungicides against *Alternaria alternata* presented in table 1. The mycelium growth of the test fungus significantly reduced by the most of them, however, among the tested fungicides Difenoconazole found most efficient to check the mycelium growth with the 98.04±1.96 % efficiency followed by Tebuconazole (50%) + Trifloxystrobin (25%) with 86.02±1.17% efficacy over the control. The mycelium growth of the fungus in difenoconazole and Tebuconazole (50%) + Trifloxystrobin (25%) treated Petri plats was insignificant, whereas in azoxystrobin and propineb the growth was recorded as 60.00±1.16 mm and 47.67±1.45 mm respectively after 7 days of incubation and were found least efficient against the *Alternaria alternata* under *In-vitro* conditions. Among the others Cuprous hydroxide inhibited the mycelium growth with 85.92±1.912 % efficacy, where 12.33±1.45 mm mycelium growth recorded as compare to the control treatment (88.00±1.53 mm). The results on the antifungal susceptibility were carried out under *In-vitro* conditions and supported by the several studies carried out on *A. alternata* (Meena *et al*., 2020).

Table 1. *In-vitro* efficacy of fungicides on the mycelium growth of *A. alternata* of leaf spot of aloe

S. No.	Treatment	Conc. (%)	Redial mycelium growth (mm)	Inhibition over control (%)
1.	Azoxystrobin	0.1	60.00±1.16	31.81±0.863
2.	Tebuconazole (50%) + Trifloxystrobin (25%)	0.1	12.33±1.20	86.02±1.17
3.	Difenoconazole	0.1	1.67±1.67	98.04±1.96

4.	Copper oxychloride	0.25	12.33±1.45	85.92±1.912
5.	Propineb	0.25	47.67±1.45	45.75±2.551
6.	Carbendazim (12%) + Mancozeb (63%)	0.25	18.33±1.20	79.15±1.436
7.	Metalaxyl (4%) + Mancozeb (64%)	0.25	22.33±1.76	74.56±2.328
8.	Metalaxyl (25%)	0.25	25.00±0.58	71.593±0.246
9.	Control	00	88.00±1.53	0
	SEm±		1.374	1.621
	CD (5%)		4.11	4.854
	CV (%)		7.44	4.41

*The redial growth of fungal isolate *AALS-1* is the mean value of three replicates.

Conclusion

Based on the results this study concluded that leaf spot disease of aloe caused by *Alternaria alternata*, is a destructive pathogen, and disease infection leads substantial quality and quantitative losses in aloe production. The pathogen was determined based on the morphological and molecular sequences of ITS region. Under *In-vitro* conditions, fungicide Difenoconazole found most efficient to restrict the mycelium growth followed by Tebuconazole (50%) + Trifloxystrobin (25%) against the test pathogen. These fungicides may be used for management of the disease in field conditions.

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Conflict of Interest

The authors declare that they have no conflict of interest about this manuscript and research.

Ethical Approval

This article does not contain any studies with human participants or animals performed by any of the authors.

Data Sharing

All relevant data are within the manuscript.

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Short Note

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Makhana (*Euryale ferox* Salisb.): Possibilities for cultivation in Haryana

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ABSTRACT

Makhana (*Euryale ferox* Salisb.) was cultivated at the Botanical Garden, CCS HAU, Hisar during 2022–23 to evaluate its cultivation potential under Haryana conditions. Seeds were sown in October 2022 in puddled clay soil at a depth of 4–5 cm in a water tank containing a 10 cm soil layer. Seedlings emerged approximately one month after sowing, with initial leaves exhibiting a reddish-purple coloration and triangular shape. Plant growth remained slow during the winter months due to low temperature stress, but accelerated from March onwards, producing large, round, spiny leaves. Vegetative growth continued until September, followed by flowering from October 2023 to January 2024. Flowering, seed set, maturation, and seed dispersal occurred simultaneously on different shoots. The seed yield ranged from 50 to 100 g per plant.

Makhana (*Euryale ferox* Salisb.) is cultivated in stagnant perennial water bodies such as ponds, land depressions, oxbow lakes, swamps, and ditches. It is commercially grown in countries like India and China. In India, Bihar is the leading state in makhana cultivation, followed by West Bengal (Kushari and Bhowmik, 2025). The seeds, commonly referred to as "black diamond," are valued for both nutritional and economic importance. Makhana is a crop suited to tropical and subtropical climates (Mandal *et al.*, 2010).

'Swarna Vaidehi' is a high-yielding variety developed by ICAR-RCER, Patna, which has been widely adopted by farmers in Bihar and has played a significant role in the commercialization of this crop. Its adoption has substantially enhanced farmers' income. Makhana cultivation is particularly popular in flood-prone districts of eastern and northeastern India. It is grown in ponds and waterlogged areas across approximately 10 states, covering about 20,000 ha. Bihar alone contributes nearly 90% of global production (Kushari and Bhowmik, 2025).

Makhana seeds possess medicinal properties, including analgesic and aphrodisiac effects. They are traditionally used in the treatment of chronic diarrhoea, vaginal discharge, impotence, premature ejaculation, nocturnal emissions, and kidney weakness associated with frequent urination. Additionally, makhana exhibits cardioprotective properties (Das *et al.*, 2006). Nutritionally, makhana is a rich source of energy (405 kcal/100 g), protein (11.5%), carbohydrates (64.7%), fat (7.6%), minerals

(2.2%), and calcium (303 mg/100 g). It is widely used in the preparation of various traditional dishes such as kheer, vermicelli, and halwa.

With the increasing extent of waterlogged areas in many regions, makhana offers a viable alternative to utilize such lands productively, particularly after fish cultivation. Considering its economic importance and potential for effective utilization of waterlogged ecosystems, the present study was undertaken to evaluate the feasibility of makhana cultivation under Haryana conditions.

An experiment was conducted at the Botanical Garden of the MAP Section, Department of Genetics and Plant Breeding, CCS HAU, Hisar during 2022–23 to assess the adaptability of makhana under Haryana agro-climatic conditions. Seeds were sown in October 2022 at a depth of 4–5 cm in puddled clay soil within a water tank, maintaining a soil depth of approximately 10 cm. Five plants were randomly selected for recording observations on growth, flowering, and yield parameters. Data were collected on seed germination (days), seedling colour and leaf shape, foliage characteristics, flower colour, number of flowers per plant, number of seeds per ovary, ovary characteristics, seed traits, and seed yield per plant.

Seedlings emerged within 25–40 days after sowing. The initial leaves were reddish-purple and triangular in shape (Table 1). Early growth was slow during the winter season due to low temperature stress, which limited active development. Growth accelerated from March onwards with rising temperatures. During the active growth phase, plants produced large, round, spiny leaves. Vegetative growth continued until September, followed by flowering from October 2023 to January 2024.

Flowering, seed formation, maturation, and seed release occurred simultaneously on different shoots. On average, 10–20 flowers per plant were recorded. Flowers were pink in colour, while the ovary turned whitish-green at maturity and was oval in shape. Each ovary produced approximately 10–25 seeds. The seed coat was initially brown, later turning dark brown to blackish. Seeds were round, with sizes ranging from 0.5 to 1.5 cm. Seed yield ranged from 50 to 100 g per plant.

Makhana cultivation represents a low-cost production system capable of generating high-value produce from otherwise unproductive waterlogged areas where conventional crops cannot be successfully grown. It has strong potential to enhance farmers' income with minimal additional investment, primarily limited to seed cost.

Table 1. Characteristics of Makhana variety Swarna Vaidehi

S. No.	Plant characters	Observation
1.	Seed germination	25-40 days
2.	Seedling colour	Reddish-purple
3.	Seedling leaf shape	Triangular
4.	Foliage leaf colour	Light-dark green
5	Foliage leaf shape	Round shape
6.	Flower colour	Pink colour
7.	No. of flowers/ plant	10-20 flowers/ plant
8.	Ovary colour	Whitish green
9.	Ovary shape	Oval shape
10.	No. of seeds/ ovary	10-25 seeds/ ovary
11.	Seed coat colour	Brown to dark brown
12.	Seed size	0.5-1.5 cm
13.	Seed shape	Round shape
14.	Seed yield/ plant	50-100 g/ plant

The present investigation demonstrates that makhana can be successfully cultivated under Haryana conditions, particularly in waterlogged areas. Its cultivation offers an effective means of converting otherwise unproductive lands into economically viable production systems. Adoption of makhana farming can enhance farmers' income by utilizing underexploited waterlogged resources with minimal additional inputs. Furthermore, the crop can be integrated into natural ponds and water bodies developed under watershed management schemes. In addition to its economic benefits, makhana cultivation may help reduce water evaporation losses due to its extensive leaf cover over the water surface.

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Conflict of Interest

The authors have no conflict of interest to declare.

Data Sharing

All relevant data are included in the manuscript.

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Short Note

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Socio-economic status of dragon fruit farmers in Haryana

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ABSTRACT

The study was undertaken to analyse the socio-economic status of dragon fruit farmers from all over Haryana. The results revealed that the majority of dragon fruit growers (58.75%) belonged to the middle-age group of 35–50 years, indicating active working participation. In terms of education, 42.5% growers attained senior secondary level, while 25% were graduates and above, reflecting relatively better literacy compared to the general farming community. About 70% of the respondents belonged to nuclear families. The small and medium farmers dominated with 26.25% and 27.5% share, respectively, while only 5% were large farmers. Annual income ranged mostly between ₹ 1.5–3.0 lakh (57.5%). Nearly half of the farmers (48.75%) cultivated dragon fruit on one acre, while 27.5% had less than one acre under the crop.

Dragon fruit (*Hylocereus spp.* and *Selenicereus spp.*) is native to Central and South America has become globally significant due to its resilience against abiotic stress, pest resistance and low input requirements, making it suitable for cultivation even on marginal lands. Being a Crassulacean Acid Metabolism (CAM) plant, it efficiently conserves moisture and performs well in arid and semi-arid environments (Wakchaure *et al.*, 2021). Its multiple annual harvests, long productive lifespan and high benefit-cost ratio make it an attractive crop for commercial cultivation (Kikon *et al.*, 2021).

It introduced in India in the late 1990s, and its cultivation area increased substantially from only 4 hectares in 2005 to over 3,000 hectares by 2020 (Wakchaure *et al.*, 2021). According to latest data for year 2023-24 it is cultivated on about 15,000 hectares with a total production exceeding 55,000 metric tonnes (Anonymous, 2025). Major producing states include Andhra Pradesh, Telangana, Maharashtra and Karnataka (Sathe *et al.*, 2024).

In Haryana, where horticultural crops cover about 6.79% of total agricultural area, dragon fruit cultivation remains limited but promising due to the state's favourable climate and proximity to metropolitan markets (Economic Survey of Haryana, 2025). Expanding its production could enhance income diversification among farmers and strengthen the state's horticultural sector. However, to exploit its full potential, there is a need for systematic assessment of production costs, marketing margins, price spread, and overall economic viability (Akhil *et al.*, 2024). Consequently, the present study has been undertaken.

The study was conducted in Haryana state during the year 2024-25. Primary data were collected from the 80 dragon fruit farmers from all over Haryana while secondary data were collected from various published/ unpublished sources. Primary data were collected by multistage purposive sampling. Data related to cost and returns of production and marketing were collected through a well-developed pre-tested interview schedule. For studying the marketing pattern of dragon fruit, 5 marketing intermediaries each from respective markets were selected for the collection of data regarding various costs involved, margins of market intermediaries in the marketing of dragon fruit.

The socio-economic characteristics of the 80 dragon fruit growers surveyed in Haryana are presented in Table 1. The age distribution revealed that 14 respondents (17.5%) were below 35 years of age, while the majority, comprising 47 farmers (58.75%), belonged to the 35-50 years age group. The remaining 19 farmers (23.75%) were above 50 years of age. Regarding educational attainment, 26 respondents (32.5%) had completed education up to the secondary level, 34 farmers (42.5%) had studied up to the senior secondary level, and 20 respondents (25%) were graduates or possessed higher qualifications. Analysis of family structure indicated that most farmers (70%) belonged to nuclear families, whereas 24 respondents (30%) were members of joint families.

The landholding pattern showed that 15 farmers (18.75%) were marginal farmers, 21 (26.25%) were small farmers, 18 (22.5%) were categorized as semi-medium farmers, and 22 (27.5%) belonged to the medium farmer category. Only four respondents (5%) were large farmers. In terms of annual income, 20 farmers (25%) reported a gross annual income below ₹ 1.50 lakh, while 46 farmers (57.5%) earned between ₹ 1.50 lakh and ₹ 3.00 lakh. The remaining 14 farmers (17.5%) reported annual incomes exceeding ₹ 3.00 lakh. Concerning dragon fruit cultivation, 22 farmers (27.5%) cultivated the crop on less than one acre, 39 farmers (48.75%) on one acre, and 19 farmers (23.75%) on more than one acre of land. The present findings are consistent with those reported by Chongloi *et al.* (2022) and Akhil *et al.* (2024) in studies conducted across different states of India. India.

Table 1. Socio-economic characteristics of sample farmers in Haryana

Variable	Category	Frequency	Percent
Age	up to 35	14	17.5
	35-50	47	58.75
	>50	19	23.75
Education	Secondary	26	32.5
	Senior Secondary	34	42.5
	Graduation or above	20	25
Family type	Nuclear	56	70
	Joint	24	30
Land holding size	Marginal (<1 ha)	15	18.75
	Small (1-2 ha)	21	26.25
	Semi-medium (2-4 ha)	18	22.5
	Medium (4-10 ha)	22	27.5
	Large (>10 ha)	4	5
Income level	<1.50 lakh	20	25
	1.50-3.00 lakh	46	57.5
	>3.00 lakh	14	17.5
Area under dragon fruit	<1 acre	22	27.5
	1 acre	39	48.75
	>1 acre	19	23.75

The study highlights the socio-economic profile of dragon fruit growers in Haryana, revealing that adoption is largely driven by young, educated farmers with small to medium landholdings. Most growers belong to nuclear families and cultivate dragon fruit on limited acreage to diversify income sources. A majority of households earn ₹ 1.50–3.00 lakh annually, and nearly half grow the crop on one acre, showing its suitability for small-scale farming. Overall, dragon fruit offers strong income potential, though further support in training, marketing, and post-harvest management is needed.

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Conflict of Interest

The authors have no conflict of interest to declare.

Data Sharing

All relevant data are included in the manuscript.

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