

Sclerotinia rot in West Bengal

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Sclerotinia rot has been recorded on twenty one plants during the winter months (December to March) of 2011 –2013 in the plains of West Bengal. It includes twelve vegetables (*Solanum melongena*, *Solanum lycopersicum*, *Capsicum annum*, *Solanum tuberosum*, *Brassica oleracea* var. *capitata*, *Brassica oleracea* var. *botrytis*, *Phaseolus vulgaris*, *Pisum sativum*, *Dolichos lablab*, *Cucumis sativus*, *Lagenaria siceraria*, *Ipomoea aquatica*), two pulse crop (*Lens esculentum*, *Cicer arietinum*), one oilseed crop (*Brassica juncea*), three ornamental plants (*Tagetes erecta*, *Bellis perennis*, *Malvaviscus arboreus* var. *arboreus*), two medicinal plants (*Psoralea corylifolia*, *Ocimum sanctum*), and one weed (*Parthenium hysterophorus*). The disease was first time recorded in India on cucumber (*Cucumis sativus*), water spinach (*Ipomoea aquatica*), marigold (*Tagetes erecta*), lanka jaba (*Malvaviscus arboreus* var. *arboreus*), and Daisy (*Bellis perennis*). In all cases white mycelium of the pathogen covered infected area and dark coloured sclerotia formed on the infected tissue. In brinjal, potato, tomato, cabbage, cauliflower and in mustard sclerotia were formed inside the infected stem. In *Malvaviscus arboreus* var. *arboreus* infection was recorded at a height of 4-6 feet on branches, in *Psoralea corylifolia* at 2-4 feet height from the ground level. The disease has also been recorded in on bean and carrot at the market. All the isolates formed sclerotia on PDA medium. Variation among the isolates in relation to mycelial growth rate and number of sclerotia produced were recorded.

Key words: *Sclerotinia* rot, *Sclerotinia sclerotiorum*, host range, morphological and cultural variability

INTRODUCTION

Sclerotinia sclerotiorum (Lib.) de Bary causing *Sclerotinia* stem and root rot (syn. white rot, watery soft rot, cottony rot, white blight) of vegetables has been considered as a ubiquitous necrotrophic soil-borne plant pathogen. The pathogen has wide host range. Purdy (1979) has reported that *S. sclerotiorum* infects 64 plant families, 225 genera and in total it affects 383 plant species. But, subsequent survey during 1994 reflects a further increase in the host range of the pathogen. Pathogen is able to infect 408 plant species pertaining

to 75 families and 278 genera and most of them belong to Dicotyledonous subclass of Angiosperm (Bolland and Hall, 1994), and later Saharan and Mehta (2008) have reported that this fungus is able to infect about 500 species of plants. The disease has been reported from different parts of India. There are reports of existence of *Sclerotinia* rot (*S. sclerotiorum*) in potato (Dutta *et al.*, 2009) and French bean (Panja and Jana, 2001) in West Bengal. During 2011-2013 many plants have been found infected by this pathogen in the plains of West Bengal. Present study includes to record the incidence of the disease with detail symptoms in dif-

ferent plants in this state and also the nature of the pathogen.

MATERIALS AND METHODS

Surveys were conducted during November 2011 to December 2013 in nine districts of West Bengal to record the incidence of Sclerotinia rot in different crops. Symptoms and severity of the disease on individual crop was recorded. Disease samples were collected for isolation of the causal pathogen. The pathogen was isolated on PDA medium amended with chloramphenicol by putting sclerotia and small mycelial bit from the collected diseased tissue. In the laboratory, pathogen was maintained in PDA medium. Pathogenicity test was conducted for every isolate on their respective host. Mycelial strip from four days old culture prepared on PDA in Petriplate was used for inoculation. Small incision (0.5 cm length and 0.5-1mm depth) was given on the branch/vine of the plant at the internode and the mycelial strip was put on the injured tissue. Mycelial strip was then wrapped with thin film of moist cotton. Water was sprayed on the branch/vine and was covered with polythene bag to maintain humidity. Such inoculation was done after sunset in the month of January. In case of cauliflower and cabbage, the inoculation was done on the petiole and the inoculated leaf was covered with polythene bag. Four days after inoculation the polythene bag was removed. Disease symptom appeared at 5 to 10 days after inoculation. Cultural and morphological characteristic of *Sclerotinia sclerotiorum* isolates, from twenty different hosts were studied by growing this pathogen on PDA medium in Petriplates (90 mm). Mycelial disc from four days old culture of individual isolate was transferred to PDA medium (at the centre of the plate) and incubated at $22\pm 1^{\circ}\text{C}$ and observation were taken at every 24 h interval after inoculation.

RESULTS AND DISCUSSION

The disease are recorded on twenty one plants during the winter months (December to March) of 2011–2013 in nine districts of plains of West Bengal (Table1). It included twelve vegetables (brinjal, tomato, chilli, potato, cabbage, cauliflower, french bean, pea, dolicos bean, cucumber, bottle gourd, water spinach), two pulse crop (lentil, gram), one oilseed crop (mustard), three ornamental plants (marigold, daisy, lanka jaba), two medicinal plants (kakuch-*Psoralea corylifolia*, tulsi-*Ocimum sanc-*

tum) and a weed parthenium (*Parthenium hysterophorus*). The disease was recorded in nine districts of West Bengal. In all the cases white mycelium of the pathogen covered infected area and dark coloured sclerotia formed on the infected tissue. In brinjal, tomato mustard, cabbage and cauliflower sclerotia were formed inside the infected stem. In *Malvaviscus arboreus* var. *arboreus* infection was found at a height of 4-6 feet, and in *Psoralea corylifolia* at 2-4 feet from the ground level (Table 1). Brief symptoms of the disease on individual plant are presented below.

Symptoms of the disease on different host

Brinjal (*Solanum melongena* L.): Infection appeared on stem and branches. Infected area turned into fade brown colours and was covered with fluffy white mycelial mat. The parts of plant above the point of infection commonly dried up. Lesion on the fruit was light brown with distinct zonations. On opening the dry portion of the stem, pith was found to be filled with sclerotia of the pathogen. These were small or large elongated to cylindrical and often attached to each other end to end. The sclerotia were also formed on the infected stem, branches and on fruit along with white mycelium sticking to the host surface. Sclerotia were brown to black in colour.

Tomato (*Solanum lycopersicum* L.): The symptoms occurred either on the main stem or on branches. Water-soaked areas developed and became light to dark brown. A white mass of mycelium with embedded black sclerotia appeared on the infected stem and branch surface and in the pith of the stem. Affected fruits were rotted.

Chili (*Capsicum annuum* L.): Symptoms of the disease were found on the basal part of the stem and lower branches. Infected area turned brown and covered by the white mycelial growth of the pathogen. Leaves were also infected.

Potato (*Solanum tuberosum* L.): Sclerotinia rot first appeared as water soaked spots, usually at the point where branch attached with stem or on branches in contact with the soil. A white cottony growth of fungal mycelium developed on the lesions and infected tissues became soft and watery. The fungus spread rapidly to nearby stems and leaves if moisture present. Lesions then expanded and girdled the stem, causing the foliage to wilt. During dry conditions, lesions became dry. Sclerotia were initially

white to creamy in colour but became black with age. These sclerotia frequently developed in hollow center of infected stems and on the surface of stem.

Cabbage (*Brassica oleracea* var. *capitata* L.) and Cauliflower (*Brassica oleracea* var. *botrytis* L.): The earliest symptoms on cauliflower and cabbage appeared as loss of turgidity of leaves during the day time but the leaves recovered during night. Affected plants became dull white to pale yellow. Stem of plant became dark brown to black and covered with fluffy growth of the fungus in cool humid weather. Rotting of the stem led up to death of the plant. Sclerotia formed inside the stem. Head rot of cauliflower was also common. Sometimes affected leaves became detached from the stem.

Beans (*Phaseolus vulgaris* L.): The prominent white growth of the pathogen appeared on stems, petiole, leaves and pods. Sclerotia were formed on the disease tissue. In humid, wet and cool condition the disease spread fast and entire field was affected followed by death of majority of plants.

Pea (*Pisum sativum* L.): The tips of pods growing on or near the soil became infected and started to rot. Eventually, a wet rot resulted in complete decay. Infection commonly spread from diseased flower parts to the stem and branches. Dark lesion were formed on the stem and branch covered with white growth of the fungus. Black sclerotia were usually evident externally on the lesion as well as within diseased pods.

Bengal gram (*Cicer arietinum* L.): Wilting, in patches in the field, was the first noticeable symptom. On inspection black lesion were found on the base of the plant. In most cases such lesion was covered with white mycelium and greenish black sclerotia were on the stem.

Dolichos bean (*Dolichos lablab* L.): Symptoms first appeared as water soaked lesion on the leaves and branch. These lesions gradually enlarged with white fluffy mycelial growth on the branch and leaf. The tips of fruits growing on or near the soil became infected and started to rot. Infection commonly spread to the inflorescence resulting rotting of flower and fruit. The white mycelium and black sclerotia were usually evident externally and also within diseased fruits.

Cucumber (*Cucumis sativus* L.): The disease initiated as water soaked area on fruits followed by a white mycelial growth covering the infected area. Three to five days later, the fruits were rotted completely. Similar symptoms appeared on stem and leaves resulting the death of the vine.

Bottle gourd (*Lagenaria siceraria* (Mol.) Standl.): The disease was noticed on this crop raised over scaffold. Fruits were severely affected. Infection started at the blossom end of the hanging fruit and gradually proceeded upward. Infection was found on very small tender and also on the large fruits. Infected area was covered with white mycelial growth embedded with sclerotia. Vines were infected at the internodes that led up to death of infected vine.

Water spinach/Kalmi (*Ipomoea aquatica* Forssk.): Symptom recorded on the stem touching the ground and white growth of the fungus was found covering the infected stem. Brown lesion was found on the leaves. The disease destroyed entire cropped area. White mycelial growth of the pathogen was found on the soil surface. A few sclerotia were formed on the infected plant.

Lentil (*Lens esculentus* Moench.): Infection occurred in the field in patches from early flowering to pod setting stage at the base of the plant. Infected areas turned brown and were covered with white fluffy growth of the pathogen. Dark brown to black sclerotia developed on the infected tissue. Infected plants wilted and dried in the field in patches.

Mustard (*Brassica juncea* L.): Symptoms on the stem became visible as elongated, water-soaked lesions, which later were covered by a cottony mycelial growth of the fungus. When the stem was completely girdled by such lesions, the plant wilted and became dry. Sclerotia were formed on the white mycelial growth and inside the infected stem.

Marigold (*Tagetes erecta* L.): Initial symptoms appeared on the main stem and branches. The affected portion underwent rotting in wet condition, covered with white fungal growth. Infection encircled the stem or branches leading to death of the stem or branches above the point of infection. Some of the infected plants were covered with white growth of the pathogen. Sclerotia were also formed in the infected stem or branches.

Table 1 : Plants infected by *Sclerotinia sclerotiorum* in West Bengal during 2011-2013

Common name of the host	Sc. name of the host	Time of recoding disease infestation	Place of incidence	Parts of plant infected	Severity of the disease
Brinjal	<i>Solanum melongena</i>	January to March (2011-12)	Nadia, Coochbehar, Jalpaiguri, Dakhin Dinajpur Birbhum, Mursidabad, Hooghly, Purulia,	Stem, branches, leaf petiole, fruit	High
Tomato	<i>Lycopersicon esculentum</i>	January (2013)	Jalpaiguri	Stem, branch, fruit	High
Chili	<i>Capsicum annum</i>	January (2013)	Jalpaiguri	Stem, branch	Moderate
Potato	<i>Solanum tuberosum</i>	January (2013)	Jalpaiguri , Nadia	Stem	Moderate
Cabbage	<i>Brassica oleracea var. capitata</i>	January (2013)	Jalpaiguri	Stem, head, leaf	High
Cauliflower	<i>Brassica oleracea var. botrytis</i>	January (2013)	Jalpaiguri	Stem, head, leaf	Moderate
French bean	<i>Phaseolus vulgaris</i>	January to February (2011- 2013)	Nadia , Purulia, Jalpaiguri	Stem, pod, leaf	High
Pea	<i>Pisum sativum</i>	January (2013)	Jalpaiguri	Stem, flower, pod	High
Dolichos bean	<i>Dolichos lablab</i>	January to Febuary (2013)	Jalpaiguri, Nadia	Branches, inflorescence, pod	Moderate
Cucumber	<i>Cucumis sativus</i>	January to Febuary (2013)	Purulia , Nadia	Vine stem, fruit	Moderate
Bottle gourd	<i>Lagenaria siceraria</i>	January (2013)	Jalpaiguri	Vine stem, fruit	High
Water spinach	<i>Ipomoea aquatica</i>	December to February (2011, 2012, 2013, 2013-2014)	Nadia	Stem, leaf	High
Gram	<i>Cicer arietinum</i>	January (2012, 2013)	Coochbehar	Stem	Moderate
Lentil		February (2011,2012,2013)	Nadia, Coochbehar	Stem	Moderate
Mustard	<i>Lens esculentus</i>	January (2013)	Jalpaiguri	Stem	Moderate
Marigold	<i>Brassica juncea</i>	January to March (2011, 2012,2013)	Nadia, Howrah	Stem, branches, leaf,flower	High
Daisy	<i>Tagetes erecta</i> <i>Bellis perennis</i>	January (2011-12)	Nadia	Leaf	Mild
Lanka jaba	<i>Malvaviscus arboreus var. arboreus</i>	January to March (2012, 2013)	Nadia	Branches	Moderate
Tulsi	<i>Ocimum sanctum</i>	January to march (2011, 2012, 2013)	Nadia	Entire plant	High
Kakuch	<i>Psoralea corylifolia</i>	January-February (2012, 2013)	Nadia	Branches	Moderate to high
Parthenium	<i>Parthenium hysterophorus</i>	Febuary (2011, 2012)	Nadia	Stem	Moderate

Daisy (*Bellis perennis* L.): Light brown patches appeared on petiole and leaf laminas and the infected leaves dried up. Stem at the soil level was also affected. A few sclerotia were formed on this plant.

Lanka Jaba (*Malvaviscus arboreus var. arboreus* Cav.) and Kakuch (*Psoralea corylifolia* L.): Infection was observed on the branches. In former plant white mycelial growth of the fungus was found at a height of 4-6 feet from the ground

Table 2 : Morphological and cultural variability of different isolates of *Sclerotinia sclerotiorum* on PDA medium

Host	Avg. radial growth of mycelia at diff. hours (cm)				Time taken for first sclerotia formation	Avg. No of Sclerotia / plate	Mycelial characters	Formation of sclerotia on the medium	Location of sclerotia in plate	Secretion of sclerotia	Wt. of 10 sclerotia (g)	
	24	48	72	96							Fresh	Dry
Brinjal	0.8	3.43	7.33	9.0	72	21	Surface*	Surface †	Peripheral	(+ve)	0.131	0.056
Tomato	3.93	9.0			48	18	Surface	Surface	Peripheral	(+ve)	0.160	0.077
Chili	0.9	5.06	9.0		96	22	Surface	Surface	Peripheral	(+ve)	0.156	0.080
Potato	0.83	3.43	7.76	9.0	96	20	Surface	Surface	Peripheral	(+ve)	0.256	0.122
Cabbage	1.36	5.6	9.0		96	17	Surface	Surface	Peripheral	(+ve)	0.178	0.082
Cauliflower	1.16	7.5	9.0		72	11	Surface	Surface	Peripheral	(+ve)	0.075	0.030
French Bean	1.1	3.16	7.36	9.0	72	11	Surface	Surface	Peripheral	(+ve)	0.152	0.065
Pea	2.26	8.13	9.0		72	19	Surface	Surface	Scattered	(+ve)	0.202	0.093
Lentil	1.36	3.56	7.7	9.0	72	21	Surface	Surface	Scattered	(+ve)	0.226	0.105
Dolichos bean	1.03	6.03	9.0		96	10	Surface	Surface	Peripheral	(+ve)	0.060	0.013
Cucumber	1.63	5.26	8.53	9.0	72	20	Surface	Surface	Peripheral	(+ve)	0.250	0.116
Bottle gourd	1.1	4.63	8.63	9.0	96	22	Surface	Surface	Peripheral	(+ve)	0.181	0.087
Water Spinach	1.76	4.6	8.56	9.0	72	12	Surface	Surface	Peripheral	(+ve)	0.103	0.056
Mustard	1.7	7.93	9.0		72	10	Surface	Surface	Peripheral	(+ve)	0.148	0.069
Marigold	1.0	3.53	7.6	9.0	72	17	Surface	Surface	Central ring	(+ve)	0.155	0.102
Daisy	1.0	2.96	6.96	9.0	72	19	Surface	Surface	Peripheral	(+ve)	0.160	0.078
Lanka jaba	0.7	4.23	8.3	9.0	72	10	Surface	Surface	Peripheral	(+ve)	0.284	0.145
Tulsi	1.16	4.23	8.3	9.0	72	17	Surface	Surface	Peripheral	(+ve)	0.168	0.084
Kakuch	1.0	4.13	7.36	9.0	96	16	Surface	Surface	Peripheral	(+ve)	0.122	0.054
Parthenium	0.5	1.13	2.73	9.0	72	17	Surface	Surface	Peripheral	(+ve)	0.129	0.084

* The fungus grew over the surface of the PDA medium

† The fungus formed on the surface of the mycelia growth

level, whereas in Kakuch (*Psoralea corylifolia*) it was 2-4 feet height. Lesions covered the branches up to a length of 2-6 inch. Branches dried up above the point of infection.

Tulsi (*Ocimum sanctum* L.): The branches, twigs, leaves and inflorescence were affected by the disease. Entire plant were covered with white powdery type growth of the causal pathogen. Ultimately the infected plant died. Very few sclerotia were produced on such plants.

Parthenium (*Parthenium hysterophorus* L.): This is a road-side weed and stem of plants were infected by this pathogen in the month of February. Many plants were wilted and dried. Dark black sclerotia were formed on the infected portion of the plant.

In addition to the incidence of the disease in field, the disease had also been recorded in on bean pod at Kalyani market and carrot at Kolkata market.

Sclerotinia rot is first time recorded in India on water spinach/kalmi (*Ipomoea aquatica* Forssk), cucumber (*Cucumis sativus* L.), marigold (*Tagetes erecta* L.), lanka jaba (*Malvaviscus arboreus* var. *arboreus* Cav.) and daisy (*Bellis perennis* L.). The disease has been recorded on rest of the plants in other states of India. The disease on potato was recorded earlier in West Bengal by Dutta *et al.*, (2009) and

french bean (Panja and Jana, 2001). Incidence of the disease on water spinach/Kalmi (*Ipomoea aquatica* Forssk), cucumber (*Cucumis sativus* L.), marigold (*Tagetes erecta* L.), lanka jaba (*Malvaviscus arboreus* var. *arboreus* Cav.), daisy (*Bellis perennis* L.) and tulsi (*Ocimum sanctum* L.) was recorded for last three consecutive years in a domestic garden at Nadia district during December to February. This is an evidence of the strong survival potential of the pathogen. Incidence of Sclerotinia rot on many plant species and repeated occurrence in the same field in West Bengal has created an alarming situation.

Morphological and cultural variability of

***Sclerotinia sclerotiorum* :** All the twenty isolates grew over the surface of the PDA medium and growth rate of tomato isolate was highest compared to other isolates and it covered the entire plates (90 mm) within 48 h and sclerotia formation started at 48 h of growth (Table 2). Isolates of *S. sclerotiorum* from chilli, cabbage, cauliflower, pea, dolichos bean and mustard covered entire medium surface at 72 h but other isolates took 96 h. Sclerotia formation started after 96 h incubation in chili, potato, cabbage, dolichos bean, bottle gourd and kakuch isolates. All other isolates formed sclerotia after 72 h. Sclerotia were formed on the surface of the mycelial growth over the agar medium. The isolates showed good variation in respect to number of sclerotia produced. Isolates from cauliflower,

french bean, dolichos bean, water spinach, mustard, lanka jaba produced 10-12 sclerotia per plate, tomato, cabbage, marigold, tulsi, kakuch and parthenium isolates produced 16-18 sclerotia, brinjal, potato, pea, cucumber, bottle gourd and daisy isolate 19-22 sclerotia per plate. No relation could be found in between growth rate and number of sclerotia produced. Seventeen isolates formed sclerotia at the periphery of the medium but isolate from pea and lentil plant sclerotia were scattered on the surface of the medium. Marigold isolate formed sclerotia towards the middle portion of the plate in the form of a ring. Iqbal *et al.* (2003) recorded sclerotia formation at the centre and periphery in brinjal isolate. The same isolate produced much more sclerotia compared to present brinjal isolate. In another report (Bag and Dutta, 2009), pattern of sclerotia formation was scattered towards periphery over the medium by *Ocimum* isolate. A few days after formation of sclerotia, a yellow colour fluid was deposited on the surface of the sclerotia in all the isolates. There was good variation in fresh weight of sclerotia of different isolates. (Table 2). Fresh weight of ten sclerotia ranged from 0.060 to 0.284 g and dry weight from 0.013 to 0.145 g. Comparatively larger (0.226-0.284 g) sclerotia were formed by potato, cucumber, lentil and lanka jaba isolates and smallest (0.060 g) sclerotia by cauliflower isolate. Weight of fresh sclerotia of other isolates varied from 0.103 to 202 g. No correlation could be established between size of sclerotia and growth rate/time of initiation of sclerotia. Further study, with these isolates is in progress. Variation of *S. sclerotiorum* strains based on colony colour on growth media, mycelial growth (radial colony growth) and growth rate, differences in sclerotia (number, size, weight etc.), and ascospores have also been reported in previous studies from different countries (Morrall *et al.* 1972; Marukawa *et al.* 1975; Price and Colhoun 1975; Kohn *et al.* 1991; Akram *et al.*, 2008; Li *et al.* 2008; Irani *et al.*, 2011).

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