

RELATION OF SCLEROTIAL INOCULUM DENSITY AND SOIL  
MOISTURE TO INFECTION OF FIELD CROPS BY  
*MACROPHOMINA PHASEOLINA* \*

A. HAKEEM SHEIKH AND A. GHAFFAR

*Department of Botany, University of Karachi, Karachi-32, Pakistan.*

Abstract

The effect of sclerotial density of *Macrophomina phaseolina* at various soil moisture levels on disease incidence in black gram, guar, okra and cotton was investigated. Infection percentage increased linearly with the increase in inoculum density of sclerotia in soil but varied inversely in relation to soil moisture regime. Infection percentage was consistently higher at 30 days compared to that at 15 days of experimental period. The susceptibility to *Macrophomina* infection varied in the test species. Whereas a sclerotial density of 5/g of soil gave 50% infection in black gram, 20 sclerotia/g of soil were required for okra, guar and 40 for cotton at 25% water holding capacity (W.H.C.) Inoculum density of upto 40 sclerotia/g soil coupled with high soil moisture (100% W.H.C.) did not produce more than 50% infection.

Introduction

*Macrophomina phaseolina* (Tassi) Goid., [*Rhizoctonia bataticola* (Taub.) Butl.] is known to produce seedling blight, charcoal rot, root rot, stem rot and pod rot on over 400 species of plants. The fungus is widely distributed in tropical and subtropical countries of the world (Young, 1949) of which atleast 40 economic hosts have been recorded from Pakistan alone (Ghaffar, *et al*, 1964). The fungus is believed to persist in soil in the form of small, black sclerotia which are produced in large numbers on infected host tissues and subsequently dispersed in soil during tillage operation (Cook *et al*, 1973 Smith, 1969).

Sclerotial inoculum of *M. phaseolina* are reported to produce greater mortality of pine seedlings than mycelial inoculum (Smith, 1969). The sclerotial densities in soil have been correlated with increased disease incidence in bean (Watanabe, *et al*, 1979) but numerical threshold for infection were not described. Meyer *et al* (1974) reported that with increase in inoculum levels the percentage infection of *M. phaseolina* on soybean seedlings also increased. Incidentally, Meyer *et al* in their experiment used 150-700 sclerotial propagules/g soil that gave a maximum of 55% infection. This appears to be an excessively

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large number since a native population of upto 29 sclerotial propagules/g have been recorded in fields of cotton (Sheikh & Ghaffar, 1975) and sunflower (Albouvette, 1976). Even Meyer *et al* (1973) have estimated population of sclerotial propagules/g of soil as: soybean, 108; corn, 31; wheat, 24; and alfalfa, 15. This study shows the infection of *M. phaseolina* on black gram, guar, okra and cotton as related to sclerotial densities in soil at different moisture levels.

### Materials and Methods

The isolate of *M. phaseolina* used in this study was obtained from root rot of cotton (Cult. No. 54, Bot. Dept. Karachi University). This was the same isolate used by Ghaffar *et al* (1969) in their investigations. The fungus was grown in corn meal sand medium (5%, w/w) for 2 weeks at 30°C and sclerotia separated by successive floatation in distilled water and decantation (Sheikh & Ghaffar, 1975). The sclerotia were dried on filter paper at room temperature and stored in glass vials for subsequent use.

Sandy loam, pH 8.3, from cotton field of Karachi University experimental plots was passed through 2 mm sieve before use. The soil was artificially infested with sclerotia of *M. phaseolina* (0.01:100; w/w) which gave a population of 80 sclerotial propagules/g of soil. This was double diluted with raw soil. Final population was 40,20,10,5,2 and 1 sclerotia/g of soil on an oven dry weight basis. Population of sclerotia was determined by our wet sieving and dilution technique (Papavizas & Klag, 1975; Sheikh & Ghaffar, 1975). Twenty g soil was wet-sieved through 120 and 300 mesh screen and the residue obtained on 300 mesh sieve was transferred in 0.5% CaClO and continuously agitated with a magnetic stirrer. One ml aliquot of the sclerotial suspension (1:5, w/v) were removed and pipetted on to the surface of 3-day old agar plates and evenly distributed. PDA supplemented with penicillin and streptomycin @ 60 mg/l; Demosan (300 mg/l) and rose bengal (100 mg/l) was used as a selective medium. The plates were incubated at 30°C, and greyish to black colonies of *M. phaseolina* were recognizable easily within 5 days. One ml aliquot of the sclerotial suspension was also distributed on the strip of filter paper. Counting the number of sclerotial propagules on filter paper and those obtained on agar plates, the recovery was 96%.

Soil infested with sclerotia of *M. phaseolina* was transferred into 7.5 cm diam plastic pots, 300 g/pot. Two-day old seedlings of black gram (*Vigna mungo* (L.) Hepper), guar (*Cyamopsis tetragonoloba* (L.) Taub.), okra (*Abelmoschus esculentus* (L.) Moench) and cotton (*Gossypium hirsutum* L.), were transplanted into soil. @ 5 seedlings/pot. Soils were adjusted and maintained at 25,50 and 100% W.H.C. (Keen & Raczkowski, 1921) by addition of water twice a day. Variation in soil water levels encountered was not more than 2%. Treatments were replicated 4 times each and non-infested soil served as control. After 15 and 30 days of growth, seedlings were taken out and number of plants infected by *M. phaseolina* was determined by transferring disinfected root pieces of the test plants on PDA.

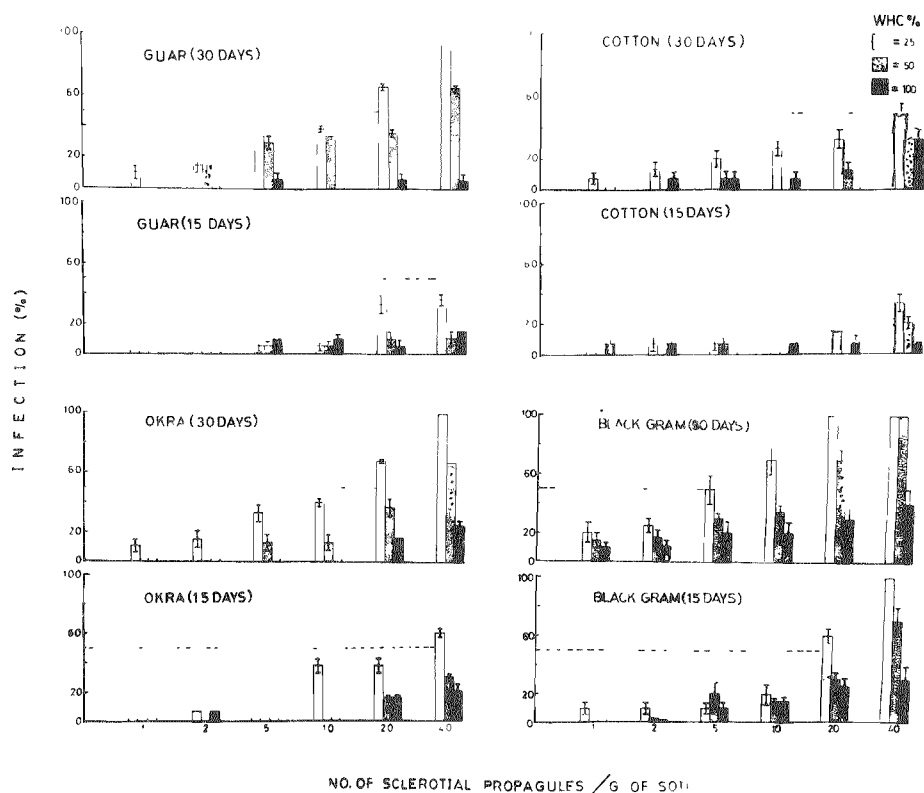


Fig. 1. Relationship of inoculum density of *Macrophomina phaseolina* sclerotia and soil moisture level on infection of black gram, okra, guar and cotton after 15 and 30 days of planting.

## Results

Infection of *M. phaseolina* was related to the inoculum densities of sclerotia in soil. Infection was generally lower at higher soil moisture; more infection being observed in soil at low moisture level of 25% W.H.C. (Fig. 1). It is interesting to note that even at low inoculum level of 1-2 sclerotial propagules/g of soil, 12-20% plants were infected at 25% W.H.C. after an experimental period of 30 days of planting. For 50% infection an inoculum level of sclerotia/g of soil was 5 for black gram, 20 for okra and guar and 40 for cotton. Infection was found to increase and the appearance of disease was rapid with the increase in sclerotial density. For 100% infection, number of sclerotia/g of soil was 10 for black gram and 40 for okra and guar. In general, greater disease incidence was noticed at low moisture level of 25% W.H.C. with a tendency to show less infection at 50 to 100% W.H.C. In soil at high moisture level (100% W.H.C.) and with an inoculum of upto 40 sclerotia/g the infection remained less than 50%.

## Discussion

The results of the present investigation indicates that even a low number of sclero-

tial propagules in soil is capable of causing root rot infection. This is contrary to the report of Meyer *et al.*, (1974) who found that 700 sclerotial propagules/g soil could produce only 55% infection on soybean. Our experiment further confirm the field observations where low number of sclerotial propagules were detected in fields of cotton (Sheikh & Ghaffar, 1975), sunflower (Albouvette, 1976), soybean, corn, wheat and alf-alfa (Meyer, *et al.*, 1973) infected with *Macrophomina* root rot. Although inoculum density was directly related to disease severity, *Macrophomina* infection was preeminently conditioned by soil moisture. Disease incidence was greater at low moisture level and increasing levels of soil moisture reduced infection on test plants. Moisture stress, in addition to high temperature, has been reported to favour charcoal rot of sorghum (Edmunds, 1964) and cotton (Ghaffar & Erwin, 1969). Our results on the reductions of sclerotial population in high soil moisture regimes (Sheikh & Ghaffar, unpublished data) are similar to those reported by Dhingra & Sinclair (1975) who showed that population of free sclerotia can be decreased by high soil moisture as compared to dry soil. This phenomenon would also explain the reduction in charcoal rot in cotton (Ghaffar & Erwin, 1969), soybean (Meyer *et al.* (1974) and pine seedlings (Hodges, 1962) by frequent irrigations or as observed during rainy season. This is similar to the reports for *Fusarium roseum* (Cook & Papendick, 1970) and for *Theilaviopsis basicola* (Papavizas & Lewis, 1979). These results would also suggest a practical cultural control of *Macrophomina* infection by keeping soil moisture at sufficiently higher level.

#### References

- Albouvette, C. 1976. Recherches Sur l'ecology des Champignons parasites dens le soi. Ann. Phytopathology, 8: 147-157.
- Cook, G.E., M.G. Boosalis, L.D. Dunkle and G.N. Odvody. 1973. Survival of *Macrophomina phaseoli* in corn and sorghum stalk residue. Plant Dis. Rept., 57: 873-875.
- Cook, R.J. and R.I. Papendick. 1970. Soil water potential as a factor in the ecology of *Fusarium roseum* f. sp. *cerealis* 'Culmorum'. Plant Soil, 32: 131-145.
- Dhingra, O.D. and G.B. Sinclair. 1975. Survival of *Macrophomina phaseolina* sclerotia in soil: Effect of soil moisture, carbon nitrogen ratios, carbon sources and nitrogen concentrations. Phytopathology, 65: 236-240.
- Edmunds, L.K. 1964. Combined relation of plant maturity, temperature and soil moisture to charcoal rot development in grain sorghum. Phytopathology, 54: 514-517.
- Ghaffar, A., and D.C. Erwin. 1969. Effect of soil water stress on root rot of cotton caused by *Macrophomina phaseoli*. Phytopathology, 59: 795-797.
- Ghaffar, A., A. Kafi and R. Mirza. 1964. Some new hosts of *Macrophomina phaseoli* (Maubl.) Ashby. Pak. Jour. Sc. Ind. Res., 7: 71-72.
- Keen, B.A. and Raczkowski, H. 1921. The relation between clay content and certain physical properties of soil J. Agri. Sci. 11: 441-449.
- Hodges, C.S. 1962. Black root rot of pine seedlings. Phytopathology, 52: 210-219.

- Meyer, W.A., J.B. Sinclair and M.N. Khare. 1973. Biology of *Macrophomina phaseoli* in soil studied with selective media. *Phytopathology*, 63: 613-620.
- Meyer, W.A., J.B. Sinclair and M.N. Khare. 1974. Factors affecting charcoal rot of soybean seedlings. *Phytopathology*, 64: 845-849.
- Papavizas, G.C. and N.G. Klag. 1975. Isolation and quantitative determination of *Macrophomina phaseolina* from soil. *Phytopathology*, 65: 182-187.
- Papavizas, G.C. and J.A. Lewis. 1971. Survival of endoconidia and chlamydospores of *Thielaviopsis basicola* as affected by soil environmental factors. *Phytopathology*, 61: 108-113.
- Reichert, I., and E. Hellinger. 1947. On the occurrence, morphology and parasitism of *Sclerotium bataticola*. *Palestine J. Bot.*, 6: 107-147.
- Sheikh, A.H. and A. Ghaffar. 1975. Population study of the sclerotia of *Macrophomina phaseoli* in the mortality of pine seedlings under varying soil conditions. *Phytopathology*, 59: 379-382.
- Watanabe, T., R.S. Smith, JR. and W.C. Snyder. 1967. Populations of microsclerotia of the soil-borne pathogen, *Macrophomina phaseoli*, in relation to stem blight of bean. *Phytopathology* 57: 1010 (Abstr.).
- Young, P.A. 1949. Charcoal rot of plants in East Texas. *Tex. Agric. Exp. Stn. Bull.* 712. 33 p.