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Multilocational validation of integrated management practices for *Sclerotinia* rot of Indian mustard (*Brassica juncea*)

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ABSTRACT

Field studies were conducted from 2008 to 2011 on integrated management of *Sclerotinia* rot of Indian mustard (*Brassica juncea* L. Czerns & Coss.) involving *Trichoderma* mixture (*T. viride* + *T. hamatum*) formulation (2×10^9 cfu/g) through seed treatment (10 g/kg), soil application (2.5 kg/ha) pre-incubated in farmyard manure and foliar sprays (0.2%) at 50 and 70 days after sowing along with improved cultural practices at Indian Agricultural Research Institute, New Delhi, which reduced the disease incidence up to a minimum of 11.1% and increased the seed yield up to a maximum of 2.72 tonnes/ha as compared to control (disease incidence 26.9% and seed yield 1.77 tonnes/ha). Validation of management practices were also undertaken during 2008 to 2011 in farmer's participatory mode under three different agro climates, Zone 1b and 3b in Rajasthan and south-west zone in Haryana. The validation studies revealed that integrated management strategies based on *Trichoderma* resulted in minimum *Sclerotinia* rot incidence (mean values of three regions 5.6%) and maximum seed yield (2.19 tonnes/ha) as compared to garlic clove extract (mean values 10.3% and seed yield 1.90 tonnes/ha) and Farmers' practices (mean values 19.2% and seed yield 1.66 tonnes/ha). Integrated management strategies based on *Trichoderma* spp. was found economically viable as indicated by incremental cost: benefit ratio ranging from 1:3.1 to 1:3.8 that was different with garlic clove extract (1:1.2 to 1:2.0), besides mustard seed was produced without any application of toxic fungicides.

Key words: Bioagent, B: C ratio, Garlic clove extract, Indian mustard, *Sclerotinia* rot, *Sclerotinia scleotiorum* and *Trichoderma* spp

Indian mustard (*Brassica juncea* L. Czernj & Coss.) is the major oilseed crop (>80% of rapeseed-mustard), widely grown in winter (*rabi*) season mostly in north and north-western India. More than 30 diseases are known to occur on *Brassica* crop in India. Among them, *Sclerotinia* rot caused by S*clerotinia sclerotiorum* (Lib.) de Bary has become a significant agricultural problem in Haryana, Himachal Pradesh, Punjab, Rajasthan, Uttarakhand and Uttar Pradesh. The maximum *Sclerotinia* rot incidence recorded in fields of mustard growers was 70% where seed yield was estimated to be reduced to 40%, which indicates the importance of the disease (NCIPM 2010). Earlier workers reported management of *Sclerotinia* rot of rapeseed and mustard mostly by foliar application of fungicides (Bradley *et al.* 2006, Ghasolia and Shivpuri 2008). The existing control measures are also the application of chemical fungicide which are often cost prohibitive, impractical and hazardous to environment and human health. Furthermore, the environmental damage and the economic cost of fungicidal application to manage this disease can not be ignored (del Rio *et al*. 2007). Keeping this in view, the need was felt for an alternative method to manage this disease in ecofriendly manner by using microbial bio-agent and plant extract. As *Trichoderma* spp and garlic (*Allium sativum* L.) clove extracts have been found effective against *S. sclerotiorum* of Indian mustard in laboratory (Yadav *et al.* 2011), therefore, they have been taken up in the integrated management of *Sclerotinia* rot. It is in this context, studies were undertaken to test the efficacy of *Trichoderma* spp and garlic clove extract for the suppression of *Sclerotinia sclerotiorum*. Hence, field studies were planned to test the efficacy of these bio-agent and garlic clove extract at Indian Agricultural Research Institute, New Delhi and also as multilocational validation studies on farmers' fields in Haryana and Rajasthan.

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MATERIALS AND METHODS

Field studies were conducted at IARI, New Delhi (28.08°N and 77.12°E, height above MSL 228.61 m) to evaluate the efficacy of bio-agent, *Trichoderma* mixture (*T. viride and T. hamatum*) and a garlic (*A. sativum* L.) clove extract during r*abi* season 2008–09 (under natural field conditions) and during 2009–10 and 2010–11 (under artificial disease conditions). Artificial inoculation of soil as well as stem was carried out as per Yadav *et al*. (2012). The field study consisting of seven different treatments (Table 1) on the basis of bio-agent *Trichoderma* (three treatments), garlic bulb extract (two treatments), fungicide carbendazim (two treatments) along with untreated control was laid out in sick field in randomized block design (plot size 5 m × 3 m) with three replications using variety Varuna. Sowing has been done between 27 and 30 October. The rows spacing was 30 cm and plant to plant spacing was 10 cm after thinning at 21 days after sowing (DAS). The recommended doses of fertilizers (N60, P40, K40, S40)/ha along with improved cultural practices were applied. To protect the crop from aphid oxy-demeton methyl (0.1%) was sprayed. The observations for per cent incidence of *Sclerotinia* rot on stem were recorded 20 days before the maturity of crop on eight rows in each plot leaving the border rows. The seed yield data was also recorded on eight rows in each plot except the border rows. Duncan's multiple range test (DMRT) was applied for analysis and comparing of results (Gomez and Gomez 1984).

Multilocational validation of two integrated management practices, viz. Trichoderma based and garlic clove extract based along with farmer's practices (Table 2) was conducted for three *rabi* crop seasons (2008–09, 2009–10 and 2010–11) at farmer's field in villages of Alwar and Sriganganagar districts of Rajasthan in collaboration with Agricultural Research Stations (ARSs), Navgaon (Alwar) and Sriganganagar (both the stations under Swami Keswanand Rajasthan Agricultural University, Bikaner) and village Sampkan (Gurgaon) in south-west zone of Haryana. Crop was sown in mid October in three large size plots measuring 5000 m^2 each. The recommended agronomic practices along with improved cultural practices were applied for raising the good crop. To protect the crop from aphid oxy-demeton methyl (0.1%) was sprayed. The observations for per cent incidence of *Sclerotinia* rot on stem were recorded 20 days before the maturity of crop. The seed yield data was recorded and thereafter, incremental cost: benefit ratio (ICBR) of two integrated management practices was calculated.

RESULTS AND DISCUSSION

Integrated management of Sclerotinia *rot of mustard*

Significantly less disease and higher yields were obtained in treated plots as compared to untreated plots at IARI, New Delhi (Table 1). *Sclerotinia* rot incidence was minimum (0.4%) in *Trichoderma* combination (seed + soil + foliar sprays) treatment, T_3 and differed significantly from garlic clove extract treatments, T_4 and T_5 (1.8 and 1.7%), carbendazim treatments-T_6 and T_7 (1.4 and 1.0%), and control,

Table 1 Effect of various treatments on *Sclerotinia* rot incidence and seed yield of mustard at Indian Agricultural Research Institute, New Delhi

Treatment	Incidence (%) of *Sclerotinia* rot				Seed yield (tonnes/ha)			
	2008–09	2009–10	2010–11	Mean	2008–09	2009–10	2010–11	Mean
T_1 = seed treatment with *T. viride* + *T. hamatum* @ 10 g/kg seed	1.1^{e}	45.8^{d}	4.4^{c}	17.1	2.36^{c}	2.25^{a}	2.43^{b}	2.35
T_2 = T_1+ soil application of *T. viride* + *T. hamatum* bioagent @ 2.5 kg mixed with 50 kg FYM/ha	1.2^{d}	41.0^{e}	3.0^{d}	15.1	2.53^{bc}	2.28^{a}	2.74^{a}	2.52
T_3 = T_2+ spray *T.viride* +*T. hamatum* @ 0.2% at 50 and 70 DAS	0.4^{f}	32.4^{f}	0.5^{e}	11.1	2.94^{a}	2.35^{a}	2.87^{a}	2.72
T_4 = Seed treatment with garlic clove extract @ 2% (W/V)	1.8^{b}	58.8^{b}	3.9^{cd}	21.5	2.25^{cd}	2.32^{a}	2.26^{c}	2.28
T_5 = T_4+ spray of garlic clove extract@ 2% (W/V) at 50 and 70 DAS	1.7^{c}	50.6^{c}	3.6^{cd}	18.6	2.58^{b}	2.33^{a}	2.62^{ab}	2.51
T_6= Seed treatment with carbendazim @ 2g/kg	1.4^{c}	50.2^{c}	5.9^{b}	19.2	1.89^{e}	2.21^{a}	2.19^{c}	2.10
T_7 = T_6+ spray with carbendazim @ 0.2% at 50 and 70 DAS	1.0^{e}	43.8^{e}	3.1^{cd}	16.0	2.15^{d}	2.23^{a}	2.80^{a}	2.39
T_8 = Control (untreated and only water spray)	2.7^{a}	70.1^{a}	7.8^{a}	26.9	1.65^{f}	1.63^{b}	2.04^{d}	1.77

DMRT analysis was followed

Figures with same letters not differ significantly

Table 2 Details of various management practices for *Sclerotinia* rot of Indian mustard

Management practice	Pre sowing	Sowing	Seedling and vegetative stage	Flowering and pod stage
I *Trichoderma*-based practices	1. Deep summer ploughing 2. Preparation of leveled and well drained field 3. Clean cultivation–removal of debris and residue of previous crop 4. Crop rotation with non-susceptible host 5. Balanced fertilizer N_{60}, P_{40}, K_{40}, S_{40}	1. Sowing from 16–31 October 2. Use of clean, certified seed devoid of sclerotia. 3. Seed treatment with *Trichoderma* (*T. viride* + *T. hamatum*) @ 10 g/kg seed 4. Soil application of *Trichoderma* (*T. viride* + *T. hamatum*) @ 2.5 kg/ha pre incubated in 50 kg FYM 5. Avoidance of narrow spacing/heavy seed rate	1. Maintenance of optimum plant population with wide spacing 2. Judicious use of irrigation depending upon crop stage, soil type rainfall etc	1. Foliar spray of *Trichoderma* (*T. viride* + *T. hamatum*) @ 0.2% at early bloom stage, i e 50 and 70 DAS 2. Rogueing out of infected plant before sclerotia formation 3. Collection and burning of infected stubbles
II Garlic clove extract based practices	1. Deep summer ploughing 2. Preparation of leveled and well drained field 3. Clean cultivation–removal of debris and residue of previous crop 4. Elimination of broad leaf weed (*Chenopodium album*) 5. Fertilizer: N_{100}, P_{40}.	1. Sowing from 16–31 October 2. Use of clean, certified seed devoid of sclerotia. 3. Seed treatment with aqueous garlic bulb extract @ 2% (w/v)	1. Irrigation and plant population as per farmer's practices	1. Foliar spray of aqueous garlic bulb extract @ 2% (w/v) at early bloom stage, i e 50 and 70 DAS 2. No rouging of infected plants 3. Collection and burning of diseased stubbles
Farmer's practices (Control)	1. No summer ploughing 2. No clean cultivation and removal of debris 3. Fertilizer as per farmer's practices	1. Sowing from 16–31 October 2. No seed and soil treatment	1. Irrigation and plant population as per farmer's practices	1. No foliar spray against *Sclerotinia* rot 2. No rouging of infected plants 3. No collection and burning of diseased stubbles

T_8 (2.7%) during 2008–09. Seed yields were also significantly higher in T_3 (2.94 tonnes/ha) as compared to control, T_8 (1.65 tonnes/ha). Dubey *et al*. (2012) reported the effectiveness of soil application and seed treatment of *Trichoderma* species for integrated management of wet root rot of chickpea. Garlic clove extract treatment T_4 was found significantly inferior to carbendazim treatments, T_6 and T_7 and to *Trichoderma* treatments, T_1, T_2 and T_3 for disease incidence. However, combination treatment (seed + sprays), T_5 of garlic was at par with seed treatment with carbendazim, T_6. Efficacy of *A. sativum* bulb extract (1% w/v) through seed treatment (ST) had resulted in improving plant stand of mustard in *Sclerotinia* infested field (Chattopadhyay *et al.* 2004). In the present investigation ST with garlic clove extract (2% w/v) was inferior to other biointensive and fungicidal treatments in terms of disease incidence and yield. Similarly during *rabi* 2009–10, *Sclerotinia* rot incidence was minimum (32.4%) in *Trichoderma* combination treatment, $T_{3,}$ with maximum in control T_8 (70.1%), and increased the seed yield (2.35 tonnes/ha) over control (1.63 tonnes/ha). Similar trends of reduction of disease incidence and increase of seed yield was found during *rabi* 2010–11. However, combination (seed + sprays) treatment, T_7 of carbendazim was at par with only seed treatment with *Trichoderma,* T_1 during 2008–09 and 2010–11 and seed + soil application of *Trichoderma,* T_2 during 2009–10. Overall mean of three years revealed that minimum incidence (11.1%) was recorded in *Trichoderma* based treatment, T_3 as compared to garlic bulb extract, T_5 (18.6%) and control, T_8 (26.9%). Seed yield was highest in *Trichoderma* based treatment T_3 (2.72 tonnes/ha) as compared to control T_8 (1.77 tonnes/ha). Results obtained here are in line with the results obtained in the laboratory under *in vitro* conditions (Amin *et al.* 2010, Yadav *et al*. 2011) as *T. viride* and *T. hamatum* significantly reduced the mycelial growth and sclerotial production of *S. sclerotiorum*. Gaur *et al.* (2010) also reported that seed treatment @10 g/kg and one

foliar spray @0.2% with mixed formulation of *T. hamatum* + *T. viride* gave minimum disease incidence (5.8%) and maximum yield (2.27 tonnes/ha) over rest of treatments.

Validation of integrated management of Sclerotinia *rot at Farmer's fields*

Multilocational validations of two integrated management practices were conducted during three crop seasons at farmer's field in Alwar and Sriganganagar district in Rajasthan and Gurgaon district in Haryana. Results of these studies, location wise are as under:

Sriganganagar region: During *rabi* 2008–09, management practice-I rendered highest average yields of 2.22 tonnes/ha and exhibited minimum (4.2%) *Sclerotinia* rot incidence (Table 3). Management practice-II was next with higher average yields of 2.01 tonnes/ha and moderate *Sclerotinia* rot incidence of 6.6%. In Farmer's practices (FP), maximum (20.2%) *Sclerotinia* stem rot incidence was observed with minimum average yield of 1.6 tonnes/ha. Similar trend of reduction of disease and increase in yield was observed during *rabi* 2009–10. During 2010–11, management practice-I rendered highest average yields of 2.44 tonnes/ha and exhibited minimum (6.0%) *Sclerotinia* rot incidence. Management practice-II was next with average higher yielding of 1.92 tonnes/ha and moderate *Sclerotinia* rot incidence of 14.0%. In Farmer's practices, maximum *Sclerotinia* rot incidence (25.7%) was recorded with minimum average yield of 1.53 tonnes/ha. Management practice-I rendered average (mean of three years) yields of 2.26 tonnes/ha and exhibited minimum (5.0%) *Sclerotinia* stem rot incidence as compared to practice-II (yield of 1.9 tonnes/ha and *Sclerotinia* rot incidence of 10.3%) and FP (yield of 1.52 tonnes/ha and *Sclerotinia* rot incidence of 22.8%). Thus *Trichoderma* based management practice differed significantly from garlic based and Farmers' practices. Maximum monetory return (Rs 52 897) was also obtained under practice-I which rendered IBCR ratio of 3.8 with maximum benefit of Rs 17 292 (Table 3). Practice-II was next in terms of monetory return (Rs 44 466) with IBCR of 2.0. Gaur *et al*. (2010) also reported that *Sclerotinia* rot of mustard can be effectively and economically managed by seed treatment @10 g/kg and one foliar spray @ 0.2% of talc based mixed formulation (2×10^8 c.f.u) of *T. hamatum* + *T. viride* with the ratio of 1:1 (w/w) at 50 DAS which gave the less disease incidence (7.9%), higher seed yield (3.26 tonnes/ha) and B: C ratio of 6.1:1 at adaptive trial centre, Srikaranpur in Sriganganagar .

Alwar region: During *rabi* 2008–09 management practice-I, rendered maximum average yield of 2.46 tonnes/ha and exhibited minimum (0.6%) *Sclerotinia* rot incidence, whereas in FP, minimum average yields of 2.07 tonnes/ha and disease incidence of 1.5% was recorded (Table 3). During *rabi* 2009–10, seed yield potential was on lower side due to heavy incidence of *Sclerotinia* rot which did not differ

Table 3 Economic viability of various management practices against *Sclerotinia* rot of Indian mustard (pooled mean of 2008-09, 2009-10 and 2010-11) in Sriganganagar and Alwar regions of Rajasthan

Management practice	*Sclerotinia* rot (%)				Yield (tonnes/ha)				Economics of management practice			
	2008–09*	2009–10*	2010–11*	Mean**	2008–09*	2009–10*	2010–11*	Mean **	Value of produce	Cost of input	Benefit over control	IBCR
Sriganganagar region												
I *Trichoderma*-based practices	4.2	4.8	6.0	5.0	2.22	2.13	2.44	2.26	52 897	4 610	17 292	3.8
II Garlic clove extract-based practices	6.6	10.4	14.0	10.3	2.01	1.78	1.92	1.90	44 466	4 414	8 861	2.0
Farmer's practices	20.2	22.5	25.7	22.8	1.60	1.44	1.53	1.52	35 605	1 325		
CD (P= 0.05)	1.8	2.0	2.5	2.1	0.09	0.13	0.16	0.13				
Alwar region												
I *Trichoderma*-based practices	0.6	19.5	8.0	9.4	2.46	1.53	2.40	2.13	50 136	2 926	10 789	3.7
II Garlic clove extract-based practices	0.8	31.3	15.0	15.7	2.28	1.19	2.16	1.88	44 572	3 183	5 225	1.6
Farmer's practices	1.5	56.5	24.5	27.5	2.07	1.03	2.06	1.72	39 347	1 480		
CD (P=0.05)	NS	13.3	7.5	11.2	0.10	0.30	0.10	0.10				

*Average of nine replication; ** average of 27 replications

significantly amongst two practices. *Trichoderma*-based management practice-I had performed best in yield (1.53 tonnes/ha) in comparison to FP (1.03 tonnes/ha) and reduced the disease intensity to 19.5 from 56.5% (FP). Average yield (mean of three years) of 2.13 tonnes/ha with disease incidence of 9.4% was recorded in *Trichoderma*-based management practice-I as compared to 1.72 tonnes/ha with maximum disease incidence of 27.5% in FP. Maximum monetory returns (₹ 50 136) over three years was also obtained in practice-I which rendered IBCR of 3.7 with net benefit of ₹ 10 789. Management practice-II was next in terms of monetory returns (₹ 44 572) with IBCR of 1.6. Chattopadhyay *et al.* (2004) also reported that seed treatments by *T. viride* and *Allium sativum* bulb extract (1% w/v) reduced the disease incidence of *Sclerotinia* rot, improved seed yield, plant height and initial plant stand over control at Bharatpur. Pathak and Godika (2010) reported the use of biofertilizers (*Azotobacter* and PSB) and *Trichoderma* along with recommended dose of fertilizer enhanced the plant growth, improved the yield and showed lesser incidence of diseases.

South-west Haryana (Gurgaon) region: During *rabi* 2008–09, management practice-I besides rendering highest average yield (2.33 tonnes/ha) also exhibited minimum (1.4%) *Sclerotinia* rot incidence. Management practice-II was next in order of production rendering average yields of 2.21 tonnes/ha. The *Sclerotinia* rot incidence under practice-II was 1.9%. Farmer's practices gave minimum average yields of 1.98 tonnes/ha. During *rabi* 2009–10 management practice-I gave maximum average yields of 1.61 tonnes/ha and also exhibited minimum (3.3%) *Sclerotinia* rot incidence. Management practice-II was next in order of production rendering average yields of 1.52 tonnes/ha. The *Sclerotinia* rot incidence under practice-II was 4.3%. Farmer's practices gave minimum average yields of 1.4 tonnes/ha, and had *Sclerotinia* rot incidence of 7.5%. During 2010–11, management practice-I again gave maximum average yields of 2.61 tonnes/ha and exhibited minimum (2.8%) *Sclerotinia* rot incidence. Management practice-II was next in order of production rendering average yields of 2.0 tonnes/ha. The *Sclerotinia* rot incidence under this practice was 8.4%. Farmer's practices gave minimum average yields of 1.85 tonnes/ha, and had *Sclerotinia* rot incidence of 10.0%. Average mean incidence (for three years) was 2.5% recorded in practice-I as compared to 7.2% in FP. Maximum (Rs 49 665) monetory returns over three years was also obtained under management practice-I, which rendered IBCR of 3.1. Management practice-II was next in terms of net returns.

From the results presented herein, *Trichoderma*-based management strategies resulted in minimum *Sclerotinia* rot incidence (mean values of three regions 5.6%) and maximum seed yield (2.19 tonnes/ha) as compared to garlic clove extract (mean values 10.3% and seed yield 1.90 tonnes/ha) and Farmers' practices (mean values 19.2% and seed yield 1.66 tonnes/ha). In this context present studies conclusively

Table 4 Economic viability of various management practices against *Sclerotinia* rot of Indian mustard (pooled mean of 2008-09, 2009-10 and 2010–11) in south-west (Gurgaon) region of Haryana

Management practice	Stem rot (%)				Yield (tonnes/ha)				Economics of management practice			
	2008–09*	2009–10*	2010–11*	Mean**	2008–09*	2009–10*	2010–11*	Mean **	Value of produce	Cost of input	Benefit over control	IBCR
I Trichoderma-based practices	1.4	3.3	2.8	2.5	2.33	1.61	2.61	2.18	49 665	3 327	10 299	3.1
II Garlic clove extract-based practices	1.9	4.3	8.4	4.9	2.21	1.52	2.00	1.91	43 454	3 359	4 088	1.2
Farmer's practices (FP)	4.2	7.5	10.0	7.2	1.98	1.40	1.85	1.74	39 366	1 350		
CD (P=0.05)	2.1	0.7	0.9	NS	0.13	0.05	0.10	0.33				

*Average of nine replication; ** average of 27 replications

demonstrate that *Sclerotinia* rot of Indian mustard can be effectively and economically managed by seed treatment @ 10 g/kg, soil application @ 2.5 kg/ha and two foliar spray @ 0.2% at 50 and 70 DAS with talc based mixed formulation (2×10^9 cfu/g) of *T. hamatum* + *T. viride* in the ratio of 1: 1 (w/w). This management practices will ultimately reduce the excessive use of chemical pesticides and side by side will save the expenditure. These findings are of immense value for mustard growing farmers.

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