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Potentiality of phytoextracts against anthracnose [*Colletotrichum Gloeosporioides*] of Dragon fruit under *in vitro* condition

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Abstract
The investigation was conducted during October to December 2024-2025 in the Department of Plant Pathology, College of Agriculture, Junagadh Agricultural University, Junagadh, with the objective of assessing the effectiveness of various phytoextracts against *Colletotrichum gloeosporioides* under *in vitro* conditions. Dragon fruit (*Hylocereus* spp.), a member of the Cactaceae family, has recently emerged as a valuable "super fruit" owing to its high nutritional content and notable antioxidant properties. Samples of infected stems and fruits showing typical anthracnose lesions were collected from the farm of the Polytechnic College of Horticulture, Junagadh Agricultural University. The pathogen was isolated through the standard tissue isolation technique and subsequently purified by the hyphal tip method on Potato Dextrose Agar. Based on cultural and morphological attributes, the organism was initially identified as *Colletotrichum* spp., and its identity as *C. gloeosporioides* was later confirmed by SLS Research Station Pvt. Ltd., Surat, Gujarat, with the validation supported by the NCBI GenBank accession number PV800860. Various phytoextracts at different concentrations were tested using the poisoned food technique. Nine phytoextracts at 5, 7.5, and 10 percent concentrations were screened *in vitro* for their suppressive effect on *C. gloeosporioides*. Among them, *Allium cepa* bulb extract demonstrated the highest mycelial inhibition (83.19%) at 10%, closely followed by *Ocimum sanctum* (82.28%), whereas *Datura stramonium* showed the lowest efficacy (36.70%). These *in vitro* evaluations of phytoextracts provided essential preliminary information that may guide the development of effective field-based management strategies for anthracnose in dragon fruit cultivation.

Keywords: Dragon fruit, Anthracnose, *Colletotrichum gloeosporioides*, *Phytoextracts*

Introduction
Dragon fruit is a climbing cactus belonging to the tropical fruit group of the Cactaceae family. It is popularly known as pitaya, pitahaya or strawberry pear. In India, it is called *Kamalam* because its outer appearance resembles a lotus flower. The crop originates from South and Central America and is now grown widely across tropical and subtropical regions. The major cultivated species include *Hylocereus undatus* (red peel with white pulp), *Hylocereus monacanthus* (formerly *H. polyrhizus*, red peel with red pulp), *H. costaricensis* (red peel with purple pulp) and *H. megalanthus* (yellow peel with white pulp), contributing roughly 94%, 4.0%, 1.5% and 0.5% of the global market share, respectively (Wakchaure *et al*., 2020) [12]. The plant is known by several common names, such as dragon fruit, pitaya, pitahaya, night-blooming cereus, strawberry pear, Belle of the Night and Cinderella plant (Perween *et al*., 2018) [11]. In Vietnam, it is referred to as *Thanh Long* (Green Dragon). Among these names, pitaya is widely used and is derived from a Haitian term meaning "scaly fruit," referring to the scale-like bracts on the fruit surface (Valero *et al*., 2025) [13].
The fruit is berry-like in nature, usually ranging from 7-14 cm in length and 5-9 cm in diameter and contains numerous tiny black seeds (Shah *et al*., 2023) [12]. Dragon fruit is recognized for its rich nutrient profile and associated health-promoting properties. Its seeds are a good source of essential fatty acids—particularly linoleic and linolenic acids—which are known to aid digestion and provide a mild laxative effect (Huang *et al*., 2021) [7]. The peel of red dragon fruit also contains significant amounts of important lipids, including linoleic, oleic and palmitic acids (Jiang *et al*., 2021) [9].

Dragon fruit is considered a highly nutritious crop because it contains an array of vital nutrients such as vitamins, minerals, carbohydrates, dietary fiber and natural antioxidants. The edible pulp is especially rich in polysaccharides (Xu *et al*., 2016) [16] and mixed oligosaccharides (Havas *et al*., 2024) [6], which help stimulate the growth of beneficial gut microorganisms like *Lactobacillus* and *Bifidobacterium.*

Dragon fruit is a semi-epiphytic cactus that flourishes in warm, dry tropical or subtropical environments where temperatures typically range between 21-29°C. However, the plant can also endure higher temperatures up to 38-40°C and brief exposure to low temperatures. Optimal growth requires abundant sunlight, 600-1300 mm of rainfall annually and a climate with alternating dry and wet periods. It can grow throughout the year in sandy, well-drained soils with adequate moisture and favorable photoperiod conditions (Jena *et al*., 2024) [8].

Dragon fruit originated primarily from the tropical and subtropical forest regions of Latin America, including North, Central and South America. According to Aztec literature, pitahaya fruits date back to the 13th century. In India, dragon fruit is cultivated in an area of 14,640 ha yielding a production of 55,780 MT in 2024-2025. The major dragon fruit growing states are Andhra Pradesh, Mizoram, Maharashtra, Karnataka, Gujarat, Kerala, Tamil Nadu, Orissa, West Bengal, Tripura, Telangana and Andaman and Nicobar Islands. (Anonymous., 2024) [2]. The major dragon fruit producing regions of the country are areas of Kutch in Gujarat. In Gujarat dragon fruit cultivated in 1214 ha with the production of 4079.3 MT and productivity of 3.36 MT.

One of the major diseases of dragon fruit is anthracnose, which is caused by six *Colletotrichum* species: *C. aenigma*, *C. gloeosporioides*, *C. siamense*, *C. truncatum*, *C. karstii* and *C. fructicola* (Meetum *et al*., 2015, Zhao *et al*., 2018, Abirami *et al*., 2019). *The objective of this study was to evaluate the effectiveness of phytoextracts against Colletotrichum gloeosporioides under in vitro conditions.*

Materials and Methods

The present research work entitled "Potentiality of phyto-extracts against anthracnose [*Colletotrichum gloeosporioides*] of dragon fruit under *in vitro* conditions" was carried out from October to December 2024-2025 at the P.G. Laboratory, Department of Plant Pathology, College of Agriculture, Junagadh Agricultural University, Junagadh, Gujarat. The experiment was conducted using a Completely Randomized Design (CRD) with a factorial concept.

Preparation of Phytoextract

Fresh leaves, rhizomes, cloves and bulbs from nine locally available plant species were thoroughly washed under running tap water, followed by rinsing with sterile distilled water. Each sample was then ground in sterile distilled water at a ratio of 1 ml per gram of tissue (1:1 v/w) using a mortar and pestle. The resulting homogenate was filtered through fine muslin cloth, and the filtrate was centrifuged at 5000 rpm for 10 minutes. The supernatant obtained was further filtered using a sterile funnel fitted with a 1-2 µm pore-size filter. This final filtrate served as the standard (100%) plant extract solution and was stored at 4°C until use. The required concentrations of each plant extract were prepared following the method described by Ansari (1995) [3]. Details of the plant extracts used and their respective concentrations are provided in Table 1.

Evaluation of phytoextracts against C. gloeosporioides *in vitro*

The inhibitory effect of different phytoextracts on the mycelial growth of *Colletotrichum gloeosporioides* was assessed *in vitro* using the poisoned food technique on Potato Dextrose Agar (PDA). The study included nine treatments, each replicated three times and data were analyzed using a Completely Randomized Design (CRD) with a factorial arrangement. To obtain final concentrations of 5%, 7.5% and 10%, specific quantities of each plant extract were incorporated into known volumes of PDA without altering the medium's composition. For every treatment, the mixtures were prepared in 250-ml conical flasks to a final volume of 100 ml and sterilized in an autoclave at 1.055 kg/cm² (15 psi) and 121.6°C for 15 minutes. After sterilization, 20 ml of the molten amended medium was poured into sterile 90-mm Petri plates. Once solidified, each plate was centrally inoculated with a 4-mm disc of actively growing *C. gloeosporioides*, placed mycelial side down to ensure proper contact with the PDA surface. Plates were incubated at 28 ± 1°C, and untreated control plates were maintained for comparison. Radial mycelial growth was recorded, and the percentage inhibition for each treatment was determined using the formula proposed by Vincent (1947) [14].

$$I = \frac{C - T}{C} \times 100$$

Where,

I = Per cent inhibition of mycelial growth

T = Mean radial growth of test pathogen in treated plate (mm) C = Mean radial growth of test pathogen in control plate (mm).

Table 1: Phytoextracts used against *C. gloeosporioides in vitro*

Sr. No.	Botanical name of plant species	Local name	Plant parts used	Concentrations (%)		
1.	*Allium cepa* L.	Onion	Bulbs	5	7.5	10
2.	*Allium sativum* L.	Garlic	Cloves	5	7.5	10
3.	*Azadirachta indica* L.	Neem	Leaves	5	7.5	10
4.	*Zingiber officinale*Rosc.	Ginger	Rhizome	5	7.5	10
5.	*Curcuma longa* L.	Turmeric	Rhizome	5	7.5	10
6.	*Ocimum sanctum* L.	Tulsi	Leaves	5	7.5	10
7.	*Aegle marmelos* L.	Bael	Leaves	5	7.5	10
8.	*Datura stramonium* L.	Datura	Leaves	5	7.5	10
9.	*Adhatodavasica Ness.*	Ardusi	Leaves	5	7.5	10

Results and Discussion

The efficacy of nine naturally available phytoextracts was assessed against *C. gloeosporioides* under *in vitro* conditions. The bioassay was conducted using the Poisoned Food Technique and the experiment was repeated three times to ensure accuracy and reproducibility of the results.

Each phytoextract was evaluated at three concentrations viz., 5%, 7.5% and 10%, along with an untreated control for comparison. Potato Dextrose Agar (PDA) medium was supplemented with the respective phytoextract concentrations and mycelial discs of the pathogen were centrally inoculated onto the plates.

The data presented in Table 2, Plate 1 and Figure 1 clearly revealed that all tested phytoextracts exerted inhibitory effects on the mycelial growth of *C. gloeosporioides* when compared with the untreated control. Among the nine phytoextracts examined *in vitro*, the bulb extract of *Allium cepa* (onion) exhibited the highest antifungal effectiveness, recording a maximum mean mycelial growth inhibition of 71.10%. This treatment proved to be significantly superior to all other phytoextracts tested. The clove extract of *Allium sativum* (garlic) also showed strong antifungal activity, achieving 61.71% mean mycelial growth inhibition and ranking as the second most effective treatment. This was followed by the leaf extract of *Azadirachta indica* (neem), which recorded a mean inhibition of (53.75%). *Ocimum sanctum* (tulsi) leaf extract also demonstrated moderate efficacy with (53.37%) inhibition of mycelial growth. Other phytoextracts such as *Curcuma longa* (turmeric), *Zingiber officinale* (ginger) and *Aegle marmelos* (bael) recorded (52.08%, 49.85% and 45.78%) inhibition, respectively. Among the least effective treatments were the extracts of *Adhatoda vasica* (ardusi) and *Datura stramonium* (datura), which exhibited (38.56%) and (36.70%) mean mycelial growth inhibition, respectively. Overall, these findings suggested that certain phytoextracts especially those from onion and garlic possessed considerable antifungal potential against *C. gloeosporioides* and might serve as promising components in integrated disease management strategies for anthracnose in dragon fruit.

Table 2: Evaluation of phytoextracts against *C. gloeosporioides in vitro*

Phytoextracts	Mycelial growth inhibition (%)			Mean mycelial growth inhibition (%)
	5	7.5	10	
Allium cepa L. (Onion)	52.43 (62.82)	55.10 (67.27)	65.80 (83.19)	57.78 (71.10)
Allium sativum L. (Garlic)	46.62 (52.82)	51.12 (60.60)	57.87 (71.72)	51.87 (61.71)
Azadirachta indica L. (Neem)	37.62 (37.25)	51.77 (61.71)	52.10 (62.27)	47.16 (53.75)
Zingiber officinale Rosc. (Ginger)	32.17 (28.35)	50.47 (59.49)	51.77 (61.71)	44.81 (49.85)
Curcuma longa L. (Turmeric)	40.22 (41.70)	40.87 (42.81)	57.87 (71.72)	46.32 (52.08)
Ocimum sanctum L. (Tulsi)	34.26 (31.69)	42.79 (46.15)	65.10 (82.28)	47.38 (53.37)
Aegle marmelos L. (Bael)	40.12 (41.52)	41.51 (43.92)	46.08 (51.89)	42.57 (45.78)
Datura stramonium L. (Datura)	29.27 (23.91)	40.87 (42.81)	41.19 (43.37)	37.11 (36.70)
*Adhatodavasica*Ness. (Ardusi)	37.40 (36.88)	38.40 (38.59)	39.36 (40.22)	38.39 (38.56)
Mean	31.83 (32.45)	37.54 (42.12)	43.38 (51.67)	-
	Phytoextract (P)	Concentration (C)		P x C
S. Em. ±	0.19	0.10		0.34
C. D. at 5%	0.55	0.30		0.96
C.V.%	1.17			

As the concentration of phytoextracts increased, the mycelial growth inhibition of *C. gloeosporioides* also showed a corresponding increase. All three concentration levels differed significantly from one another in terms of their inhibitory efficacy.

Among these, the highest concentration (10%) of all phytoextracts produced markedly greater inhibition, recording a mean mycelial growth reduction of 51.67%, compared with the lower concentrations. Considering the interaction effect between various phytoextracts and their concentrations, the bulb extract of onion at 10% concentration exhibited the highest mycelial growth inhibition (83.19%) and was significantly superior to the other treatments. The leaf extract of tulsi at 10% concentration ranked next, showing 82.28% inhibition of mycelial growth. The onion bulb extract at 10% (83.19%) was statistically comparable to the tulsi leaf extract at 10% (82.28%). The results obtained in the present study were in close agreement with the findings reported by Bal and Behera (2012) [4], who observed similar trends in the effectiveness of certain plant extracts against *Colletotrichum capsici*, the causal agent of anthracnose or ripe fruit rot in chilli. In their study, various phytoextracts demonstrated significant antifungal activity, thereby supporting the current findings that plant-based treatments could effectively inhibit the mycelial growth of this pathogen. The consistency of results across studies reinforced the potential role of botanical extracts as eco-friendly and sustainable alternatives to synthetic fungicides in integrated disease management strategies for chilli crop.

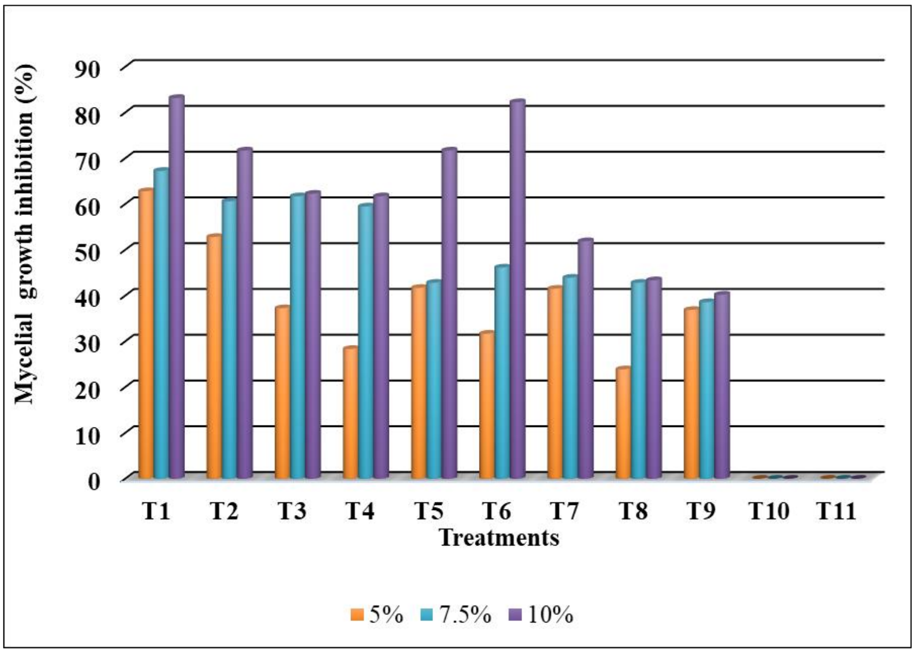

Fig 1: Evaluation of phytoextracts against *C. gloeosporioides in vitro*

Plate 1: Evaluation of phytoextracts against *C. gloeosporioides in vitro*

Treatments

T1: *Allium cepa* L. (Onion)
T2: *Allium sativum* L. (Garlic)
T3: *Azadirachta indica* L. (Neem)
T4: *Zingiber officinale* Rosc (Ginger)
T5: *Curcuma longa* L. (Turmeric)
T6: *Ocimum sanctum* L. (Tulsi)
T7: *Aegle marmelos* L. (Bael)
T8: *Datura stramonium* L. (Datura)
T9: *Adhatoda vasica Ness* (Ardusi)
T10: Control

Conclusions

Among the phytoextracts tested, the bulb extract of *Allium cepa* (onion) showed the greatest mean inhibition of mycelial growth, significantly exceeding the effectiveness of the other plant extracts. In contrast, the leaf extract of *Datura stramonium* at 5% concentration exhibited the lowest activity, causing the least inhibition of *C. gloeosporioides* growth.

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Data Availability Statement
Data will be provided whenever it is needed

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