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Effect of Wetting on the Pathogenesis and Survival of Sclerotia of *Rhizoctonia solani*

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Sclerotia of *Rhizoctonia solani* grown on maize-meal sand medium when subjected to wetting either continuously or alternately in a cycle of wetting and drying lost the ability to cause infection on excised rice leaves and their germination was also reduced. Materials were leaked out from submerged sclerotia resulting in lose in weight. Although 35-day old sclerotia cultured either on MSM or PDA behaved similarly in respect of pathogenecity and germination, 75-day old sclerotia grown on MSM lost these activities but those cultured on PDA still remained pathogenic.

Key Words - Drying Infection Leakage Sclerotia Wetting

Wetting of sclerotia and/or their burial in waterlogged soil, in certain fungi, has been implicated for the loss of their viability and capacity to incite infection (Sewell, 1970; Coley-Smith 1979; Gladders & Coley-Smith 1980; Sharm et al. 1983). Similar response of the sclerotia of *Rhizoctonia solani* Kuhn (teleomorph *Thanatephorus sasakii* (Shirai) Tu and Kimbrough = *T. cucumeris* (Frank) Donk, the causal fungus of the sheath blight of rice has been observed (Roy, 1986) which is considered responsible for fluctuation of the disease in the typical wet-land rice field. Coley-Smith (1979) and Gladders and Coley-Smith (1980) recorded that leakage of material from the sclerotia was responsible for the loss of viability. Therefore, experiments were conducted to find out whether such a mechanism existed in the sclerotia of *R. solani*, and also the capacity of the treated sclerotia to incite infection on rice leaves.

MATERIALS & METHODS Sclerotia, 2-month old, grown on 2% maize-meal sand medium (MSM) were subjected to continuous immersion for 6 days and alternate wetting and drying in a cycle of 3-3-3 days. After the treatment, the sclerotia were used to inoculate excised rice leaves (cv. Mashuri) kept in transparent plastic petri dishes, 13 cm diameter, each containing 7 to 8 leaf pieces. Three plates were used for each treatment. The excised pieces were arranged in parallel on a moist cotton pad inside a petri dish and both the cut ends were covered with moist cotton wool drawn in the form of wick (Fig. 1) to keep the leaf pieces in position as well as continuously keeping them moist. A sclerotium was then placed on each leaf piece. As bacterial growth developed in water in a few tubes in which the sclerotia were dipped. Sclerotia in one lot were surface sterilized with 2% sodium hypochlorite before inoculation. The test was repeated with the treatment of continuous wetting for 8 days and continuous wetting for the same period but changing the water daily with 70-day old sclerotia.

Loss of weight of the treated and control sclerotia was determined. The treated sclerotia after 7-day wetting in sterile distilled water were dried in an oven to constant weight. Similarly, the sclerotia in control treatment were dried in oven to constant weight.

Estimation of sugars in the leaked out material was determined by Shaffer Somogyi micro method (AOAC 1975). Sclerotia 2/3 were steeped in 25 ml sterile distilled water in a tube for 7 days and after removing them, the water was analysed for sugar. In another treatment a few drops o toluene were added to the water in the tube. After using the water of the first treatment (i.e. without toluene), the sclerotia were removed, air-dried and sugars retained in them were determined. Sugars present in the air-dried sclerotia of the control treatment were also estimated.

To find out the effect of age of sclerotia and media on the sclerotial survival and pathogenicity, sclerotia were grown on 2% MSM and PDA for 35 and 75 days designated as 'young' and 'old', respectively. They were dislodged from the plates with the help of a pair of forceps and subjected to wetting treatment (Table 1). After treatment, germination of the sclerotia was recorded on PDA fortified with streptomycin sulphate, and their capability to infect rice leaves was demonstrated by the methods described above. Four plates of 9.5 cm diameter each containing 4 sclerotia and 3 plates of 13 cm diameter each containing 7 leaf pieces were used to test viability of the sclerotia and their efficacy to incite infection, respectively. The sclerotia were surface sterilized with 2% sodium hypochlorite before use in both the tests.

RESULTS In the first test, while all the sclerotia (60-day old) maintained in the laboratory caused infection on the leaves, 59% (13 out of 22) of the sclerotia subjected to continuous wetting for 6 days caused infection. When the sclerotia were subjected to alternate wetting and drying, they lost the capability to cause infection (Fig. 1). Only one sclerotia out 22 produced lesion. There was no difference between sterilized and unsterilized sclerotia in inciting in-

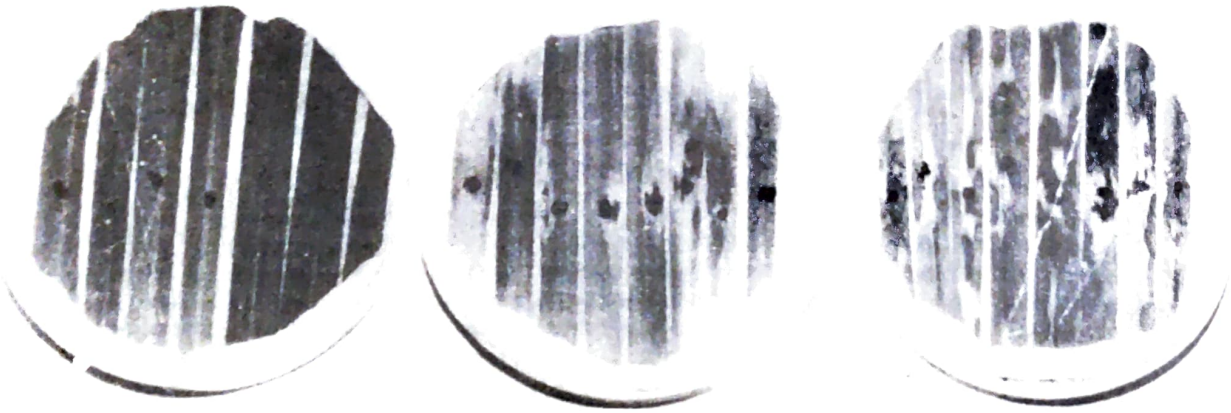

Fig. 1 Infection on detached rice leaves by sclerotia of *R. solani* (from left) infection on the leaves by air-dried sclerotia (control), reduced infection by sclerotia continuously submerged for 9 days, and no infection by sclerotia subjected to alternate wetting and drying in a cycle of 3-3-3 day.

fection. When the test was repeated with 70-day old sclerotia kept continuously immerged for 8 days, only one out of 21 caused infection irrespective of whether the water was changed daily or not. In the control, all the 21 sclerotia incited infection.

Loss of material from 2-month old sclerotia grown on MSM when kept immersed continuously for 6 days varied from 12.1 to 14.6% in the two tests, and that in the treatment of alternate wetting and drying in a cycle of 3-3-3 days from 16.0 to 16.1%, respectively. In the first test, another treatment of 3-day continuous wetting was included in which the loss was 11.7%. Of the leaked out materials, 0.53% comprised of sugar (Table 2). Sugars equivalent to 0.16 to 0.20% of sclerotial weight were lost in water during 7-day wetting.

Table 1 shows that PDA-cultured irrespective of their age (35 or 75 days, respectively) germinated equally well in the treatment of continuous wetting for 12 days and alternate wetting and drying in a cycle of 3-3-3-3 day as in the control. Similar was the result in inciting infection on rice leaves. Although the young MSM-cultured sclerotia behaved similarly, the old sclerotia lost the capacity to germinate and infect rice leaves - out of 7 leaf pieces per plate, almost all were infected by the young and old PDA-grown and young MSM-grown sclerotia but none by old MSM grown sclerotia.

Table 1 Number of Sclerotia (old and young) Germinated after Different Treatments of Immersion

Treatment	PDA		Maize-meal sand	
	Young	Old	Young	Old
Alternate immersion and drying (3-3-3-3 day cycle)	4	4	4	0
Continuous immersion for 12 days	4	4	4	0
Control (dry under lab condition)	4	4	4	4

Table 2 Sugar Constituent of the Sclerotia and (sugar) Fraction Leaked out in Water in which the Sclerotia were steeped.

Treatment	% reducing sugar	% sucrose
Dried sclerotia	0.32	0.21
Dried sclerotia after 7-day wetting	0.27	0.15
Water in which sclerotia steeped for 7-days in the above treatment	0.05	0.11
Same as above but toluene added	0.09	0.11

DISCUSSION Carbohydrates constitute a major group of storage products in sclerotia (Coley-Smith 1979); therefore, leaking out of such substances resulting in loss of

weight might be a factor for loss of pathogenicity of the sclerotia of *R. solani*. Quantity of sugar in the leaked out material from the sclerotia when toluene was added in water in which the sclerotia were immersed was more than without toluene (Table 2). This might be due to the utilization of a part of the sugar component by the contaminant bacteria in the latter treatment. Greater weight loss in the treatment of alternate wetting and drying than in continuous wetting is in accord with the observations of Gladders and Coley-Smith (1980) on the sclerotia of *R. tuliparum*.

Difference in aggressiveness of the sclerotia of *R. solani* grown on different media had been noted by Dath (1984) and PDA was a favoured medium but she did not find any significant difference on the age level of the sclerotia on aggressiveness upto 40 days. However, this study shows that 75-day old sclerotia loss viability to incite infection when grown on MSM and subjected to wetting but sclerotia of the same age cultured on PDA still retain the capacity to infect. These studies indicate that sclerotia of *R. solani* cannot remain viable in water-logged condition in wetland rice field so as to cause infection in the succeeding rice crop; however, in upland situation (and on the bunds in rice fields), these (sclerotia) remain viable and retain their capacity to incite infection (Roy 1984).

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