Research Article

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Studies on weather parameters, screening of cultivars and efficacy of fungicides on *Alternaria alternata* causing leaf spot in bael (*Aegle marmelos*)

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ABSTRACT

Thirteen bael cultivars were evaluated for resistance to Alternaria leaf spot disease. Three cultivars were classified as resistant, nine as moderately resistant, and one as moderately susceptible. The disease prevalence was influenced by weather conditions, particularly temperatures ranging from 27.60°C to 35.75°C, relative humidity between 58.00% and 80.40%, 7.80 hours of daily sunshine and 1.50 mm of rainfall. The disease spread rapidly after seasonal rains, peaking during the 35th meteorological week with 60.60% intensity under temperatures of 24.95°C to 31.80°C and relative humidity of 74.05% to 90.25%. Among all the treatments, Propiconazole (@ 0.1%) was the most effective, reducing disease intensity (7.78%) and 71.41% disease control followed by Difenconazole (@ 0.1%) 10.69% disease intensity and 60.35% disease control, respectively.

Introduction

Bael (*Aegle marmelos* Correa) is one of the most important indigenous fruits of the arid tropics belongs to the Rutaceae family. It is also known as 'Shriphal' and 'Bengal Quince' and is widely cultivated in various parts of Eastern Uttar Pradesh. It has been known in India since prehistoric time and holds significant mythological value. It is considered sacred, dedicated to Lord Shiva and referenced in ancient texts such as the Ramayana, Yajurveda, Buddhist and Jain scriptures. Bael leaves are also traditionally used in worship during the Vinayaka chaturthi festival, particularly for offerings to Lord Ganesha.

The bael fruit is known by various name across different regions: Bengal quince, Indian quince, golden apple, holy fruit, stone apple, Bel, Bela, Sriphal, Belger, Baelpatra, Bilva, maredu, Bilpandu, Bil, Katori, and Maredoa in India; Matum and Mapin in Thailand; Phneover Pnoi in Cambodia; Baunav in Vietnam; Oranger du Malabar in French; and Marmelos in Portuguese. Although the bael tree grows in most Indian states, it is commonly found in Uttar Pradesh, Bihar, West Bengal, Orissa, and Madhya Pradesh. The plant thrives abundantly in Eastern Uttar Pradesh, especially in the districts of Mirzapur, Varanasi, Gorakhpur, Basti, Gonda, Faizabad, Sultanpur, Etawah as well as the Sewan district of Bihar (Teaotia *et al.,* 1963). According to Jauhari and Singh (1971) varieties such as 'Kaghi Etawah,' 'Sewan Large,' 'Mirzapur,' and 'Deoria Large' were found to be superior in taste and quality.

Bael fruits contain 61.5 g of water, 1.8 g of protein, 0.39 mg of thiamine, 1.19 mg of fat, 87 mg of riboflavin, 1.1 mg of niacin, 55 mg of Vitamin A, 85 mg of calcium, 50 mg of phosphorus, 600 mg of potassium, and 8 mg of Vitamin C per 100 grams of the edible portion (Gopalan *et al.,* 1971; Kumar *et al.,* 2023). Among the several fruits containing riboflavin, bael contains highest vitamin B-2 (riboflavin). The ripe fruit serves as a tonic, restorative, astringent, laxative, and is beneficial for the heart and brain. The unripe fruit is recognized for its

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astringent, digestive, and stomachic properties and is often used to treat diarrhea and dysentery. Pitre and Srivastava (1987) studied the anti-diarrheal activity of bael root.

In India, bael trees typically flower in April and May, shortly after new leaves emerge, with fruit maturing between March and June of the following year. Bael has a rich history of ethno-medicinal uses and is recognized as a valuable source of medicine for various human and animal ailments (Kala, 2006). Different parts of the bael plant can be employed to address numerous health issues, including diabetes, liver toxicity, fungal and microbial infections, inflammation, fever, and pain relief (Sharma *et al.,* 2011; Purohit and Vyas, 2005). The fruit is utilized in pickles and sweetened preserves known as murabba when unripe, and once ripe, it is processed into jellies, shakes, and juices, providing a cooling effect, especially during summer. Bael fruit is generally resistant to pests and diseases, though it can be affected by fungi during storage (Orwa *et al.,* 2009). Observations of conidiophores showed dark structures with chains of conidia, which had both cross and longitudinal septa, characteristic of *Alternaria* species. The average size of the conidia ranged from 25-60 × 10-16 µm. The first documentation of Alternaria blight on bael occurred in the Eastern Plateau and Hill region of India (Maurya *et al.,* 2016).

Nursery plants are prone to several fungal foliar diseases, such as root rot caused by *Fusarium solani*, leaf spot caused by *Myrothecium roridum (*Kumar, S. and Singh, H.K. (2021) and *Alternaria alternata* (Maurya *et al.,* 2016 and Singh *et al.,* 2023), as well as dieback and leaf spot caused by *Fusarium pallidoroseum* (Anonymous, 2010 and Kumar *et al.* (2021). Among them, Alternaria leaf spot of bael is now becoming an important menace in Uttar Pradesh and other parts of the country. Madaan and Gupta (1985) first time reported leaf spot disease of bael, caused by *Alternaria alternata* (Fr.) Keissler from Hisar, Haryana. Hence, present study was undertaken to manage the leaf blight of bael by screening, epidemiological studies and through different contact and systemic fungicides.

Material and Methods

Field experiments were conducted on a bael cultivar at the Main Experimental Station latitude 26.55^0 N and longitude 81.84^0 E, Department of Fruit Science, Acharya Narendra Deva University of Agriculture & Technology, Kumarganj, Ayodhya during the year 2021-22 and 2022-23 season. The experiment was conducted in Randomized Block Design with three replications to assess the resistance in bael cultivars, along with epidemiological studies and effect of fungicides for Alternaria leaf spot. The treatments included: T_1= Copper Oxychloride 50% WP at 0.2%; T_2= Mancozeb 75% WP at 0.2%; T_3= Carbendazim 50% WP at 0.2%; T_4= Propiconazole 25 EC at 0.1%; T_5= Difenconazole 35 EC at 0.1%; T_6= Trifloxystrobin at 0.1%; T_7= Antracol at 0.2%; T_8= Iprodione at 0.2%; T_9= Fludioxonil at 0.1%; and T_{10} = Control. Each treatment was applied in 8x8m^2 distance with three replications. Fungicides were applied in three foliar sprays at 15 days interval, starting with the first spray immediately after disease onset. Disease development was assessed 20 days after the final spray, and per cent disease index (PDI) was calculated using a 0-5 rating scale.

Table 1. Scale for disease intensity

Rating	Average disease intensity in (%)	Per cent area covered with disease infection	Disease Reaction
0	0%	No infection	Immune
1	0.1-5 %	0.1-5% area covered	Resistant
2	5.1-20 %	5.1-20% area covered	Moderate Resistant
3	20.1-50 %	20.1-50% area covered	Moderately Susceptible
4	50.1- 75%	50.1-75% area covered	Susceptible
5	75.1-100%	75.1% or above	Highly Susceptible

Per cent disease intensity and per cent disease control were calculated by using the following formula (Vincent, 1947).

$$\text{Per cent disease incedence}= \frac{\text{No of infected Leaves}}{\text{Total no of examined leaves}} X100 \frac{\text{No of infected Leaves}}{\text{Total no of examined leaves}} X100$$

$$\text{Per cent disease index}= \frac{\text{Sum of all numerial rating scale}}{\text{Totl no of examined Leves x maximum grade}} X100$$

$$\frac{\text{Sum of all numerial rating scale}}{\text{Totl no of examined Leves x maximum grade}} X100$$

$$\text{Per cent disease control}= \frac{\text{\% disease of Control} - \text{\% disease of Treatent}}{\text{\% disease of Control}} X100$$

Results and Discussion

Thirteen genetically diverse genotypes of bael were tested and per cent disease incidence and severity (%) were recorded in each cultivar for resistance against Alternaria leaf spot of bael caused by *Alternaria alternata* (Table 2 and Fig. 1). Among the cultivars, ND/AH-11 exhibited the lowest average incidence and severity of Alternaria leaf spot, recording 8.99% and 2.42%, respectively. This was followed by NB-4 with 10.37% and 3.34%, ND/AH-8 with 16.59% and 4.14%, NB-9 with 21.85% and 5.17%, and NB-16 with 27.47% and 7.08%, respectively. Conversely, CISH B-2 had the highest incidence and severity, at 70.29% and 22.18%. These findings align with those reported by Singh *et al.* (2023). Among 13 cultivars, 3 were classified as resistant, 9 as moderately resistant, and 1 as moderately susceptible against Alternaria leaf spot (Table 2).

Table 2. Identification of resistance sources against Alternaria leaf spot of bael

S. No.	Variety	Disease incidence (%)		Pooled mean	Disease severity (%)		Pooled mean	Disease reaction
		2021-22	2022-23		2021-22	2022-23		
1	CIAH B 1	57.3	70.58	63.94	11.00	16.35	13.68	MR
2	CISH B 2	63.33	77.25	70.29	21.07	23.30	22.18	MS
3	N B-5	37.65	49.27	43.46	9.73	10.48	10.11	MR
4	N B-9	17.37	26.33	21.85	3.27	7.07	5.17	MR
5	N B-16	22.33	32.6	27.47	5.83	8.33	7.08	MR
6	N B-17	42.33	53.65	47.99	9.82	11.00	10.41	MR
7	Pant Aparna	60.00	73.93	66.97	16.17	17.80	16.98	MR
8	Pant Sujata	48.17	58.55	53.36	11.37	12.17	11.77	MR
9	N B-7	53.37	65.13	59.25	11.70	14.23	12.96	MR
10	N B-4	10.41	10.33	10.37	3.10	3.57	3.34	R
11	ND/AH-8	15.35	17.83	16.59	4.36	3.93	4.14	R
12	ND/AH-17	31.83	44.13	37.98	7.91	8.34	8.13	MR
13	ND/AH-11	7.91	10.08	08.99	1.99	2.86	2.42	R

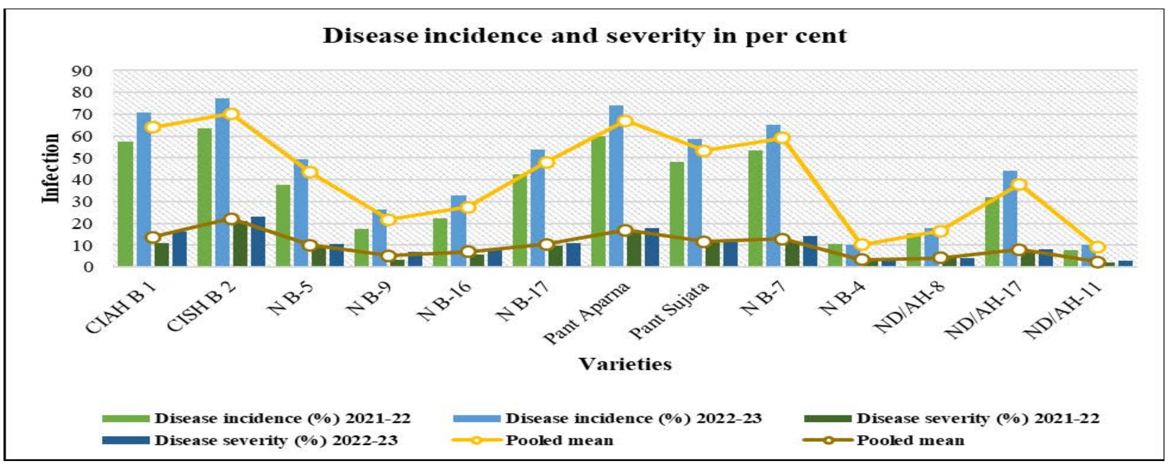

Fig. 1. Screening of bael cultivars against Alternaria leaf spot

Disease development of Alternaria leaf spot of bael in relation to weather parameters

During the first week of July (28th meteorological week), Alternaria leaf spot symptoms were observed on bael nursery plants, starting from the initial growth stage. The symptoms began at the leaf margins and tips, appearing as irregular, light brown spots that gradually darkened to brown and gray. As the disease progressed, several spots merged, forming larger necrotic areas, eventually covering significant portions of the leaves. Under severe conditions, these coalesced spots formed large patches, leading to leaf defoliation (Kumar *et al.*, 2015).

The initial onset of the disease coincided with weather conditions including temperatures ranging between 27.60°C and 35.75°C, relative humidity between 58.00% and 80.40%, daily sunshine of 7.80 hours, and 1.50 mm of rainfall. It was noted that the disease spread rapidly following seasonal rains (Table 3 and Fig. 2).

Table 3. Alternaria leaf spot of bael disease and its correlation with weather parameters

SMW	Date	Max Temp	Min Temp	Max RH	Min RH	Rf	SS	PDI
28	09July - 15July	35.75	27.60	80.40	58.00	1.50	7.85	1.21
29	16July - 22July	34.95	26.90	85.50	67.25	34.40	4.25	3.69
30	23July – 29July	33.40	26.50	89.90	73.30	60.50	2.80	7.13
31	30July – 05Aug.	33.00	25.90	89.05	70.40	12.10	4.20	9.88
32	06Aug. – 12Aug.	32.90	26.20	92.25	73.70	66.80	4.25	11.96
33	13Aug. -19Aug.	33.15	25.80	90.45	75.10	25.30	5.55	14.68
34	20Aug. - 26Aug.	32.20	25.50	92.75	77.95	44.90	5.10	16.87
35	27Aug. - 02Sep.	33.20	25.80	88.40	71.25	9.90	4.75	21.30
36	03Sep. - 09Sep.	34.05	25.45	89.30	64.25	26.90	5.90	25.05
37	10Sep. - 16Sep.	31.80	24.95	90.25	74.05	52.30	4.70	27.19

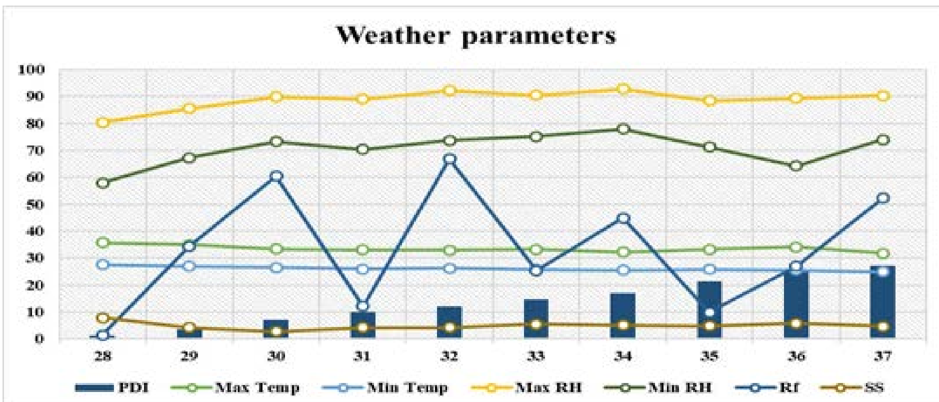

Fig. 2. Alternaria leaf spot of bael disease and its correlation with weather parameters

The highest disease intensity (60.60%) was observed during the 35th meteorological week, when weather conditions included temperatures between 24.95°C and 31.80°C, and relative humidity ranging from 74.05% to 90.25%. Additionally, there were 4.70 hours of sunshine per day and 52.30 mm of rainfall, which favored the first appearance of the disease. A negative and highly significant correlation (at the 1% level) was noted between PDI (Per cent Disease Index) and minimum temperature, while the correlation between PDI and maximum temperature was also negative but only significant. Other weather parameters showed non-significant correlations with PDI, whether positive or negative (Table 4). Similarly, Jayalakshmi *et al.* (2022) and Shivaji *et al.* (2017) recorded disease symptoms in the 29th meteorological week (2nd week of July), with weather conditions of maximum temperature (32.80°C), minimum temperature (27.00°C), morning relative humidity (84.80%), evening relative humidity (70.70%), sunshine hours/day (4.70), and rainfall (28.00 mm). Other studies, such as Ansari *et al.* (1989) and Degenhardt *et al.* (1982), also found that environmental factors, including temperatures of 15-25°C, relative humidity above 80%, and leaf wetness (rain or dew) for 4-24 hours, contributed to disease development and epidemics by supporting pathogen survival (Table 4).

Table 4. Correlation coefficient of PDI of Alternaria leaf spot of bael with weather parameters

	Max. Temp.	Min Temp	Max RH	Min RH	Rf	SS	PDI
Max Temp	1						
Min Temp	0.853**	1					
Max RH	-0.876**	-0.772**	1				
Min RH	-0.881**	-0.632*	0.885**	1			
Rf	-0.513	-0.280	0.654*	0.606	1		
SS	0.496	0.266	-0.587	-0.639*	-0.629*	1	
PDI	-0.658*	-0.917**	0.564	0.376	0.149	-0.038	1

***Correlation is significant at the 0.01 level*
**Correlation is significant at the 0.05 level*
Rf-Rainfall
SS=Sunshine hours/ day

The results of the foliar application of fungicides, presented in Table 5 and Fig. 3 that Propiconazole @ 0.1% recorded the lowest disease intensity with 7.78%. This treatment was statistically comparable to other treatments, including Difenoconazole @ 0.1%, Antracol @ 0.2%, Copper Oxychloride @ 0.2%, Trifloxystrobin @ 0.1%, Iprodione @ 0.2%, Mancozeb @ 0.2%, Carbendazim @ 0.2%, and Fludioxonil @ 0.1%. The corresponding disease intensities for these treatments were 10.69%, 13.99%, 14.79%, 15.77%, 17.88%, 21.34%, 22.33%, and 23.80%, respectively. Highest disease control was observed in Propiconazole @ 0.1% (71.41%), followed by Difenoconazole @ 0.1%, Antracol @ 0.2%, Copper Oxychloride @ 0.2%, Trifloxystrobin @ 0.1%, Iprodione @ 0.2%, Mancozeb @ 0.2%, Carbendazim @ 0.2%, and Fludioxonil @ 0.1%, with corresponding disease control of 60.35%, 48.20%, 45.23%, 41.62%, 33.78%, 21.07%, 17.50%, and 12.18%, respectively.

Similarly, Shivaji *et al.* (2017) found that Propiconazole at 0.1% was the most effective fungicide with disease severity of 7.78%, followed by Difenoconazole at 0.1% (PDI 8.34%) and Mancozeb at 0.2% (PDI 11.96%). Highest disease control (75.12%) was recorded in Propiconazole, followed by Difenoconazole (73.67%) and Mancozeb (57.71%). In another study by Maurya *et al.* (2016), successful disease management was achieved through foliar application of Chlorothalonil 75 WP @ 3g/l, followed by Carbendazim, 50 WP @1-1.5g/l at 15-day intervals. Vishwakarma *et al.* (2021), Mishra *et al.* (2017) and Kumari (2019) found that the lowest disease intensity (16.05%) was observed with Propiconazole at 0.1%, which was statistically similar to treatments with Difenoconazole at 0.1%, Copper Oxychloride at 0.2%, and Mancozeb at 0.2%, which had disease intensities of 17.10%, 18.47%, and 19.18%, respectively.

Table 5. Efficacy of fungicides against Alternaria leaf spot of bael

S No.	Treatments	Disease severity (%)		Pooled mean	Per cent disease control		Pooled mean
		2022	2023		2022	2023	
1	Copper oxychloride @ 0.2%	15.25 (22.96)	14.32 (22.22)	14.79	41.07	49.39	45.23
2	Mancozeb @ 0.2%	20.83 (27.13)	21.85 (27.85)	21.34	19.22	22.92	21.07
3	Carbendazim @ 0.2%	21.24 (27.43)	23.42 (28.92)	22.33	17.71	17.31	17.51
4	Propiconazole @ 0.1%	6.83 (15.13)	8.73 (17.17)	7.78	73.64	69.17	71.41
5	Difenconazole @ 0.1%	11.16 (19.48)	10.22 (18.62)	10.69	56.52	64.18	60.35
6	Trifloxystrobin @ 0.1%	15.91 (23.49)	15.63 (23.27)	15.77	38.21	45.02	41.62
7	Antracol @ 0.2%	14.05 (21.99)	13.94 (21.89)	13.99	45.51	50.89	48.20
8	Iprodione @ 0.2%	18.15 (25.20)	17.61 (24.79)	17.88	29.68	37.87	33.78
9	Fludioxonil @ 0.1%	22.73 (28.46)	24.87 (29.89)	23.80	11.92	12.45	12.18
10	Control	25.87 (30.54)	28.50 (32.24)	27.19	0.00	0.00	0.00
	CD at 5%	1.846	2.020	2.09			6.56
	SE(m)	0.616	0.675	0.71			2.22
	SE(d)	0.872	0.954	1.00			3.13

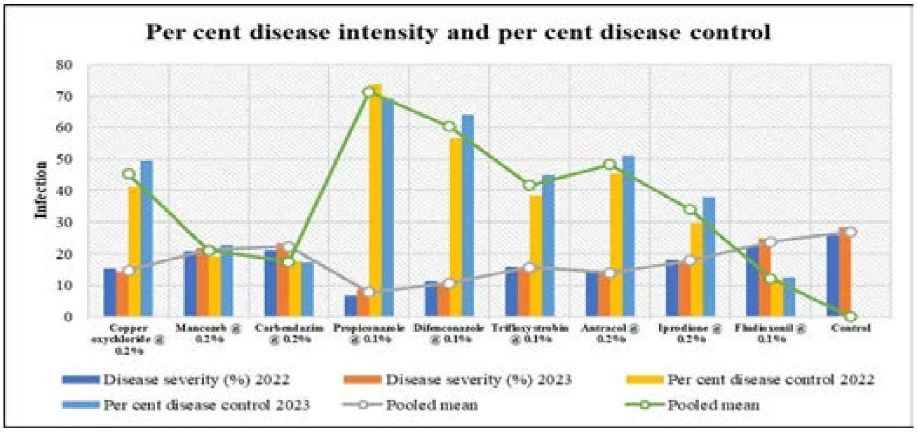

Fig. 3. Effect of fungicides on Alternaria leaf spot of bael

The fungal leaf spot disease causes significant vegetative loss in bael nurseries and is prevalent in this region. An *in vitro* study assessing the efficacy of fungicides on Alternaria leaf spot in bael nurseries showed that all treatments were significantly more effective than the control. Maximum fungal inhibition (100%) was observed with the application of Propiconazole at 0.1%, Difenoconazole at 0.1%, Antracol at 0.2%, and Copper Oxychloride at 0.2%. Similarly, Mishra *et al.* (2017) reported that Mancozeb and Carbendazim achieved 87.05% and 79.76% growth inhibition, respectively. After 72 hours, growth inhibition in Carbendazim and Mancozeb-treated plates was 69.54% and 86.00%, respectively. By 96 hours, complete (100%) growth inhibition was recorded in plates treated with Propiconazole and Difenoconazole, while Carbendazim, Mancozeb, and Copper Oxychloride treatments showed growth inhibition of 65.00%, 82.80%, and 95.32%, respectively.

Conclusion

The evaluation of bael cultivars for resistance to Alternaria leaf spot reveals significant variability in disease susceptibility among the 13 tested cultivars, with three classified as resistant and nine as moderately resistant. Weather conditions play a crucial role in disease prevalence, highlighting the importance of monitoring temperature and humidity levels, especially after seasonal rains when disease intensity peaks. The findings underscore the effectiveness of Propiconazole (0.1%) in managing Alternaria leaf spot, demonstrating a substantial reduction in disease intensity and control. These insights can inform both cultivation practices and disease management strategies, promoting the sustainable production of Bael in arid tropical regions. Further, research is recommended to explore the genetic basis of resistance in the identified cultivars and the long-term efficacy of fungicide treatments.

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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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