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Effect of bio-resources against *alternaria* leaf spot (*Alternaria brassicae*) of broccoli (*Brassica oleracea* var. italica)

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Abstract
Broccoli is an economically important vegetable and India is the second largest producer in the world. *Alternaria* leaf spot is one of the most important disease in crucifers which causes serious yield and quality loss in production. Microalgal bio-stimulants are able to stimulate the growth and protect from many diseases of several crops under both optimal and stressful condition. It helps in generating multiple benefits, such as enhanced rooting, higher crop yields and quality and tolerance to drought and salt stress. The bioactive ingredients of neem have also shown to be efficacious in controlling several crop diseases. Keeping in view of the above mentioned facts, the following treatments like – Microalgae at different doses, Neem oil and SMC were utilized for controlling *Alternaria* leaf spot of broccoli. Neem oil @ 3% and Microalgae at different doses was applied three times at an interval of 15 days after appearance of the disease. Spent mushroom compost was applied 7 days before transplanting broccoli. Among all the treatments highest percent reduction of disease intensity in *Alternaria brassicae* was found in T6 @8% microalgae, 3% neem oil and SMC. T6 was recorded to significantly increase in higher plant height (cm), width of curd (cm) and weight of curd (g) at 26.60 cm, 28.06 cm and 247.46 g respectively.

Keywords: *Alternaria brassicae*, broccoli, microalgae, neem oil, SMC

Introduction

Broccoli (*Brassica oleracea* var. italica) belongs to the family Brassicaceae or Cruciferae and is native to the Mediterranean region. It is a highly nutritious crop containing high number of vitamins (A and C) and minerals (K, P, Ca and Fe). In India it is mostly cultivated in hilly areas of Himachal Pradesh, Uttar Pradesh, Jammu and Kashmir, Nilgiri Hills and Northern plains of India. India holds second positions in the list when it comes to the production of broccoli vegetables after China. Broccoli production according to the FAOSTAT, 2019 is 9.1 million tonnes in India. Broccoli like many other vegetables gets infected by a number of bacterial, fungal, viral diseases and nematodes. *Alternaria* leaf spot or blight caused by *Alternaria brassicae* has become a problematic disease due to the soil-borne survival of the fungus, local over-wintering of inoculum, cultivation of susceptible varieties and favourable environmental conditions. The typical symptoms appear as circular to irregular shaped spots with concentric rings (target board appearance). Individual spots coalesce into large necrotic areas and leaf drop can occur. Lesions can occur on petioles, stems, flower pedicels on broccoli plant (Ellis, 1968) [4].

Microalgae are photosynthetic microorganisms grown in marine, fresh or wastewaters with different nutrient sources and used in fuel, agriculture, food, pharmaceutical, lipids, proteins and phyto-hormones as reported by Kumar and Sahoo (2011) [13]; Godleweska *et al*., (2016) [8] and Uithirapandi *et al*. (2018) [25], their intracellular extracts can be considered for enhancing the plant yield (Ellaroussia *et al*., 2016) [3]; Garcia-Gonzalez and Sommerfeld, 2016) [6]. Microalgae are used in agriculture in different applications, such as amendment, foliar application, seed priming and fertigation (application of microalgae along with irrigation). Microalgal bio-stimulants are able to stimulate the growth and development of several crops under both optimal and stressful condition. It helps in generating multiple benefits, such as enhanced rooting, higher crop yields and quality and tolerance to drought and salt stress (Ronga *et al*. 2019) [21]. Microalgae have potential for enhancing soil fertility and promote plant growth, fruit quality and also increase the availability of macro and micro nutrients (Ellaroussia *et al*., 2016; Garcia-Gonzalez and Sommerfeld, 2016) [3, 6].

Moreover, microalgae is thought to be beneficial to plant growth because of its ability to produce a variety of valuable compounds such as polyunsaturated (Lin *et al*., 2017) [18]. The application of microalgae products in plants have been reported to have had different responses, such as robust growth, increased yield, improved nutrient uptake, and increased biotic and abiotic stress resistance (fungal infections, pest attacks, and frost), increased quality, and fruit with longer shelf life. Neem oil is an effective and preventive fungicide used in the control of various diseases like leaf spot, *Alternaria* blight, downy and powdery mildews, rust, scab and flower, twig and tip blight, anthracnose and Botrytis blight (Kuepper, 2003) [12]. Spent mushroom compost (SMC) improves the soil structure, aeration, slow release nutrient which support root development leading to higher growth and yield of broccoli (Abou *et al*., 2006) [1]. SMS is characterized by a high content of organic matter, good availability of macro- and micro-nutrients, neutral pH, a favorable narrow C/N ratio, and a low content of heavy metals (Kwiatkowski *et al*., 2018 and Elsakhawy *et al*., 2020) [17, 5]. The use of spent mushroom compost in agriculture for the fertilization of arable land and permanent grassland has been proposed in fruit and vegetable growing, as well as for the establishment and maintenance of green spaces (Maher *et al*., 2000) [20]. Song *et al*., (2007) [23] and Jordan *et al*., (2007) [11] have reported that spent mushroom compost has a positive effect on the productivity of grasses, cereals, some vegetable plants, and flowers.

Materials and Methods

The present study was conducted at the Experimental field of Department of Plant Pathology in Central Research Farm, Sam Higginbottom University of Agriculture, Technology and Sciences, during the Rabi season of 2021-2022. Field experiment was laid-out in Randomized block design with three replications in a plot size of 2×2 m2 at a spacing of 45 cm × 40 cm with all the recommended package of practices.

Spent Mushroom Compost was applied as soil application during soil preparation at a rate of 1 kg per plot. Microalgae were applied at different doses @1%, 3%, 5%, 6%, 7% and 8% v/v, i.e., 10 g, 30 g, 50 g, 60 g, 70 g and 80 g per litre of water, and applied in each plot through irrigation at 30, 45 and 60 DAT followed by foliar spray of Neem oil @ 3%. Seven treatments along with untreated control check were tested against *Alternaria brassicae* under field condition. Observations were recorded at 30, 45 and 60 DAT on 5 randomly selected plants in each plot to see the disease intensity of *Alternaria brassicae* and plant growth parameters – Plant height, width of curd and weight of curd. Treatments were imposed after the first appearance of the disease symptoms i.e., 15-20 DAT.

Per cent disease intensity was recorded at 30, 45 and 60 days after incidence of *Alternaria* leaf spot. Percentage of Disease intensity will be calculated in accordance with following formula.

$$\text{Disease intensity} = \frac{\text{Sum of all disease ratings}}{\text{Total no. of ratings} \times \text{Maximum disease grade}} \times 100$$

Table 1: Description of disease rating scale for *Alternaria* leaf spot of broccoli (Hansen and Earle 1997; Doullah *et al*., 2006) [9, 2]: Categorization of disease intensity:

Grade	Description of Symptoms
0	No spots and no Chlorosis on the investigated plant organ
1	Disease symptoms visible on up to 10% area of the investigated plant organ
2	Disease symptoms visible on 11 to 23% area of the investigated plant organ
3	Disease symptoms visible on 26 to 50% area of the investigated plant organ
4	Disease symptoms visible on 51 to 73% area of the investigated plant organ
5	Disease symptoms visible on more than 76% area of the investigated plant organ

a) Microalgae 8%, Neem oil 3% & SMC

b) Microalgae 7%, Neem oil 3% & SMC

c) Broccoli curds from untreated control

Results and Discussion

The data regarding disease intensity of *Alternaria brassicae* of broccoli, mean of (30, 45 and 60 DAT) presented in table 2 showed that all the treatments were significantly superior over control. The results indicated that T6 (Microalgae 8% + Neem oil 3% + SMC) significantly reduced the percentage of *alternaria* leaf spot as compared to T5, T4, T3, T2, T1 and T0. The disease intensity of *Alternaria brassicae* was recorded to be lowest in T6 – (2.81%) followed by T5 – (3.69%), T4 – (4.69%), T3 – (5.59%), T2 – (6.37%), T1 – (7.45%) as compared to untreated check T0 – Control (10.95%)

Table 2: Effect of Microalgae, Neem oil and SMC on disease intensity (%) of *Alternaria brassicae* of broccoli.

Treatments		Disease Intensity (%)			
		30 DAT	45 DAT	60 DAT	MEAN
T0	Control (Untreated check)	5.26	9.73	17.86	10.95
T1	Microalgae 1% + Neem oil 3% + SMC	4.61	7.41	10.34	7.45
T2	Microalgae 3% + Neem oil 3% + SMC	4.10	6.62	8.38	6.37
T3	Microalgae 5% + Neem oil 3% + SMC	3.64	6.06	7.07	5.59
T4	Microalgae 6% + Neem oil 3% + SMC	3.19	4.99	5.89	4.69
T5	Microalgae 7% + Neem oil 3% +SMC	1.73	4.19	5.14	3.69
T6	Microalgae 8% + Neem oil 3% + SMC	1.47	2.91	4.03	2.81
	C.D. (5%)	0.73	0.54	0.83	3.27
	S.Ed ±	0.34	0.25	0.38	1.50

The data presented in table 3 reveals the response of Microalgae, Neem oil and SMC on plant height of broccoli at 30, 45 and 60 DAT under field condition. The results indicated that among all the treatments T6 (Microalgae 8% + Neem oil 3% + SMC) was found to significantly increase the height of plant from all the treatments. The plant height was recorded to be the highest in T6 – (26.60 cm) followed by T5 – (25.71 cm), T4 – (24.71 cm), T3 – (23.68 cm), T2 – (22.57 cm), T1– (21.28 cm) as compared to untreated check T0 – (18.06 cm).

Table 3: Effect of Microalgae, Neem oil and SMC on Plant Height (cm) of broccoli.

Treatments		Plant Height (cm)			
		30 DAT	45 DAT	60 DAT	Mean
T0	Control (Untreated check)	6.13	11.33	36.73	18.06
T1	Microalgae 1% + Neem oil 3% + SMC	7.40	14.20	42.26	21.28
T2	Microalgae 3% + Neem oil 3% + SMC	7.26	15.86	44.60	22.57
T3	Microalgae 5% + Neem oil 3% + SMC	7.9	16.33	46.80	23.68
T4	Microalgae 6% + Neem oil 3% + SMC	8.40	17.1	48.60	24.71
T5	Microalgae 7% + Neem oil 3% + SMC	8.73	18.13	50.26	25.71
T6	Microalgae 8% + Neem oil 3% + SMC	8.86	18.86	52.00	26.60
	C.D. (5%)	0.74	0.93	0.91	3.84
	S.Ed ±	0.34	0.41	0.39	1.76

The data presented in table 4 reveals the response of Microalgae, Neem oil and SMC on width of curd of broccoli at 30, 45 and 60 DAT under field condition. Among the different concentrations maximum width of curd was found in T6 – (11.53 cm) followed by T5 – (10.06cm), T4 – (9.26cm), T3 – (8.46cm), T2 – (7.46cm), T1 – (6.26cm) and T0 – Control (4.20cm).

Table 4: Effect of Microalgae, Neem oil and SMC on Width of Curd (cm)

Treatments		Width of Curd (cm)			
		60 DAT	75 DAT	90 DAT	Mean
T0	Control (Untreated check)	4.20	17.00	26.33	15.84
T1	Microalgae 1% + Neem oil 3% + SMC	6.26	25.93	31.26	21.15
T2	Microalgae 3% + Neem oil 3% + SMC	7.46	26.73	33.46	22.55
T3	Microalgae 5% + Neem oil 3% + SMC	8.46	27.93	35.00	23.80
T4	Microalgae 6% + Neem oil 3% + SMC	9.26	28.86	37.06	25.06
T5	Microalgae 7% + Neem oil 3% + SMC	10.06	29.46	38.86	26.13
T6	Microalgae 8% + Neem oil 3% + SMC	11.53	32.06	40.00	28.06
	C.D. (3%)	0.64	0.97	0.51	2.76
	S.Ed±	0.29	0.45	0.23	1.26

The data presented in table 5 reveals the response of Microalgae, Neem oil and SMC on weight of curd of broccoli after harvest under field condition. Among the different concentrations maximum weight of curd was found in T6 – (247.46 g) followed by T5– (225.10 g), T4 – (212.23 g), T3 – (188.13 g), T2 – (177.30 g), T1 – (157.60 g) and T0 – Control (122.13 g).

Table 5: Effect of Microalgae, Neem oil and SMC on Weight of Curd (g)

Treatments		Weight of Curd (g)			
		R1	R2	R3	Mean
T0	Control (Untreated check)	121	119.4	126	122.13
T1	Microalgae 1% + Neem oil 3% + SMC	157.8	155	160	157.60
T2	Microalgae 3% + Neem oil 3% + SMC	179.8	183.6	168.6	177.30
T3	Microalgae 5% + Neem oil 3% + SMC	182	185.2	197.2	188.13
T4	Microalgae 6% + Neem oil 3% + SMC	218.4	207	211.3	212.23
T5	Microalgae 7% + Neem oil 3% + SMC	220	229.1	226.2	225.10
T6	Microalgae 8% + Neem oil 3% + SMC	251	233.4	268	247.46
	C.D. (3%)				12.93
	S.Ed ±				5.93

The above results are in agreement with the findings of Kumar *et al*., (2018) [14] where he studied the efficacy of micro algae as biofertilizer of onion plants. Microalgal dry biomass of C. vulgaris and S. platensis was mixed with cow dung and applied to the soil separately. The growth parameters, yield attributes, bio-chemical composition, anti-nutritional composition and minerals were found to be higher in platens is and cow dung mixed biofertilizer treatment compared to the other treatments. This finding is supported by Supraja *et al*., (2020) [24] where he applied a mixed algal extract on tomato as seed treatment and foliar spray, which is found to have positive influence over the seed germination and plant growth rate, the total chlorophyll content was found to increase as well. Similarly, Kusvuran *et al*., (2019, 2021) [15, 16] investigated the effectiveness of microalgae (Chlorella vulgaris Beijerinck) through foliar application on drought dressed and salt stressed broccoli plants. Microalgae could be recommended as a sustainable strategy to improve the defense system of drought-stressed broccoli plants. Neem extracts were found highly effective against *Alternaria* brassisicola causing *alternaria* leaf spot of cabbage at 15% and 25% concentration and mycelial inhibition was recorded 68% at 25% concentration through poison food technique. Gupta *et al*., (2018) [7]. Similarly, Sasode *et al*., (2012) [22] also evaluated the effectiveness of some botanicals against *Alternaria brassicae* under *in vitro* condition where neemoil @3% was found to be significantly superior over Tulsi, Lantana, Datura and Pudina. Lopes *et al*., (2015) [19] also concluded that there is great potential of spent A. subrufescens compost (SMC) for use in organic tomato production because of the better quality of harvested fruit and Islam *et al*., (2014) [10] conducted an experiment to evaluate the growth and yield performance of broccoli influenced by organic manures such as Cowdung, SMC and mulching. He concluded that SMC in combination with water hyacinth mulching was superior over other treatments and best suited for broccoli cultivation.

Conclusion

From the above study it is concluded that T6 - Microalgae (8%) in combination with Neem oil (3%) and SMC is superior over all the treatments for the control of disease intensity and all the growth parameters. It also showed to produce the highest yield. *Alternaria brassicae* is a destructive fungus for broccoli but with the utilization of bio-resources and environmental friendly fertilizer it becomes easier to control this cosmopolitan fungus with much lower costs and is found as a better alternative to fungicides due to their fewer negative impacts on the environment and easy availability. However, the results are of one cropping season of October, 2021 – February, 2022 under the Prayagraj agro-climatic conditions. As such to validate the findings, more such experiments should be taken up in the future.

References

1. Abou El-Magd, Hoda MM, Mohamed A, Fawzy ZF. Effect of organic manure with or without chemical fertilizers on growth, yield and quality of some varieties of broccoli plants. Journal of Applied Science Research. 2006;2(10):791-798.
2. Doullah M, Meah M, Okazaki K. Development of an effective screening method for partial resistance to *Alternaria* brassicicola (dark leaf spot) in Brassica rapa. European Journal of Plant Pathology. 2006;116:33–43.
3. Elarroussia H, Elmernissia N, Benhimaa R, Kadmiria IME, Bendaou N, *et al*. Microalgae polysaccharides a

promising plant growth biostimulant. Journal of Algal Biomass Utilization. 2016;7(4):55-63.
4. Ellis MB. Dematiaceous hyphomycetes. Common wealth Mycological Institute; c1971. p. 464- 497.
5. Elsakhawy TA, Abd El-Rahem WT. Evaluation of Spent Mushroom Substrate Extract as a Biofertilizer for Growth Improvement of Rice (*Oryza sativa* L.). Egypt Journal of Soil Science. 2020;60:31–42.
6. Garcia-Gonzalez J, Sommerfeld M. Biofertilizer and biostimulant properties of the microalga Acutodes musdimorphus. Journal of Applied Phyvology. 2016;28(2):1051-1061.
7. Gupta P, Chandra R, Dubey P, Bio-efficacy of plants extract against *Alternaria* brassicicola causing *Alternaria* leaf spot of cabbage. Journal of Pharmacology and Phytochemistry. 2019;8(1):2332-2334.
8. Godlweska K, Michalak I, Tuhy L, Chojnacka K. Plant Growth Biostimulants Based on Different Methods of Seaweed Extraction with Water. Frontiers in the Expansion of Bioproducts; c2016. p. 11.
9. Hansen LN, Earle ED. Somatic hybrids between *Brassica oleracea* L. and *Sinapis Alba* L. with resistance to *Alternaria brassicae* (Berk.) Sacc. Theoretical and Applied Genetics. 1997;94(8):1078-1085.
10. Islam MD, Majumdar M, Sultana F, Islam T, S Md, *et al*. Growth and yield potential of broccoli in response to organic manures and mulching. International Journal of Business, Social and Scientific Research. 2014;2(1):68-73.
11. Jordan SN, Mullen GJ. Spend mushroom legislation in Ireland. Environmental Science Association of Ireland; c2007. p. 37–41.
12. Kuepper G. Downy mildew in cucurbits. Appropriare Technology Transfer for Rural Areas (ATTAR), USA, 2003. p. 1-6.
13. Kumar G, Sahoo D. Effect of seaweed liquid extract on growth and yield of Triticum aestivum var. Pusa Gold. Journal of Applied Phycology. 2011;23:251-255.
14. Dineshkumar R, Subramanian J, Arumugam A, Ahamed A Rasheeq, Sampathkumar P. Exploring the microalgae bio-fertilizer effect on onion cultivation by field experiment. Waste Biomass Valoriz. 2018;11:77-87.
15. Kusvuran S. Microalga (*Chlorella vulgaris* Beijerinck) alleviates salt stress of Broccoli plants by improving secondary metabolites and antioxidative defense system. Sciences Tome. 2019;72(9):1205-1212.
16. Kusvuran S. Microalgae (*Chlorella vulgaris* Beijerinck) alleviates drought stress of broccoli plants by improving nutrient uptake, secondary metabolites, and antioxidative defense system. Horticultural Plant Journal. 2021;7(3): 221-231.
17. Kwiatkowski CA, Harasim E, Yakimovich A, Kołodziej B, Tomczynska-Mleko M. Evaluation of spent mushroom substrate, mineral NPK fertilization and manure fertilization on chamomile (*Chamomila recutita* L. Rausch) yield and raw material quality. Acta Scientiarum Polonorum Hortorum Cultus. 2018;17:3–16.
18. Lin B, Ahmed F, Du H, Li Z, Yan Y, *et al*. Plant growth regulators promote lipid and carotenoid accumulation in Chlorella vulgaris. Journal of Applied Phycology. 2017;30:1549-1561.
19. Lopes Raul Xavier, Zied Diego Cunha, Martos Emerson Tokuda, de Souza, Rovilson José, *et al*. Application of spent Agaricus subrufescens compost in integrated production of seedlings and plants of tomato. International Journal of Recycling of Organic Waste in Agriculture. 2015;4(3):211-218.
20. Maher MJ, Smyth S, Dodd VA, McCabe T, Magette WL, *et al*. Managing Spent Mushroom Compost; Teagasc: Dublin, Ireland; c2000. p. 111-121.
21. Ronga D, Biazzi E, Parati K, Garminati D, Garminati E. *et al*. Microalgal Biostimulants and Biofertilisers in Crop Productions. Agronomy. 2019;9(4):146-163.
22. Sasode RS, Prakash S, Gupta A, Pandya RK, Yadav A. *In vitro* study of some plant extracts against *Alternaria brassicae* and *Alternaria brassicicola*. Journal of Phytology. 2012;4(1):44-46.
23. Song L, Siu-Wai C. Dual roles of spent mushroom substrate on soil improvement and enhanced drought tolerance of wheat Triticum aestivum. In Proceedings of the International Conferences, 3rd QLIF Congress, Crop Production, Soil Management, Stuttgart, Germany, 20–23 March 2007.
24. Supraja KV, Behera B, Balasubramanian P. Efficacy of microalgal extracts as biostimulants through seed treatment and foliar spray for tomato cultivation. Industrial Crops and Products. 2020;151(1):112-453.
25. Uithirapandi V, Suriya S, Boomibalagan P, Eswarna S, Ramya SS, *et al.*, Bio-fertilizer potential of seaweed liquid extracts of marine macroalgae on growth and bio-chemical parameters of Ocimum sanctum. J. Pharmacogn Phytocam. 2018;7(3):3528-3532.