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Comparative Effectiveness of Various Fungicides in Managing Mustard Clubroot Disease Caused by *Plasmodiophora Brassicae*

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Abstract

The experiment was accomplished to find out which fungicide or fungicides work best against clubroot disease and how well they protect mustard plants. It also looked at how these fungicides affect the yield and other important growth factors of mustard. The study took place in the research field and lab of the Plant Pathology Division at the Regional Agricultural Research Station (RARS), Bangladesh Agricultural Research Institute (BARI), located in Jamalpur-2000, Bangladesh, during the Rabi season of 2024-2025. The experiment took place both in natural field settings and in a controlled lab environment. The study includes 8 (eight) treatments, namely, T_1= Acrobat MZ (Dimethomorph + Mancozeb) @ 2 g/L, T_2= Ridomil Gold MZ 68 WG (Mancozeb + Metalaxyl) @ 2 g/L, T_3= Dolochune @ 5 t/ha, T_4=Luna sensation 50 SC (Fluopyrum + Trifloxystrobin) @ 2 ml/L, T_5 = Amistar Top 325 SC (Azoxystrobin + Difenoconazole) @ 2 ml/L, T_6= Nativo 75 WG (Tebuconazole + Trifloxystrobin) @ 2 g/L, T_7= Blitox 50 WP (Copper oxychloride) @ 2 g/L and T_8= Control. BARI Sarisha-14 was used in this study as test variety. In case of field experiment, only 0.59% of infected plants were found in the control treatment (T_8) where disease severity is 1 and disease status was a few small clubs in the infected plants. Other treatments had no infection (no clubbing). These findings indicate that fungicides application for controlling clubroot disease of BARI Sarisha-14 is not effective because very few percentage infections were found in control treatment under natural field conditions. The highest seed yield per hectare (1768.52 kg) noted in the T_7 treatment followed by the T_8 treatment (1720.37 kg), T_1 and T_2 treatments (1703.70 kg). These findings indicate that although seed yield performance exhibits significant variations among the treatments but only one treatment showed better performances compared to control. On the other hand, two treatments showed at par seed yield performance with the control and four treatments showed inferior performance to control that might be due to the uneven population and land heterogeneity. In case of lab experiment, compared to the control treatment, six fungicides and one soil amendment reduced clubroot disease incidence and severity after six weeks of inoculation.

Keywords: Mustard/rapeseed, Clubroot disease, screening, fungicides, disease incidence (%), disease severity (%), disease severity status, seed yield and artificial inoculation

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INTRODUCTION

The soil-borne disease called clubroot is caused by a type of pathogen called *Plasmodiophora brassicae*, which is a necessary biotrophic organism

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that lives on living plant cells. It is currently documented from every continent in the world and is especially common in temperate climates under a variety of soil environmental stress conditions. Plants stop living completely in a very short time because of serious problems like leaves turning yellow, wilting that can't be reversed, and growth of swollen, club-like structures on the main and secondary roots of Brassica plants **(Figure 1.b**) Baskey S et al, (2018) [1]. This disease has been growing alarmingly in rape and mustard since the early 1980s, when widespread cultivation of these crops began in Bangladesh, particularly in areas with acidic soil. Clubroot disease is right now the biggest and only major problem for growing mustard and rapeseed, especially in areas with acidic soil, where crops can be completely lost. In areas where the disease is common, farmers must stop growing mustard and rapeseed. The virulence and aggressiveness of the pathogen population show significant variation both within and across clubroot-affected fields in different agroclimatic zones of Bangladesh. Bangladesh primarily grows three main oilseed crops: sesame, which is called *Sesamum indicum*, mustard, which belongs to the Brassica family, and groundnut, which is also known as *Arachis hypogaea*. Compared to other countries around the world, Bangladesh produces very little mustard, only 1.32 tons for each hectare of land. The low mustard yield in Bangladesh is caused by several factors. Diseases are among the most important factors that have been identified. Clubroot disease, which is brought on by *Plasmodiophora brassicae*, is a recent and growing threat to mustard. The usual ways to deal with disease in unhealthy soil include using soil improving substances like lime to make the soil less acidic, calcium cyanamide which acts as both fertilizer and a pesticide, and other soil treatments like boron and molybdenum. Also, methods such as heating the soil with the sun and adding compost are commonly used to help the soil get better. It turned out that these management methods didn't always work. Using fungicides can change the physical and chemical properties of soil and affect the types of microorganisms living in the soil Zhang H, et al (2022) [2]. Bacteria play important roles in helping the soil manage nutrients, support plant growth, and stop diseases that come from the soil, all because there are lots of different types of microbes in the vegetable-soil system Zhang H, et al (2020) , Kong X et al (2018) [2, 3]. Fungicide use could damage the stability of the vegetable-soil ecosystem by changing the physicochemical characteristics and microbial populations of the soil either directly or indirectly. Different kinds of soil react differently to fungicides, which affects the bacteria living in the soil and how the soil is structured Zhang H, et al (2022) [2]. During *P. brassicae* infection causes major changes in soil features and the types of microbes present Lebreton L et al (2019) [4]. The effects of applying fungicides to *P. brassicae*-contaminated soil, it is not known how brassicae-contaminated soil affects microbial populations and soil characteristics. However, Bangladesh doesn't have any control methods available. So, it's important to pick an effective fungicide or fungicides that work well to stop mustard clubroot disease from happening. The current study aims to identify effective fungicide or fungicides and evaluate their effectiveness in managing clubroot disease in mustard both in the field and lab conditions.

MATERIALS AND METHODS

Experiments were conducted to find out an efficient fungicide or fungicides and the efficacy of fungicides against clubroot disease and for yield and yield contributing characters of Mustard at the research field and lab of Plant Pathology Division, Regional Agricultural Research Station (RARS), Bangladesh Agricultural Research Institute (BARI), Jamalpur-2000, Bangladesh during the Rabi season of 2024-2025 **(Figure 1.a).** The experiment was conducted under natural field conditions and lab conditions. The study consists of 8 (eight) treatments viz., T_1= Acrobat MZ (Dimethomorph + Mancozeb) @ 2 g/L, T_2= Ridomil Gold MZ 68 WG (Mancozeb + Metalaxyl) @ 2 g/L, T_3= Dolochune @ 5 t/ha, T_4=Luna sensation 50 SC (Fluopyrum + Trifloxystrobin) @ 2 ml/L, T_5 = Amistar Top 325 SC(Azoxystrobin + Difenoconazole) @ 2 ml/L, T_6= Nativo 75 WG (Tebuconazole + Trifloxystrobin) @ 2 g/L, T_7= Blitox 50 WP (Copper oxychloride) @ 2 g/L and T_8= Control. BARI Sarisha-14 was used in this study as test variety. The experiment was arranged in a Randomized Complete Block Design with three repetitions. The size of the plot was 2.0 meters by 1.8 meters. Seeds were planted on November 23, 2024, and they were sown continuously with the rows placed 30 centimeters apart from each other. Once the seedlings started growing, they were thinned out to leave 5 cm of space between each one. Fertilizers were used in the amounts of 120 kg, 80 kg, 60 kg, 40 kg, 4 kg, and 1 kg per hectare

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for nitrogen, phosphorus, potassium, sulfur, zinc, and boron respectively. These were given using Urea, TSP, MOP, Gypsum, Zinc

Figure 1. (a) Field view of the experiment and (b) clubroot infected mustard plant.

sulphate, and Boric acid. In the final part of getting the land ready, half of the urea and every other fertilizer was spread. The leftover urea was applied at the beginning of the flowering period. All the cultural activities among different cultures were done on time to make sure the crops grew at the same rate. Data was collected on days to flowering, days to maturity, plant height, primary branches per plant, secondary branches per plant, number of siliquae per plant, length of siliqua, 1000-seed weight, yield per plot, yield per hectare, total number of plants per plot, and the incidence and severity of clubroot disease in Mustard (percentage of infected plants, disease severity, and disease status). In this study, disease severity was measured on a scale from 0 to 3, where 0 means no clubbing, 1 means a few small clubs, 2 means moderate clubbing, and 3 means severe clubbing. Finally, figure out how well the fungicides work in preventing Clubroot disease in Mustard. The data were statistically analyzed using the open-source program 'R' [5].

Procedures of Artificial Inoculation of *Plasmodiophora Brassicae* under Lab Conditions

The entire study was carried out in the laboratory and growth chamber of the Plant Pathology Division at the Regional Agricultural Research Station (RARS), which is part of the Bangladesh Agricultural Research Institute (BARI), located in Jamalpur-2000 **(Figure 2),** Bangladesh. The research used samples of clubroot disease collected from Brassica plants. The soil used in the lab tests was taken from the top 20 centimeters of soil from a farm field where mustard was grown before. The soil was crushed and passed through a sieve with 3 mesh openings, then put into pots that measured 12 centimeters in length, 12 centimeters in width, and 10 centimeters in height. The seeds of BARI Sarisha-14 were dipped in 70% ethanol for a short time and then rinsed three times with sterilized water, each rinse lasting one minute. Seeds were planted straight into each pot, and the young plants were spaced out so that only two plants were left in each pot after two weeks from when the seeds were planted. The infected clubbed roots were first taken from the infected plants. Fresh, firm and mature clubbed roots were collected and stored in a deep freezer. Later, the roots were washed well, then frozen and kept in a freezer at -20 degrees Celsius. Resting spores of *P. brassicae* were separated from clubroot galls. The galls were washed with tap water to remove dirt and other unwanted materials, and then they were rinsed three times using sterilized water. The inoculation solution was prepared using the method outlined by Peng *et al.* (2019) [6], with a slight modification. The galls were broken into smaller pieces and placed into a tissue crusher together with sterilized water, using a volume ratio of 1-part galls to 1 part water. After mixing everything together, the clear liquid on top was collected by pouring it through

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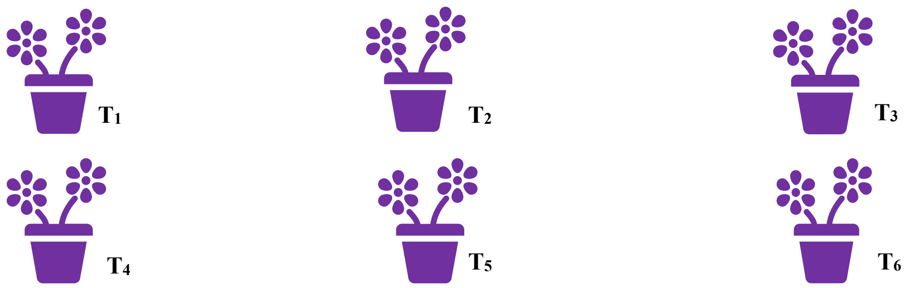
eight layers of gauze. Then, it was spun in a centrifuge at 100 times gravity for 5 minutes. The count of resting spores in the supernatant was done four times with a hemocytometer, and each time, five squares were examined. Finally, the resting spores were mixed with sterile water to reach a final concentration of 1×10^8 per milliliter, making it the inoculation solution. The lab experiment was carried out at the Plant Pathology Laboratory, Regional Agricultural Research Station (RARS), Bangladesh Agricultural Research Institute (BARI), Jamalpur-2000, Bangladesh from February to April 2025. The temperature inside the growth chamber stayed the same at 24 degrees Celsius, and the humidity was kept at 90%. Eight (8) treatments viz., T_1= Acrobat MZ (Dimethomorph + Mancozeb) @ 2 g/L, T_2= Ridomil Gold MZ 68 WG (Mancozeb + Metalaxyl) @ 2 g/L, T_3= Dolochune @ 5 t/ha, T_4=Luna sensation 50 SC (Fluopyrum + Trifloxystrobin) @ 2 ml/L, T_5 = Amistar Top 325 SC(Azoxystrobin + Difenoconazole) @ 2 ml/L, T_6= Nativo 75 WG (Tebuconazole + Trifloxystrobin) @ 2 g/L, T_7= Blitox 50 WP (Copper oxychloride) @ 2 g/L and T_8= Control were applied after inoculation for all set up for the study. Each treatment was done with six copies, and each copy had ten to twelve plants. Inoculation of resting spores and fungicide application were done according to the methods described by Peng *et al.* (2019) [6]. Plants were inoculated with 2 mL resting spores (1×108 mL−1) three weeks after seeding by pouring the solution around each plant's base. Control treatment received 2 mL of sterilized water. The treatment T_3 was applied during pot preparation using Dolochune @ 5 t/ha. After 24 hours since the initial infection (24 hpi), the plants were given their specific fungicide treatments. These were made by dissolving the mentioned fungicides of tap water, following the recommended amounts. Each plant's base was sprayed with ten milliliters of either tap water or fungicide solution. The treatment was applied three times, at 1 day, 8 days, and 18 days after the infection was introduced, following the recommended schedule for application times and intervals. Plants were collected during the six weeks after they were infected. Paper towels were used to dry the roots after they had been cleaned with PBS solution. Then, the level of clubroot disease was assessed using standard disease rating guidelines. Grade 0 means there are no signs of disease. Grade 1 shows small clubs and a few galls, mostly on the fine roots. Grade 2 has more galls, mainly on the main root and fewer on the fine roots. Grade 3 involves serious galls on both the main root and the fine roots. Grade 4 indicates large galls or decay Agarwal A, et al (2011) [7]. The following formula was used for calculating the Disease Index (DI) for each treatment at each evaluation date Han X, et al (2022) [8]: DI (%) is equal to Σ (nx0 + nx1 + nx2 + nx3 + nx4) x100 / (Nx4). where N is the total number of plants, n is the number of plants in each grade, and the classes that indicate the severity of the symptoms are 0, 1, 2, 3, and 4.



T_1= Acrobat MZ (Dimethomorph + Mancozeb) @ 2 g/L, T_2= Ridomil Gold MZ 68 WG (Mancozeb + Metalaxyl) @ 2 g/L, T_3= Dolochune @ 5 t/ha, T_4=Luna sensation 50 SC (Fluopyrum + Trifloxystrobin) @ 2 ml/L, T_5 = Amistar Top 325 SC(Azoxystrobin + Difenoconazole) @ 2 ml/L, T_6= Nativo 75 WG (Tebuconazole + Trifloxystrobin) @ 2 g/L, T_7= Blitox 50 WP (Copper oxychloride) @ 2 g/L and T_8= Control.

Pot size: length × width× height was 12× 12 × 10 cm

Inoculation: 2 mL 1×10^8 mL^{-1} resting spore suspension of *P. brassicae* per plant was applied in all treatments (T_1–T_8).

Treatment of fungicides: 10 mL tap water per plant was applied in T_8 treatment, T_3 treatment was applied according to dose of Dolochune and 10 mL solution of different fungicide per plant was applied according to other treatments. All the treatments were repeated 3 times at 1-, 8- and 18-days post inoculation (dpi).

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Sampling: Each treatment was done with six replicates, and ten plants were collected from each treatment after six (6) weeks inoculation.

Figure 2. Lab experimental design with detailed methods presented diagrammatically.

RESULTS AND DISCUSSION

Results of Field Experiment

Effects of Fungicides and Soil Amendment Products on Clubroot Disease of Mustard

BARI Sarisha-14 was evaluated to find out suitable fungicide/fungicides and the efficacy of different fungicides against clubroot disease and for yield and yield contributing characters. The performances of different fungicides along with control are stated in Table 1, 2 and 3. Significant differences were observed for total plants per plot, but percentage infected plants, disease severity and disease status of BARI Sarisha-14 showed no significant variations Table 1. The total number of plants per plot varied from 132 to 178. The highest total number of plants (178) was found in the control treatment (T_8) and the lowest in the T_5 treatment (132). The incidence of clubroot disease was observed among the treatments. Only 0.59% of infected plants were found in the control treatment (T_8) where disease severity is 1 and disease status was a few small clubs in the infected plants. Other treatments had no infection (no clubbing).

These findings indicate that fungicides application for controlling clubroot disease of BARI Sarisha-14 is not effective because very few percentage infections were found in control treatment under natural field conditions. Artificial inoculation of clubroot inoculum in the plants afterwards application of fungicides would be effective. Liao et al (2022) [9] reported that after three and six weeks of treatment, the three fungicides [Fluazinam (500 g/L, suspension concentrate), metalaxyl-mancozeb (53%, contain 5% metalaxyl and 48% mancozeb, wettable powder), and carbendazim (50% wettable powder)] somewhat reduced the incidence and severity of clubroot disease when compared to water treatment. Compared to the water control and the other two fungicide treatments, the fluazinam treatment showed the highest control efficacy (59.81%) and the lowest disease incidence and DI after six weeks of treatment. The control effect did not differ substantially between the metalaxyl-mancozeb (15.95%) and carbendazim (29.80%) treatments; however, the DI and disease incidence were significantly lower than those of the water treatment. Three weeks following inoculation, the amount of *P. brassicae* in rhizosphere soil (RS) was associated with the severity of clubroot disease. Six weeks after inoculation, *P. brassicae* Rose was significantly more prevalent in RS treated with water and metalaxyl-mancozeb than in those treated with carbendazim and fluazinam. Bulk soil did not significantly change between the two sampling times. Peng *et al.* (2014) [10] also demonstrated that the potential of a few fungicides and bio fungicides, either alone or in combination with crop rotation or cultivar resistance, was evaluated for integrated clubroot disease control. *Plasmodiophora brassicae* was susceptible to the synthetic fungicides pentachloronitrobenzene, fluazinam, and cyazofamid. Under controlled environmental settings, the bio fungicides Serenade® and Prestop® both developed host resistance and inhibited the illness on canola through antibiosis. Formulations for granular and seed treatment were created to make it easier to apply bio fungicide in field tests. Both synthetic and bio fungicides were insufficiently effective in the seed furrow when *P. brassicae* resting spore levels were high in the soil. On Chinese cabbage, they occasionally lessened the harshness of clubroot. According to the findings of our research, the use of synthetic fungicides or bio fungicides for the treatment of clubroot disease on canola is not currently commercially practical until better formulations are created and greater efficiency is attained.

The effects of different fungicides on days to flowering, days to maturity and plant characteristics of BARI Sarisha-14 are presented in Table 2. Days to flowering and days to maturity do not differ significantly (Table 2). Days to flowering of BARI Sarisha-14 varied from 33 to 34 days in different treatments. Depending on the treatments, BARI Sarisha-14 took 74–76 days to get to maturity. There

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were notable differences in plant height, primary branches per plant, and secondary branches per plant (Table 2). The range of plant heights was 92.60 to 99.20 cm. There were between six and eight primary branches per plant. There were between 0.00 and 2.00 secondary branches per plant.

The effects of different fungicides on yield and yield contributing characteristics of BARI Sarisha-14 are stated in Table 3. Significant differences were observed in the number of siliquae per plant, 1000-seed weight, and seed yield per plot and hectare except siliqua length. The number of siliquae per plant varied from 70.40 to 100.33. The T_4 treatment had the lowest (70.40) number of siliquae per plant. On the other hand, The T_7 treatment got the highest number of siliquae per plant followed by the T_2 treatment (93.70 cm). The length of siliqua ranged from 4.04 to 5.23 cm. The T_2 treatment attained the highest 1000-seed weight (3.97 g) and the T_2 treatment gained the lowest 1000-seed weight (3.70 g). Seed yield per plant varied from 0.54 to 0.64 kg among the treatments. The highest seed yield per plant (0.64 kg) was noted in the T_7 treatment followed by the T_2 and T_8 treatments (0.62 kg). Seed yield per hectare ranged from 1494.44 kg to 1768.52 kg. The highest seed yield per hectare (1768.52 kg) is noted in the T_7 treatment followed by the T_8 treatment (1720.37 kg), T_1 and T_2 treatments (1703.70 kg). These findings indicate that although seed yield performance exhibits significant variations among the treatments but only one treatment showed better performances compared to control. On the other hand, two treatments showed at par seed yield performance with the control and four treatments showed inferior performance to control that might be due to the uneven population and land heterogeneity.

Table 1. Effect of different fungicides on total number of plants per plot, percent infected plants, disease severity and disease status of BARI Sarisha-14.

Treatments	Total no. of plants/plot	Infected plants (%)	Disease severity	Disease status
T_1	170 ab	0	0	No clubbing
T_2	158 c	0	0	No clubbing
T_3	148 d	0	0	No clubbing
T_4	158 c	0	0	No clubbing
T_5	132 e	0	0	No clubbing
T_6	169 b	0	0	No clubbing
T_7	172 ab	0	0	No clubbing
T_8	178 a	0.59	1	A few small clubs
Level of significance	**	-	-	-
CV (%)	3.01	-	-	-

** Significant at 5% level (p≤0.05), ** Significant at 1% level (p≤0.01), NS: Not significant (p≥0.05). At the 5% level of significance, there is no statistical difference between the same letter in the same column. Square root transformed values are indicated by values enclosed in parenthesis. Disease severity (0-3 scale): 0 = No clubbing, 1 = a few small clubs, 2 = moderate clubbing, 3 = severe clubbing. T_1= Acrobat MZ (Dimethomorph + Mancozeb) @ 2 g/L, T_2= Ridomil Gold MZ 68 WG (Mancozeb + Metalaxyl) @ 2 g/L, T_3= Dolochune @ 5 t/ha, T_4=Luna sensation 50 SC (Fluopyrum + Trifloxystrobin) @ 2 ml/L, T_5 = Amistar Top 325 SC(Azoxystrobin + Difenoconazole) @ 2 ml/L, T_6= Nativo 75 WG (Tebuconazole + Trifloxystrobin) @ 2 g/L, T_7= Blitox 50 WP (Copper Oxychloride) @ 2 g/L and T_8= Control.*

Table 2. Effect of different fungicides on days to flowering, days to maturity and plant characteristics of BARI Sarisha-14.

Treatments	Days to flowering	Days to maturity	Plant height (cm)	Primary branches/plant	Secondary branches/plant
T_1	33	74	92.93 bc	6 e	0.60 c
T_2	34	75	95.53 abc	8 a	0.55 c
T_3	33	76	92.60 c	7 c	0.00 e
T_4	34	76	93.07 bc	8 a	2.00 a
T_5	33	75	95.20 abc	7 c	0.20 d
T_6	33	75	86.20 d	6 d	0.00 e
T_7	33	75	99.20 a	7 b	1.00 b
T_8	33	75	97.33 ab	7 a	0.20 d

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Treatments	Days to flowering	Days to maturity	Plant height (cm)	Primary branches/plant	Secondary branches/plant
Level of significance	NS	NS	**	**	**
CV (%)	3.4	2.09	2.55	2.51	15.20

** Significant at 5% level (p≤0.05), ** Significant at 1% level (p≤0.01), NS: Not significant (p≥0.05). The same letter in the same column does not differ statistically at the 5% level of significance. T_1= Acrobat MZ (Dimethomorph + Mancozeb) @ 2 g/L, T_2= Ridomil Gold MZ 68 WG (Mancozeb + Metalaxyl) @ 2 g/L, T_3= Dolochune @ 5 t/ha, T_4=Luna sensation 50 SC (Fluopyrum + Trifloxystrobin) @ 2 ml/L, T_5 = Amistar Top 325 SC(Azoxystrobin + Difenoconazole) @ 2 ml/L, T_6= Nativo 75 WG (Tebuconazole + Trifloxystrobin) @ 2 g/L, T_7= Blitox 50 WP (Copper oxychloride) @ 2 g/L and T_8= Control.*

Table 3. Effect of different fungicides on yield and yield-contributing characters of BARI Sarisha-14.

Treatments	No. of siliqua/plant	Length of siliqua (cm)	1000-seed weight (g)	Seed yield/ plot (kg)	Seed yield (kg/ha)
T_1	73.20 c	5.23	3.87 ab	0.61 ab	1703.70 ab
T_2	93.70 ab	4.32	3.97 a	0.62 ab	1703.70 ab
T_3	72.80 c	4.43	3.70 b	0.56 c	1566.67 bc
T_4	70.40 c	4.04	3.90 ab	0.58 bc	1611.11 abc
T_5	90.60 b	4.47	3.93 ab	0.54 c	1494.44 c
T_6	74.07 c	4.48	3.90 ab	0.58 bc	1608.33 abc
T_7	100.33 a	4.55	3.73 ab	0.64 a	1768.52 a
T_8	92.73 b	4.55	3.77 ab	0.62 ab	1720.37 ab
Level of significance	**	NS	*	**	**
CV (%)	4.61	5.04	2.28	3.97	4.04

**Significant at 5% level (p≤0.05), ** Significant at 1% level (p≤0.01), NS: Not significant (p≥0.05). The same letter in the same column does not differ statistically at the 5% level of significance. T_1= Acrobat MZ (Dimethomorph + Mancozeb) @ 2 g/L, T_2= Ridomil Gold MZ 68 WG (Mancozeb + Metalaxyl) @ 2 g/L, T_3= Dolochune @ 5 t/ha, T_4=Luna sensation 50 SC (Fluopyrum + Trifloxystrobin) @ 2 ml/L, T_5 = Amistar Top 325 SC (Azoxystrobin + Difenoconazole) @ 2 ml/L, T_6= Nativo 75 WG (Tebuconazole + Trifloxystrobin) @ 2 g/L, T_7= Blitox 50 WP (Copper oxychloride) @ 2 g/L and T_8= Control.*

RESULTS OF LAB EXPERIMENTS

Effects of Fungicides and Soil Amendment Products on Clubroot Disease of Mustard

Compared to the control treatment, the six fungicides and one soil amendment product reduced the occurrence and severity of clubroot disease to some degree after six weeks of treatment Figure 3. The results show disease incidence, disease index, and control effectiveness from different treatments for clubroot disease in mustard are shown in the figure. 4. The disease occurred in 75 to 100% of the occurrences. The control treatment had the highest number of diseases, which was 100%, while the T_4 treatment was the lowest at 75%. The T_5 treatment was 80%, T_6 treatment had 82%, and T_1 treatment had 85%. The other three treatments have a disease incidence rate higher than 90%. The disease was very severe, ranging from 64 to 100% of cases. The worst disease severity, which is 100%, is seen in control treatment. The least severe disease is found in the T_4 treatment with 64%, then comes T_5 at 69%, T_6 at 71%, and T_1 at 74%. The other three treatments have more than 80% disease severity. The effectiveness of the disease control ranged from 12 to 36% across the different treatments. After 6 weeks of treatment, the T_4 treatment had the highest control effectiveness at 36%, then T_5 at 31%, T_6 at 29%, and T_1 at 26%. The lowest disease control effectiveness was seen in the T_7 treatment with 12%, followed by T_2 at 15% and T_3 at 18%. In this study, it was discovered that Luna sensation 50 SC (Fluopyrum + Trifloxystrobin) had a disease control effectiveness of 36%, which was better than the other treatments used. It works by targeting fungi throughout the system, stopping them from making energy. This happens because of the two main ingredients, fluopyram and trifloxystrobin. This lets it manage various diseases by stopping the spores from starting to grow, slowing down the fungus' spread, and preventing it from creating new spores. Another fungicide called Amistar Top 325 SC works against a wide range of diseases. It helps prevent, treat, and control difficulties in plants.

It has two main ingredients: Azoxystrobin, which stops fungi from breathing, and Difenoconazole, which prevents fungi from growing by messing up how they make certain chemicals. This two-step

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process offers ongoing protection and helps increase both the amount and the quality of the crops. Nativo 75 WG is a fungicide that mixes tebuconazole and trifloxystrobin, and it's used to manage various types of fungal diseases. Its work depends on two ways it affects the fungus: tebuconazole stops the fungus from building its cell wall, and trifloxystrobin stops the fungus from breathing properly. It can be used as a spray on the leaves at certain amounts. The first use is usually suggested before the disease starts, and then more sprays are needed every 15 to 21 days as part of a plan to help plants resist the disease. Our results match what Suzuki et al. (1995) [11] found. He showed that fluazinam can stop the resting spores from germinating and prevent the root hair infection stage, which stops the formation of clubs.

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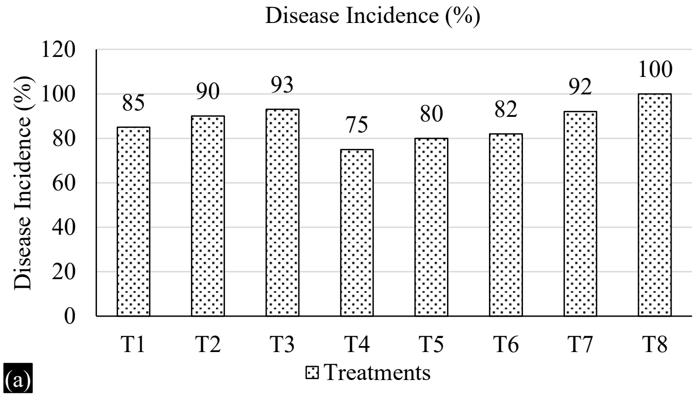
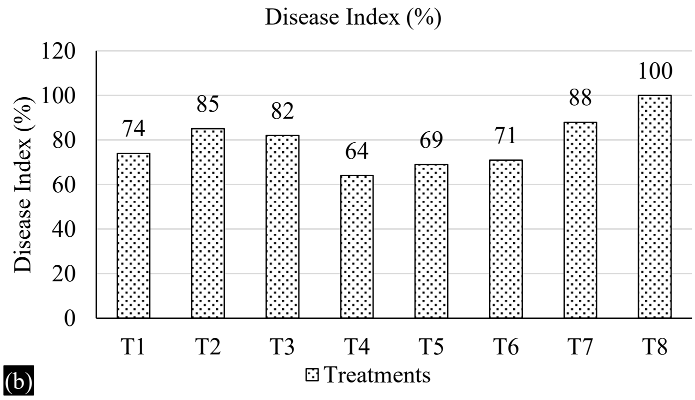
(f)

Figure 3. (a–h) Phenotype of Clubroot disease severity in different treatments of mustard. T_1= Acrobat MZ (Dimethomorph + Mancozeb) @ 2 g/L, T_2= Ridomil Gold MZ 68 WG (Mancozeb + Metalaxyl) @ 2 g/L, T_3= Dolochune @ 5 t/ha, T_4=Luna sensation 50 SC (Fluopyrum + Trifloxystrobin) @ 2 ml/L, T_5 = Amistar Top 325 SC (Azoxystrobin + Difenoconazole) @ 2 ml/L, T_6= Nativo 75 WG (Tebuconazole + Trifloxystrobin) @ 2 g/L, T_7= Blitox 50 WP (Copper oxychloride) @ 2 g/L and T_8= Control.





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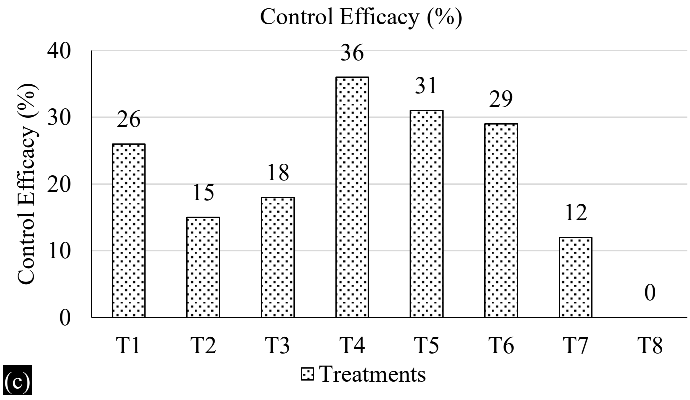

Figure 4. (a–c) Disease incidence, disease index and control efficacy in different treatments of clubroot disease of mustard. T_1= Acrobat MZ (Dimethomorph + Mancozeb) @ 2 g/L, T_2= Ridomil Gold MZ 68 WG (Mancozeb + Metalaxyl) @ 2 g/L, T_3= Dolochune @ 5 t/ha, T_4=Luna sensation 50 SC (Fluopyrum + Trifloxystrobin) @ 2 ml/L, T_5 = Amistar Top 325 SC (Azoxystrobin + Difenoconazole) @ 2 ml/L, T_6= Nativo 75 WG (Tebuconazole + Trifloxystrobin) @ 2 g/L, T_7= Blitox 50 WP (Copper oxychloride) @ 2 g/L and T_8= Control.

CONCLUSION

BARI Sarisha-14 was evaluated for yield and yield-contributing characteristics as well as to find out the appropriate fungicide/fungicides and the efficacy of fungicides against the clubroot disease of mustard both in the field and lab conditions. In case of field conditions, with a disease severity of 1 and a disease status of a few small clubs in the infected plants, only 0.59% of the infected plants were detected in the control treatment (T_8). There were no infections from other treatments. Because a relatively low percentage of infections were identified in the control treatment under natural field conditions, our findings suggest that the application of fungicides to suppress the clubroot disease of BARI Sarisha-14 is ineffective. It would be beneficial to artificially inoculate the plants with clubroot inoculum before applying fungicides. However, later, lab experiments were conducted through artificial inoculation. Among the T_7, T_8, T_1, and T_2 treatments, the maximum seed yield output per hectare (1768.52 kg, 1720.37 kg, and 1703.70 kg) was observed, respectively. Although there are notable differences in seed yield performance amongst the treatments, these results suggest that just one treatment outperformed the control. However, two treatments produced seed yields that were comparable to the control, and four treatments performed worse than the control, which may have been brought on by the unequal population and land heterogeneity. In case of lab experiment, after six weeks of inoculation, six fungicides and one soil amendment significantly reduced clubroot disease incidence compared to the control treatment, which had a 100% disease occurrence. Disease severity varied, with T_4 showing 64% severity versus 100% in control. The effectiveness peaked at 36% for T_4 and dropped to 12% for T_7. Luna sensation 50 SC was the most effective, targeting fungi's energy production, while Amistar Top 325 SC and Nativo 75 WG also managed disease by disrupting fungal respiration and growth. Regular application of these treatments boosts crop resilience against diseases. In addition, it should also consider broader integrated disease management perspectives, including recent advances in nanotechnology or biocontrol.

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