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Antifungal potentiality of selected micronutrients, botanicals and fungicides against purple blotch complex pathogens of onion

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Abstract

A field study was undertaken at central research farm of Sher-e-Bangla Agricultural University's (SAU), Dhaka during November 2022 to April 2023.The experiment was configurated in RCBD design to assess the antifungal potentiality of micronutrients (Zinc, Boron, Magnesium), botanicals (Neem, Vet, Nishinda leaves extract) and chemical fungicides (Dithane M-45, Rovral, Ridomil Gold) against purple blotch complex pathogens of onion. Data were recorded on percent disease incidence, percent disease index and different morphological parameters. From the results of the study, it was revealed that among the micronutrients and botanicals, boron and neem gave the satisfactory results. But plots treated with fungicides, Ridomil Gold gave superior performance by lowering percent disease incidence (38.88%) and percent disease index (36%) with higher yield and yield attributes. However, from the findings, it can be concluded that although the chemical fungicides showed the best performance, but in combination of micronutrients and botanicals may be used as bio-rational management.

Keywords: Antifungal potency, botanicals, fungicides, micronutrients, purple blotch complex

1. Introduction

The Onion, scientifically known as *Allium cepa* L. and classified under the *Alliaceae* family, is a crucial element of Bangladeshi cuisine and commonly consumed food items [1]. It is the most commonly grown and consumed spice vegetable crop worldwide [2, 3]. Its distinct and highly valued flavor, aroma, and taste have earned it the title of "Queen of the Kitchen" [4]. The bulb is the onion's primary consumable component, typically grown from seeds [5]. Onions are versatile in their preparation, as they can be consumed raw and sliced in salads or cooked alongside other vegetables and meats. Onion bulbs contain significant nutrients, including phosphorus, calcium, and carbohydrates, as well as protein and vitamin C. being a low-latitude horticultural crop, onions have a relatively brief growing season [3].

The onion is a crucial and ancient crop cultivated in Bangladesh for a long time. It contains the lachrymatory principle, a potent antibiotic (quercetin) with antifungal and antibacterial properties [6]. This antibiotic has several health benefits for humans, including lowering cholesterol levels, acting against cancer and serving as a source of antioxidants [7]. The medicinal properties of the onion have made it a widely grown vegetable and spice crop in Bangladesh. Major onion-growing areas in Bangladesh include Faridpur, Cumilla, Manikgonj, Dinajpur, Jessore, Pabna, Rajshahi, Mymensingh, Jamalpur, and Rangpur [8]. In Bangladesh, the commonly grown onion varieties are Faridpuri and Taherpuri. Although Bangladesh is a significant onion producer, accounting for only 2% of the global production annually, China and India are the world's largest producers, accounting for 26% and 21%, respectively [9]. The Bangladesh Bureau of Statistics (BBS) reported that in our country, onion production amounted to approximately 2517070.76 mt from 507557.05 acres of land in 2022 [8]. Even though the average standard onion production in Bangladesh is 18 tons per hectare, the average yield is only 11 tons per hectare [9]. Bangladesh imports significant onions from India, Egypt, Turkey, Myanmar, and Pakistan, resulting in a significant outflow of foreign currency. Unfortunately, the onion crisis has recently worsened, and prices have skyrocketed. The government has imposed a 5% import duty on this year's (2020-2021) budget [9]. According to the Bangladesh Bureau of Statistics [10], multiple factors contribute to our country's low productivity of onions. However, the primary factor is related to disease problems. The most significant diseases that affect onions worldwide are purple leaf blotch (PLB) and *stemphylium* leaf blotch (SLB), which can cause severe damage to the foliage, leading to significant crop losses in both bulb and seed production. Various studies have documented these losses, ranging from 30 to 100% [11, 12, 13]. It has been observed that *Stemphylium vesicarium* is responsible for initiating the infection that causes the disease, and it facilitates the subsequent infection of *Alternaria porri*, leading to purple blotch. This disease is known as the purple blotch complex.

In Bangladesh, few efforts have identified effective strategies for managing this disease in onion bulb and seed production [14, 15, 16]. Moreover, there is a growing global awareness of the environmental damage caused by excessive and indiscriminate use of harmful chemicals. Therefore, it is crucial to use fungicides judiciously in order to protect nature and prevent environmental pollution. In order to protect the environment and preserve nature, it is crucial to use fungicides wisely. Various fungicides such as Chlorothalonil 75% WP, Mancozeb 75% WP, Propineb 70% WP, Difenoconazole 25% EC, Propiconazole 25% EC, and Hexaconazole 5% EC have been used globally and proven to be effective against plant diseases [17]. However, many fungicides still need to be tested for their effectiveness against these diseases. Nutrients play a significant role in the growth and development of plants, as well as disease management. All the essential nutrients are vital, and some, such as Zn, B, Mn, Mg, and Mo, are particularly sensitive in onion production [18, 19]. Zinc and boron are especially important for promoting plant growth through the biosynthesis of endogenous hormones [20]. Apart from fungicides and nutrients, it has been observed in laboratory and field trials that extracts from various parts of higher plants have antifungal, antibacterial, and insecticidal properties [21, 22]. For example, extracts from neem, nishinda, vet and eucalyptus globules leaves have been effective in controlling various fungal plant pathogens, including *Alternaria alternata*, *Alternaria porri*, *Aspergillus flavus*, *Fusarium oxysporum. f.* sp. *phaseoli*, *Fusarium solani*, *Monilinia fructicola*, *Rhizoctonia solani*, and *Sclerotinia sclerotiorum*. Based on the results of previous reports, the present investigation was aimed to evaluate the effectiveness of various micronutrients, fungicides and botanicals in controlling the purple blotch complex disease of onion through the estimation of percent disease incidence and disease index.

2. Materials and Methods

2.1 Experimental site and duration

The field experiment was carried out in the central research field and lab research in the Molecular Biology and Plant Virology Laboratory (MBPVL) at Sher-e-Bangla Agricultural University (SAU), Dhaka. The experiment was carried out during the winter season from November 2022 to April 2023.

2.2 Experimental design

A randomized complete block design (RCBD) was followed to conduct the field experiment with three (3) replications. Size of each unit plot was 2.0 × 2.5 m^2. There was a total of 27 plots with the following spacing maintained: plant to plant: 20 cm, row to row: 30 cm, and plot to plot (lengthwise and breadthwise): 0.5 m & 0.65 m.

2.3 Treatments

In total nine treatments were codified for conducting the research work.

Table 1: List of the treatments used in the experiment

T_1 = Zinc (1g/L)	T_4 = Rovral 50 WP (2g/L)	T_7 = Neem leaves extract (1:4 w/v)
T_2 = Boron (1.5g/L)	T_5 = Ridomil gold MZ 68 WDG (5g/L)	T_8 = Nishinda leaves extract (1:4 w/v)
T_3 = Magnesium (1.56g/L)	T_6 = Dithane M-45 (5g/L)	T_9 = Vet leaves extract (1:4 w/v)

2.4 Planting material

For this experiment, a particular type of onion variety namely “Taherpuri” was chosen because of its appealing size, popularity, and ability to fetch a higher price in the market but susceptible to purple blotch complex disease.

2.5 Collection and transplanting of onion seedlings

Healthy onion seedlings with 45 days old were collected from Manikganj, Bangladesh. The seedlings were transplanted into the main experimental plot in the evening while maintaining a distance of 30 cm between rows, 20 cm between individual plants, and 0.5 m & 0.65 m (lengthwise & breadthwise) between adjacent plots.

2.6 Preparation of micronutrients

Micronutrients (zinc, boron, and magnesium) were collected from SAU farm. Micronutrients were prepared with the concentration of 1g/L,1.5g/L,1.56g/L for zinc, boron and magnesium respectively [23].

2.7 Preparation of fungicides

The fungicides (Dithane M-45, Rovral 50 WP and Ridomil Gold MZ 68 WDG) were collected from Siddik bazar, Dhaka, Bangladesh. Fungicidal solution was made at recommended dose by combining the required amount of chemical with sterilized water. The concentrations of the fungicidal spray solution were applied which present in Table 2.

Table 2: The concentration of the fungicidal spray solution

Sl. No.	Trade name	Group	Concentration
1.	Rovral 50WP	Iprodione	2 g/L
2.	Ridomil Gold MZ 68 WDG	Mancozeb + Metalaxyl 68% a.i/Kg	5 g/L
3.	Dithane M- 45	Mancozeb 75% WP	5 g/L

2.8 Preparation of botanical extract

Nishinda, neem, and vet leaves were collected from Sher-e-Bangla Agricultural University to obtain the botanical extracts. The extracts were prepared using the method outlined by Ashrafuzzaman and Khan (1992) [24], whereby 100 g of plant parts were added to 400 ml of distilled water to maintain a ratio of 1:4 (w/v).

2.9 Application of treatments

The treatments chosen were applied as foliar spraying techniques for administering the experiment. The spraying process began on the 20th day following transplanting when the plants were 65 days old. A total of three spraying sessions were conducted and each session was taking place at 10 days interval.

2.10 Isolation of causal agents
The infected leaves of onion were collected from the experimental field. Then leaf samples were thoroughly cleaned under running tap water. The leaves were sliced into small pieces, both damaged and healthy portions should be present in these small pieces. The small pieces were placed on the blotter paper and this work was done under a laminar airflow cabinet. Then, plates were incubated at (25±1) °C temperature until the pathogenic growth was observed. During incubation, the blotter paper was sprayed with distilled water to maintain a moist condition.

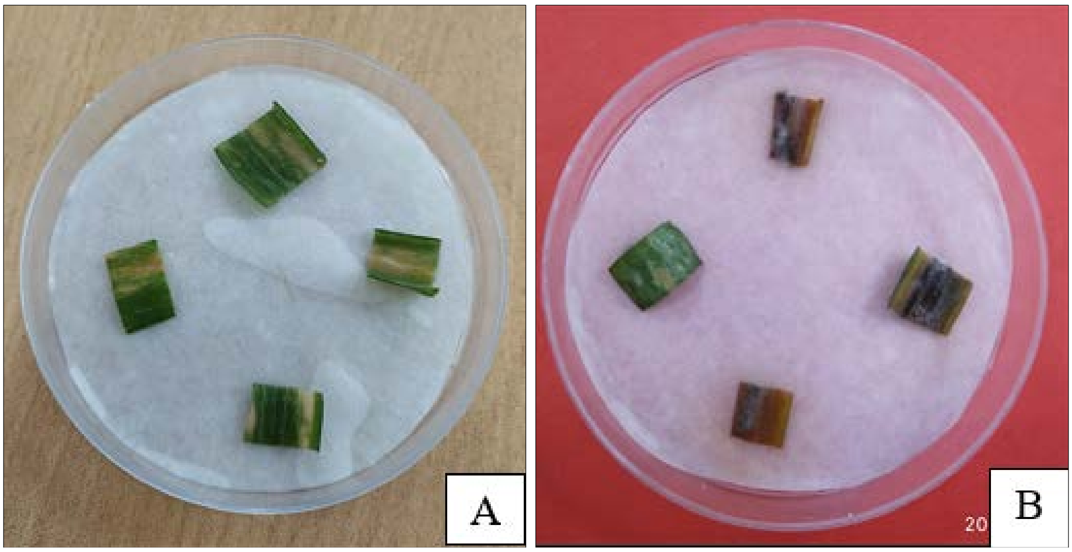

Fig 1: Blotter paper method (A) before incubation and (B) After incubation

2.11 PDA media preparation
A PDA (potato dextrose agar) medium was used to isolate fungi. The media was prepared according to standard protocol.

2.12 Pure culture preparation
Using the sterile inoculating needle, fungal feature, mycelia or conidia, was extracted from the blotter paper samples and transferred to the media. The plate was then sealed and incubated upside down at (25±1) °C temperature until the mycelial growth was observed. Each fungus was treated in the same way. Each time the needle was used, it was heated in a spirit lamp flame until red; at this point, it was removed and allowed to cool before being cleaned again. The principal site for the inoculation procedure was performed in the laminar airflow cabinet.

2.13 Identification of fungus
Pure cultures were made from the blotter plates to purify the fungal isolates. Fungi slides were produced with a needle and forceps, as well as apparent tapes, and then placed beneath the lens of a compound microscope. The slides were examined with a 10X lens and a 40X lens for a more complete examination. Under the microscope, the mycelial and conidial features were observed more closely on the pure culture plates and the fungus was identified on the basis of morphological characteristics.

2.14 Data collection
During the experiment, data were collected by observing diseased plants in the field and recorded data of the observations in a notebook according to specific guidelines. Five plants were randomly selected from every plot and marked using sticks. Data collection began after the onset of the disease and continued until maturity at 10-day intervals. The following parameters were recorded for data collection.

2.14.1 Measurement of disease incidence and percent disease index
Disease incidence was calculated by the following formula [25, 26]:

$$\text{Disease incidence (\%)} = \frac{\text{Number of infected plants}}{\text{Number of inspected plants}} \times 100$$

The level of disease severity in the chosen plant per plot in the field was visually assessed by utilizing a scale ranging from 0 to 5 [27].
After that the percent disease index (PDI) was calculated by using the following formula-

$$\text{PDI} = \frac{\text{The sum of total disease ratting}}{\text{Total no. of observation} \times \text{Maximum grade in the scale}} \times 100$$

2.16 Counting of leaves number per plant
Randomly five chosen diseased plants were selected for data collection of a specific parameter. Data were collected at 10 days interval from 70 days after transplanting (DAT) up to 90 DAT.

2.17 Measurement of yield attributes of onion
Bulb diameter (cm), Weight (fresh and dry) in Kg per plot, Individual bulb weight (g) and Yield (t ha^{-1}) were measured. After harvesting, data on the weight (kg) of freshly collected bulbs from each plot were recorded. The bulbs' dry weight (kg) was obtained by sun-drying fresh samples for a few days and then measuring them with the help of a weight machine.

2.18 Statistical analysis
The most recent edition of the software "Statistix-10" was employed to analyze the data. The data was compared based on the LSD test at a 5% level of significance using the mean value.

3. Results and Discussion

3.1 Effect of different treatments on percent disease incidence (DI) and percent disease index (PDI): In this study, some selected micronutrients (Zinc, Boron, Magnesium), botanicals (Neem leaves extract, Nishinda leaves extract, Vet leaves extract) and chemical fungicides (Dithane M-45, Rovral, Ridomil gold) were assessed to evaluate the antifungal potentiality against the pathogens of purple blotch complex of onion. For evaluation of the antifungal potentiality, percent disease incidence and percent disease index were estimated at 70, 80 and 90 DAT. Among the treatments of micronutrients, the highest disease incidence (51.38%, 54.16% and 56.94%) and percent disease index (38%, 44% and 52%) were recorded in magnesium treated plots at different DAT which was statistically similar and identical with zinc treated (DI=50.16%,53.23% & 54.62%; PDI=34%,40% & 50%) plots and the lowest disease incidence (DI=45.36%,49.07% &51.84%) and PDI (28%,36.26% & 42%) was recorded in boron treated plots at different DAT. These results agreed one recent report where the combination of micronutrients @ 0.25% followed by borax @ 0.5% at 30 and 45 DAPS is superior and boron gave the best performance in suppressing the purple blotch complex [28]. Among the treatments of fungicides, the highest disease incidence (38.88%, 42.12% & 44.44%) and PDI (26.33%, 36.00% & 40.00%) were recorded in Rovral treated plots at different DAT which was statistically identical and similar with Dithane M-45 treated (DI=34.30%,37.49% & 39.81%;PDI=24.66%,34.00% & 38.00%) plots at different DAT and the lowest disease incidence (33.33%, 36.10% & 38.88%) and PDI (24.00%, 30.00% & 36.00%) were recorded in Ridomil gold treated plots showing the best heath condition of onion seedlings [29]. Reported comparable results, where Ridomil MZ (Metalazyl + Mancozeb) was identified as the most efficient fungicide in decreasing disease incidence and severity and boosting the yield of bulbs and seeds [30]. Similar results also found in reported work, where Hexaconazole and Mancozeb + Cymoxanil were the most effective in managing the complex disease of onion, resulting in percent disease control (PDC) of 84.45% and 80%, respectively. Among the treatments of botanicals, the highest disease incidence (63.14%, 66.58% & 69.16%) and PDI (40.00%, 48.00% & 55.33%) were recorded in Vet leaves extract treated plots at different DAT which was statistically identical and different with Nishinda leaves extract treated (DI=60.18%,62.96% & 65.27%; PDI=32.66%,42.00% & 50.00%) plots and the lowest disease incidence (56.94%,59.25% & 61.10%) and PDI (32.00%,38.00% & 46.00%) were recorded in Neem leaves extract treated plots. These findings are in close conformity with those earlier reported by [31] and [32] discovered that neem, mehandi, and ginger extracts were efficient for controlling *Stemphylium* blight of onion, garlic and blights in other crops caused by *A. porri* (Table 3).

Table 3: Efficacy of different treatments on percent disease incidence (DI) and percent disease index (PDI) of purple blotch disease of onion

Treatments	DI (%) at			PDI (%) at		
	70 DAT	80 DAT	90 DAT	70 DAT	80 DAT	90 DAT
T_1(Zinc)	50.16 bc	53.23 bcd	54.62 bcd	34.00 abc	40.00 bcd	50.00 bc
T_2(Boron)	45.36 cd	49.07 cd	51.84 cd	28.00 cde	36.26 de	42.00 de
T_3(Magnesium)	51.38 bc	54.16 bc	56.94 bc	38.00 ab	44.00 ab	52.00 ab
T_4(Rovral)	38.88 de	42.12 de	44.44 de	26.33 de	36.00 de	40.00 ef
T_5(Ridomil gold)	33.33 e	36.10 e	38.88 e	24.00 e	30.00 f	36.00 f
T_6(Dithane M-45)	34.30 e	37.49 e	39.81 e	24.66 e	34.00 ef	38.00 ef
T_7(Neem leaves extract)	56.94 ab	59.25 abc	61.10 abc	32.00 bcd	38.00 cde	46.00 cd
T_8(Nishinda leaves extract)	60.18 ab	62.96 ab	65.27 ab	32.66 bc	42.00 bc	50.00 bc
T_9(Vet leaves extract)	63.14 a	66.58 a	69.16 a	40.00 a	48.00 a	55.33 a
LS	**	**	**	**	**	**
LSD(0.05)	10.45	11.51	11.40	6.31	4.83	4.29
CV(%)	12.53	12.99	12.30	11.74	7.22	5.45

In a column means having a similar letter(s) are statistically identical, and those having dissimilar letter(s) differ significantly as per 0.05 level of probability, LS = level of significance, ** = Significant at 1% level of probability.

3.2 Identification of the causal organisms of purple blotch complex on the basis of cultural and morphological characters

Two fungal pathogens; *Alternaria porri* and *Stemphylium vesicarium* were identified and characterized on the basis of their cultural and morphological characteristics.

In case of *A. porri*, different cultural characters like fast growing, slightly fluffy, uniformly spreading, slightly depressed at the centre with distinct concentric zonation of light to dark brown, off-white to olive green colour were observed. The majority of the isolates of *A. porri* grew fluffy on potato dextrose agar with colony colors ranging from pinkish white through dull orange to olivaceous and black and definite to diffuse zonation patterns [33]. The similar cultural characteristics also studied in recent published report. From the microscopic study, it was observed that the fungal mycelium was first hyaline, then became pale brown to olivaceous brown or smoky with a purple tint. The hyphae were septate and irregularly branched. Conidiophores appeared alone or in clusters, straight or flexuous light to mid brown, septate and blunt at the ends. Conidia were single, straight to curved, obclavate, muriform, and light to golden brown in color. Maximum longitudinal septation of conidia was observed in *Alternaria porri* having a prominent conidial beak (Figure 1). These findings are consistent with those previously reported by [34-37].

In case of *Stymphylium vesicarium*, the colony color ranged from greenish brown to dirty white, deep grey to whitish, light grey to whitish, deep greenish white, light grey and dirty white to greenish with elevation ranging from umbonate to raise to flat and texture ranging from cottony, fluffy to velvety. These findings coincided previously by [38]. From the microscopic study, it was observed that the fungal mycelium was initially sub-hyaline, then pale brown to dark brown. Conidiophores were unbranched, dispersed, straight to flexuous, smooth, septate, and light to medium brown in color. Mature conidia were oblong to broadly elliptic, thickly verrucose, golden brown to olive brown rounded at the ends and having reduced or absent conidial beak.Maximum transverse septation of conidia was recorded in *Stemphylium vesicarium* (Figure 2). These findings align with those of [39-41].

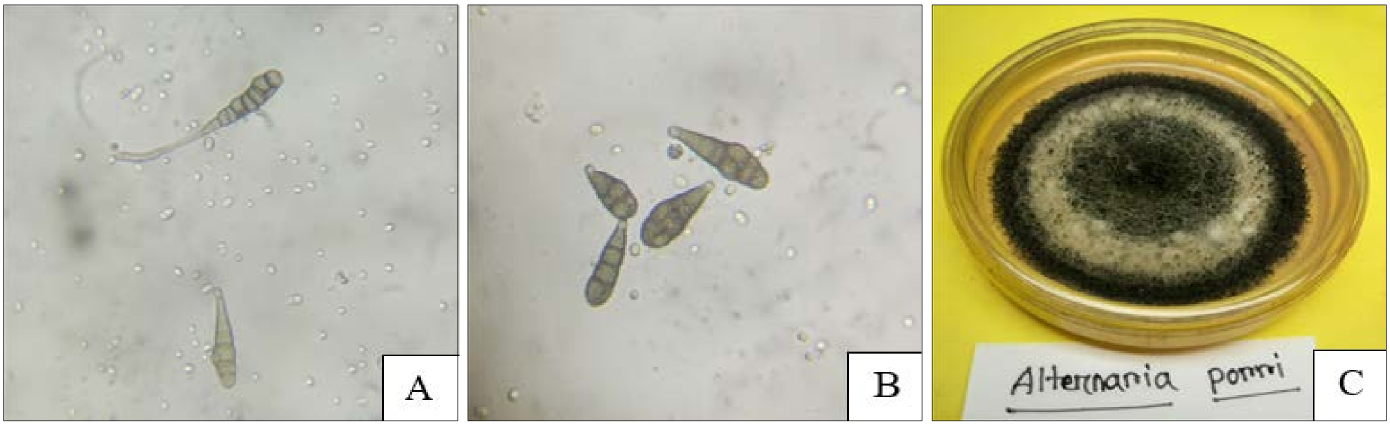

Fig 2: (A) Conidia of *Alternaria porri* (10x); (B) Conidia of *Alternaria porri* (40x); (C) Pure culture of *Alternaria porri*

Fig 3: (D) Conidia of *Stemphylium vesicarium* (10x); (E) Conidia *of Stemphylium vesicarium* (40x); (F) Pure culture of *Stemphylium vesicarium*

3.3 Effect of different treatments on number of leaves per plant: The number of leaves per plant varied significantly among treatments (Table 4). At 70, 80 and 90 DAT, the onion seedlings treated with Ridomil gold had a significant impact in producing the highest number of leaves (4.53, 7.00 and 7.16) and it was observed that the seedlings in the plot treated with vet leaf extract treatment had the lowest number of leaves (3.60, 5.70 and 5.90) at all data recording dates. However, the remaining treatments gave a moderate number of leaves in all observations [23]. Almost found similar results and they reported that onion seedlings treated with ridomil gold fungicides had provided maximum number of leaves per plant.

Table 4: Efficacy of different treatments on the number of leaves per plant

Treatments	Number of Leaves at		
	70 DAT	80 DAT	90 DAT
T_1(Zinc)	4.33 ab	6.53 ab	6.60 ab
T_2(Boron)	4.20 ab	6.56 ab	6.70 ab
T_3(Magnesium)	3.80 bc	6.10 ab	6.50 ab
T_4(Rovral)	4.33 ab	6.60 ab	6.86 ab
T_5(Ridomil gold)	4.53 a	7.00 a	7.16 a
T_6(Dithane M45)	4.30 ab	6.43 ab	6.60 ab
T_7(Neem leaf extract)	4.03 abc	6.60 ab	6.60 ab
T_8(Nishinda leaf extract)	4.00 abc	6.30 ab	6.40 ab
T_9(vet leaf extract)	3.60 c	5.70 b	5.90 b
LS	**	**	**
$LSD_{(0.05)}$	0.59	0.96	1.06
CV (%)	8.33	8.67	9.34

In a column means having a similar letter(s) are statistically identical, and those having dissimilar letter(s) differ significantly as per 0.05 level of probability, LS = level of significance, ** = Significant at 1% level of probability.

3.4 Effect of different treatments on yield and yield attributes of onion

The effects of different treatments on onion single bulb weight and diameter, fresh and dried bulb weight and yield varied significantly (Table 5). The highest single bulb weight (43.53 g), bulb diameter (14.64 cm) were recorded in treatment T_3 (Ridomil gold) which were statistically alike to treatment T_1 (Zinc; bulb weight 43.10 g and bulb diameter 14.53 cm respectively) and T_4 (Rovral; bulb weight 42.16 g and bulb diameter 14.54 cm respectively). The highest fresh weight (3.03 kg plot^{-1}), dry weight (2.80 kg plot^{-1}), yield (5.61 tha^{-1}) were recorded in T_5 (Ridomil gold). On the other hand, the lowest single bulb weight (20.40 g) and bulb diameter (10.99 cm), fresh (1.45 kg plot^{-1}) and dry weight (1.44 kg plot^{-1}), yield (2.88 t ha^{-1}) were recorded in the T_9 (vet leaf extract) treatment [23]. Almost found similar results and they reported that onion seedlings treated with Ridomil gold fungicides had increased the yield of the onion by 54.34% [30]. Reported similar results, where Hexaconazole and Mancozeb + Cymoxanil resulted in the highest yield of 878.7 kg/ha and 878.3 kg/ha, respectively, and the highest thousand seed weight of 3.72 gm and 3.64 gm, respectively. Similarly [42], observed a 71.95% reduction in disease incidence in plots treated with fungicides compared to the control. Furthermore, they found a 10.6% and 50.9% increase in bulb weight and yield per plot in the fungicide-treated plots. The finding also supported the findings of [43] who have reported that spraying Ridomil MZ-72 (0.2%) or Rovral 50WP (0.2%) at 7-day intervals reduced the incidence and severity of purple blotch complex of onion and boosted bulb production.

Table 5: Efficacy of different treatments on yield attributes and yields of onion

Treatments	Single bulb weight (g)	Bulb diameter (cm)	Fresh weight (kg plot^{-1})	Dry weight (kg plot^{-1})	Yield (t ha^{-1})
T_1(Zinc)	43.10 a	14.53 a	2.61 ab	2.48 ab	4.96 ab
T_2(Boron)	38.70 a	13.34 ab	2.11 cd	1.97 cd	3.94 cd
T_3(Magnesium)	28.90 b	11.68 bc	2.27 bc	2.18 bc	4.36 bc
T_4(Rovral)	42.16 a	14.54 a	2.75 a	2.48 ab	4.96 ab
T_5(Ridomil gold)	43.53 a	14.64 a	3.03 a	2.80 a	5.61 a
T_6(Dithane M45)	41.90 a	13.31 ab	2.69 ab	2.45 ab	4.91 ab
T_7(Neem leaf extract)	24.10 b	11.20 c	2.01 cd	1.84 cde	3.68 cde
T_8(Nishinda leaf extract)	23.10 b	11.18 c	1.71 de	1.63 de	3.27 de
T_9(Vet leaf extract)	20.40 b	10.99 c	1.45 e	1.44 e	2.88 e
LS	**	**	**	**	**
$LSD_{(0.05)}$	9.35	1.82	0.45	0.44	0.88
CV(%)	15.90	8.22	11.47	11.87	11.89

In a column means having a similar letter(s) are statistically identical, and those having dissimilar letter(s) differ significantly as per 0.05 level of probability, LS = level of significance, ** = Significant at 1% level of probability.

3.5 Relationship between percent disease index (PDI) and yield of onion

A correlation study was conducted to determine the relationship between the percent disease index and yield of onion at 70 DAT, 80 DAT and 90 DAT. From the relationship studies, it was revealed that the percent disease index negatively correlated with the yield of onion which are clearly depicted in figure 4-6.

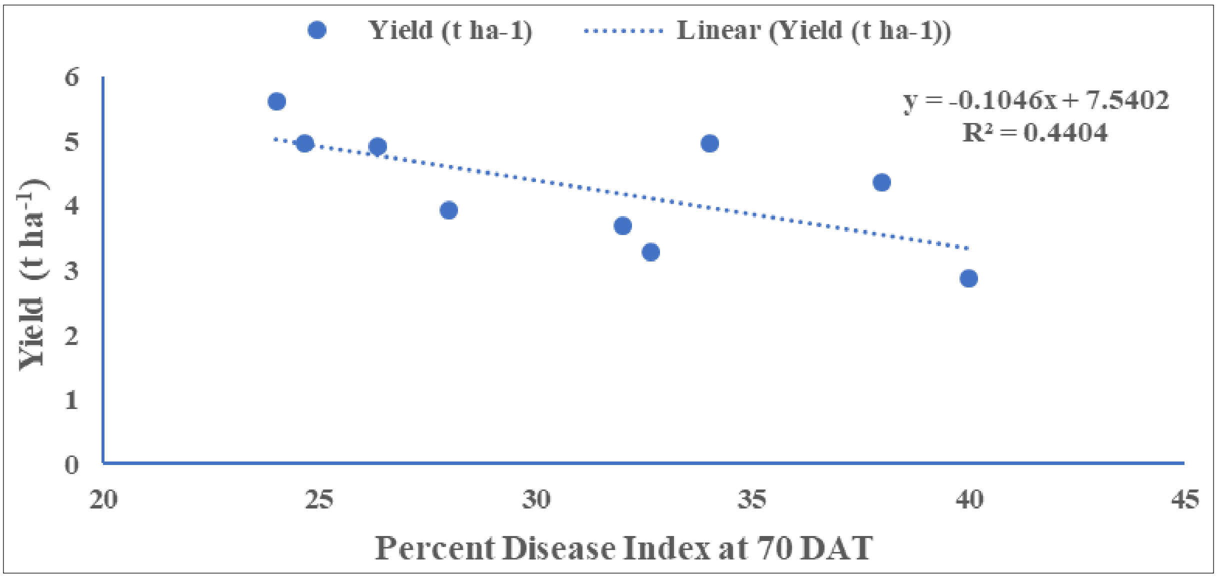

Fig 4: Relationship between PDI and yield of onion at 70 DAT

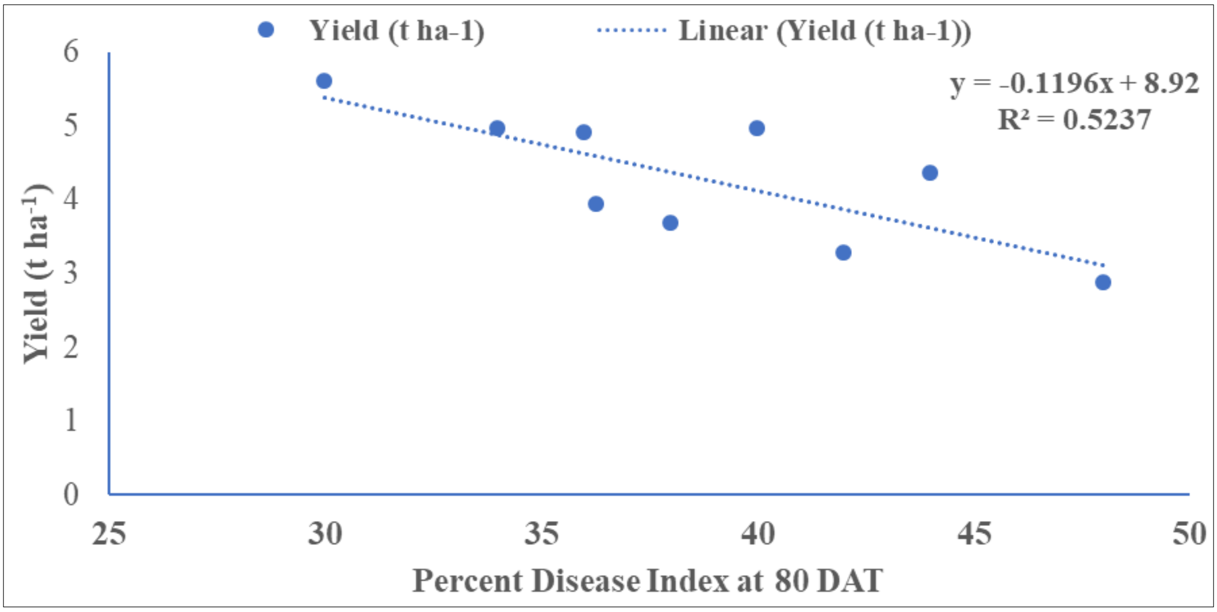

Fig 5: Relationship between PDI and yield of onion at 80 DAT

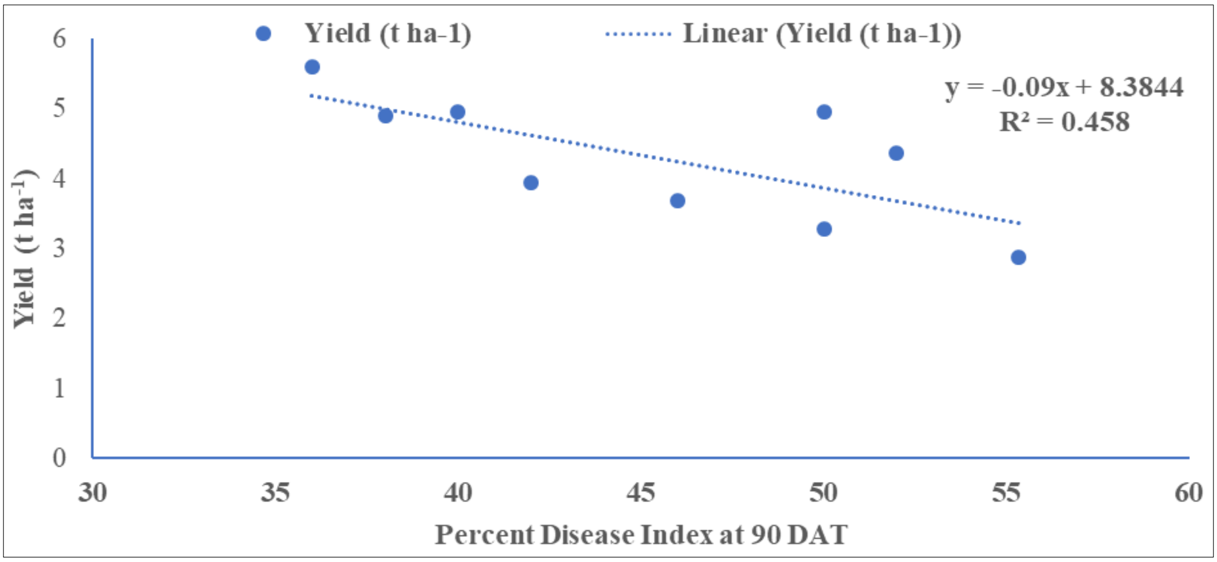

Fig 6: Relationship between PDI and yield of onion at 90 DAT

4. Conclusion

In the present study, it was revealed that evaluated micronutrients and botanicals gave the satisfactory results, but plots treated with fungicides gave superior performance in terms of reducing the percent disease incidence and percent disease index with higher yield and yield attributes. However from the study, it may be concluded that considering the overall results, the combined application of botanical extracts and judicious use of micronutrients may be recommended as an eco-friendly approach for controlling the purple blotch complex of onion.

5. Funding

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6. Conflict of interest

The authors state that they have no conflicts of interest.

7. References

1. Hossain AKMA, Islam MJ. Status of Allium production in Bangladesh. Acta Horticulturae. 1994;358:33-36.
2. Food and Agriculture Organization of the United Nations. World Food and Agriculture-Statistical Yearbook, Rome; c2021. Available from: https://doi.org/10.4060/cb4477en. 2021.
3. Brewster JL. Onions and other vegetable alliums. CABI; c1994, 15.
4. Selvaraj S. Onion: Queen of the Kitchen. Kisan World. 1976;3:32-34.
5. Jones HA, Mann LK. Onions and their allies. Soil Science.1964;98:68.
6. Purseglove JW. Tropical crops: Monocotyledons. Longman Press; c1972.
7. Uddin N, Naseer A, Shahid M, Muhammad N, Ali N. *In vitro* evaluation of fungicides and plant extracts to control purple blotch disease of onion in Pakistan. Plant Pathology Journal. 2020;19:77-88.
8. Bangladesh Bureau of Statistics. Yearbook of Agricultural Statistics; c2023 May. Available from: http://www.bbs.gov.bd.2022.
9. Mahmud I. The Prothom Alo. Increased onion production brings hope to farmers. Available from: https://en.prothomalo.com/business/local/increased-onion-production-brings-hope-to-farmers; c2020 Sep 17.
10. Bangladesh Bureau of Statistics. Yearbook of Agricultural Statistics. Available from: http://www.bbs.gov.bd; c2006.
11. Awad MA, El-Shenawy Z, Omran AF, Shatla MN. Cultural practices in relation to purple blotch disease of onion. Scientia Horticulturae. 1978;9(3):237-243.
12. Everts KL, Lacy ML. The influence of dew duration, relative humidity and leaf senescence on the conditional formation and infection of onion by *Alternaria porri*. Phytopathology. 1990;80(11): 1203-1207.
13. Cramer CS. Breeding and genetics of *Fusarium* basal rot resistance in onion. Euphytica. 2000;115:159-166.
14. Rahman ML. Efficacy of fungicides in controlling purple leaf blotch (*Alternaria porri*) of onion (*Allium cepa*). Master's thesis, Plant Pathology; c1990.
15. Ashrafuzzaman M, Ahmed MU. Control of foliage disease of onion by spray fungicides. Bangladesh Hort. 1976;4(2):25-30.
16. Rahman ML, Ahmed HU, Mian IH. Efficacy of fungicides in controlling purple leaf blotch of onion. Bangladesh J Plant Path. 1988;4(1&2):71-76.
17. Bachkar SACPB, Bhalckar MN. Control of *Alternaria porri* Causing Purple Blotch of Onion during Kharif Season. J Allium Res. 2018;1:57-59.
18. Huber DM, Graham RD. The role of nutrition in crop resistance and tolerance to disease, In Rengel Z (ed) Mineral nutrition of crops fundamental mechanisms and implications. Food Product Press, New York; c1999. p. 205-226.
19. Havlin JL, Beaton JD, Tisdale SL, Nelson WL. Soil fertility and fertilizers: An introduction to nutrient management. 6th ed. Prentice Hall, Upper Saddle River; c2007. p. 244-289.
20. Bhatt B, Srevastava K, Singh MP. Studies on the Effect of Foliar Application of Micronutrients on Growth, Yield, and Economics of Tomato (*Lycopersicon esculentum* Mill). Progressive Horticulture. 2004;36:331-334.
21. Ergene A, Guler P, Tan S, Mirici S, Hamzaoglu E, Duran A, *et al*. Antibacterial and antifungal activity of Heracleum sphondylium subsp. artvinense. African J of Biotech. 2006;5:1087-1089.
22. Islam MDM, Begum F, Nahar N, Habiba UA, Fakruzzaman KM. *In-Vivo* Management of Purple Blotch of Onion Caused by *Alternaria porri* (Ellis) Cif. Through

Fungicides. American Journal of Plant Sciences. 2020;11:1847-1859.
23. Kanta SA, Nisha HAC, Ferdous R, Tohura T, Hossain MB. Evaluation the Effect of Fungicides, Micronutrients and Botanicals on Purple Blotch Complex of Onion in Bangladesh. European Journal of Agriculture and Food Sciences. 2023;5(1):97-102.
24. Ashrafuzzaman MH, Khan AR. Antifungal activity *in vitro* of some plant extracts on *Rhizoctonia solani*. Bangladesh J Sci. Res. 1992;10(2):243-244.
25. Agrios GN. Plant Pathology. 5th ed. Elsevier Academic Press, Burlington, Mass; c2005. p. 952.
26. Kranz J. Measuring Plant Disease. In: Experimental techniques in plant disease epidemiology, Springer, Berlin Heidelberg; c1988. p. 35-50.
27. Horsfall JG, Barratt RW. The grading system for measuring plant disease, Phytopathology. 1945;35:655.
28. Biswas P, Das S, Bar A, Maity TK, Mandal AR. Effect of micronutrient application on vegetative growth and bulb yield attributes of rabi onion (*Allium cepa* L.). Int. J Curr. Microbiol. App. Sci. 2020;9(3):556-565.
29. Georgy NI, Radwan JA, Mohamed HA, Shabi AE. Chemical control of downy mildew and purple blotch of onion in Egypt. Agril. Res. Rev. 1986;61:25-41.
30. Paneru N, Pragya A, Puja T. Management of purple blotch complex of onion (*Allium cepa* Cv red creole) under field condition in Rukum-West, Nepal. MJSA. 2020;4(2):71-74.
31. Kumari, L, Shekhawat KS. Rai PK. Efficacy of fungicides and plant extracts against Alternaria blight of periwinkle. J Mycol. Pl. Pathol. 2006;36(2):134-137.
32. Sharma A, Dass A, Paul MS. Antifungal effect of neem extract on some common phytopathogenic fungi. Adv. Pl. Sci. 2007;20(2):357-358.
33. Shahnaz E, Razdan VK, Andrabi M, Rather TR. Variability among *Alternaria porri* isolates. Indian Phytopath. 2013;66:164-167.
34. Ellis MB. Dematiaceous Hyphomycetes. Commonwealth Mycological Institute, Kew, England; c1971. p. 608.
35. Udhand S. Survey for incidence and management of purple blotch of onion. M.Sc. Thesis, Junagadh Agricultural University, Junagadh, Gujrat; c2009.
36. Singh P. Studies on purple blotch (*Alternaria porri* (Ellis) Cif.) of onion (*Allium cepa* L.) and its management. M.Sc. Thesis, Anand Agricultural University, Anand, Gujrat; c2012.
37. Vijayalakshmi M, Madhavi M, Kavitha A. Studies on *Alternaria porri* (Ellis) Cif. pathogenic to onion (*Allium cepa* L.). Arch Appl Sci Res. 2012;4:1-9.
38. Chowdhury NHA. Cultural, morphological and molecular characterization of *Stemphylium vesicarium* causing white blotch of onion. M.Sc. Thesis, Sher-e-Bangla Agricultural University, Dhaka, Bangladesh; c2013.
39. Simmons EG. Perfect states of *Stemphylium*. 1969;61:1-26. DOI: 10.2307/3757341
40. Basallote-Ureba MJ, Prados-Ligero AM, Melero-Vara JM. Aetiology of leaf spot of garlic and onion caused by *Stemphylium vesicarium* in Spain. Plant Pathol. 1999;48:139-45.
41. Arzanlou M, Khodaei S, Babai-Ahari A. *Helianthus annuus* as a natural host for *Stemphylium vesicarium* in Iran. Australas. Plant Dis Notes. 2012;7:167-170.
42. Sultana N, Ayub A, Islam M. Yield loss assessment of onion bulb due to purple blotch disease. Annual Research Report. Plant Pathology Division, BARI, Joydevpur; c2008. p. 40-41.
43. Rahman AM. Study on purple blotch of onion and its management. M.S. Thesis. Department of Plant Pathology. BAU, Mymensingh; c2004.