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ROLE OF INTERNAL SEED BORNE MYCELIUM OF DOWNY MILDEW OF PEARL MILLET IN THE TRANSMISSION OF DISEASE IN THE WESTERN RAJASTHAN

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Downy mildew disease caused by *Sclerospora graminicola* (Sacc.) Schroet, has become a major obstacle to harness the high yield potential of newly developed varieties/hybrids of pearl millet (*Pennisetum glaucum* (L.) R.Br.) To develop a suitable control measure, knowledge on exact mode of transmission of this pathogen is of paramount importance. The pathogen is present as oospore in the soil and on the seed surface, and as a dormant mycelium in extra and intra embryonal tissues (Williams 1979). Circumstantial evidences are only known regarding the viability of seed borne mycelium and very little is known about their role in the transmission of disease in Western Rajasthan. Experiments were therefore, undertaken for detection and testing of the viability of seed borne mycelium and their role in the transmission of disease.

Seeds formed close to the affected parts of the partially transformed ears of downy mildew infected plants of cv. HB 3 were seleted for the study in rainy season of 985, at Central Arid Zone Research Institute, Jodhpur. Seeds having moisture conent between 6.0 to 7.5% were kept at temperature of 28°C ± 2°C till sowing. Further studies were also carried out at the Danish Government Institute of Seed Pathology for Developing Countries and Institute of Spore Plants, Copenhagen, Denmark. For detection of the mycelium in the seed tissues and for testing their viability, 1000 seeds were taken and modified embryo test procedure of Shetty et al. (1978) was used in combination with the feulgen staining technique for fungal nuclei of germinating resistant sporangia (Olson 1984) with slight modification. Before switching over to embryo count method the seeds were plated overnight (14-16 hrs) at 26°C on moistened blotting paper to soften the tissue and possibly to activate the mycelium inside the seed. The Seeds were then fixed for six-seven hours in Schaudinn's/2 fixative. To demonstrate seed transmission of downy mildew the procedure of Shetty et al. 1977, was used, which is reported to be very conducive and critical in triggering off infection. 2000 seeds from the same infeeted lot were sown in pots containing a moist

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mixture of peat-soil and sand, and held in an environment controlled room at 23-25°C and submitted to 12/12 hr alternate cycles of darkness and artificial day light (Philips TLF 40W/34). After sowing each pot was covered with plastic bags for two weeks to provide high humidity. Before sowing seeds were treated with 2% sodium hypochloride solution for 10 minutes, followed by 5 minutes treatment in 0.2% mercuric chloride solution and finally washed in tap water for 2-3 minutes (Shetty et al. 1977), though no oospores were detected from the seeds in washing test.

About 150 seeds (15%) were carrying seed borne mycelium of *S. graminicola*. The identity of the fungus was confirmed under the compound microscope. The characleristic net like, coenocytic, bluish to light violet mycelium was found in scutellum, membrane covering scutellum, endosperm, aleurone layer, pericarp and plumular bud region (Fig. 1 to 4). Though many workers have reported the presence of mycelium with in the pearl millet seed, no body could detect mycelium in the plumular bud region.

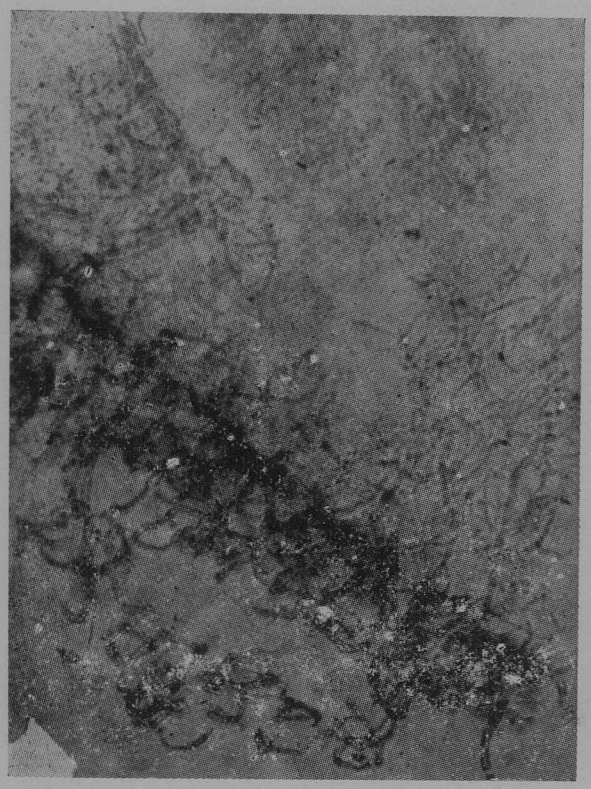

Figure 1. Scutellar region of pearl millet seed showing *Sclerospora graminicola* mycelium (x200)

In all the preparations of the embryos of infected seeds in feulgen staining technique, examined under phase contrast microscope, the host embryonal nuclei took the red colour stain in basic fuchsin showing their viability. In contrast, the nuclei of internal seed borne fungal mycelium were not stained suggesting the probability of presence of nonviable or empty mycelium (Fig. 3-4).

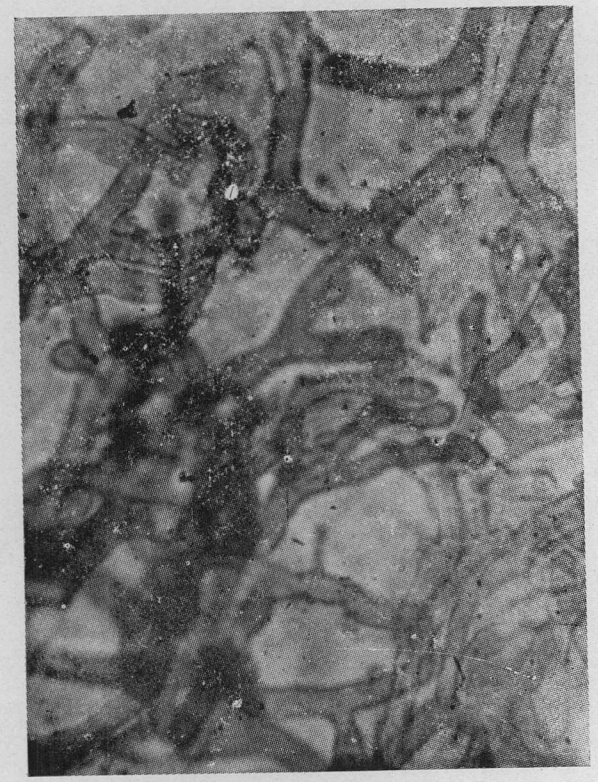

Fig. 2. Net like mycelium of *sclerospora graminicola* (x750)

In seed transmission study no downy mildew was seen in any of the 2000 plants grown in environmental controlled room, observed upto 45 days. To further confirm leaves of 20 pale looking plants were cut in to 2-3 cm pieces and put on wet blotter paperlpetri dishes with the abaxia in side up and incubated in darkness for 5-6 hrs. No sporangiophores or sporangia were noticed under the compound microscope. Though, Shetty et al. (1980), however, observed three out of 998 plants infected after 37, 38 and 43 days of sowing in HB3 and one plant out of 1242 plants in NHB_3 after 20 days of sowing in environmental controlled room under similar conditions. Suryanarayana (1962) opined that mycelium in the seed tissues was nonviable but Shetty et al. (1977) believed that resting mycelium in the embryo is capable of inducing downy mildew infection. Williams (1979) also stated that Sclerospora type mycelium was not viable

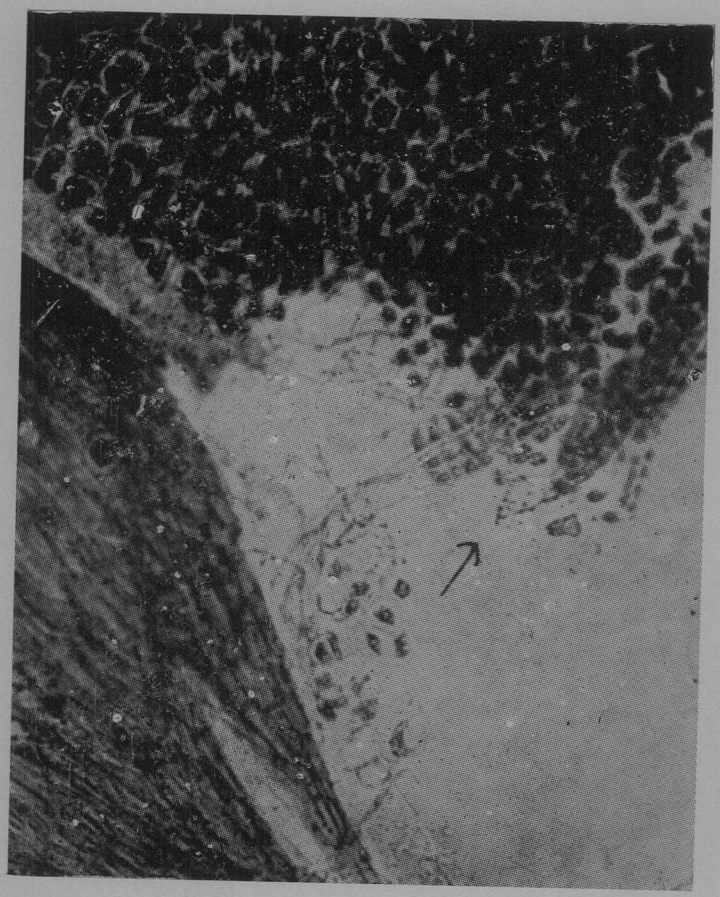

Figure 3. Plumular bud region with stained host embryonal nuclei and mycelium(x200)

in mature, dry (10-5.6% moisture), thoroughly surface sterilized pearl millet seed since in separate experiments conducted at Maryland (USA) and Kew (England) no diseased plants were obtained from the seeds carrying downy mildew mycelium.

In the present study no viable mycelium of *S. graminicola* was found inside the pearl millet seed, therefore, there is very remote possibility of transmission of this disease through seed borne mycelium in Western Rajasthan as higher temperature during harvest and storage leave very low moisture content in the seed which in turn may make such mycelium nonviable.

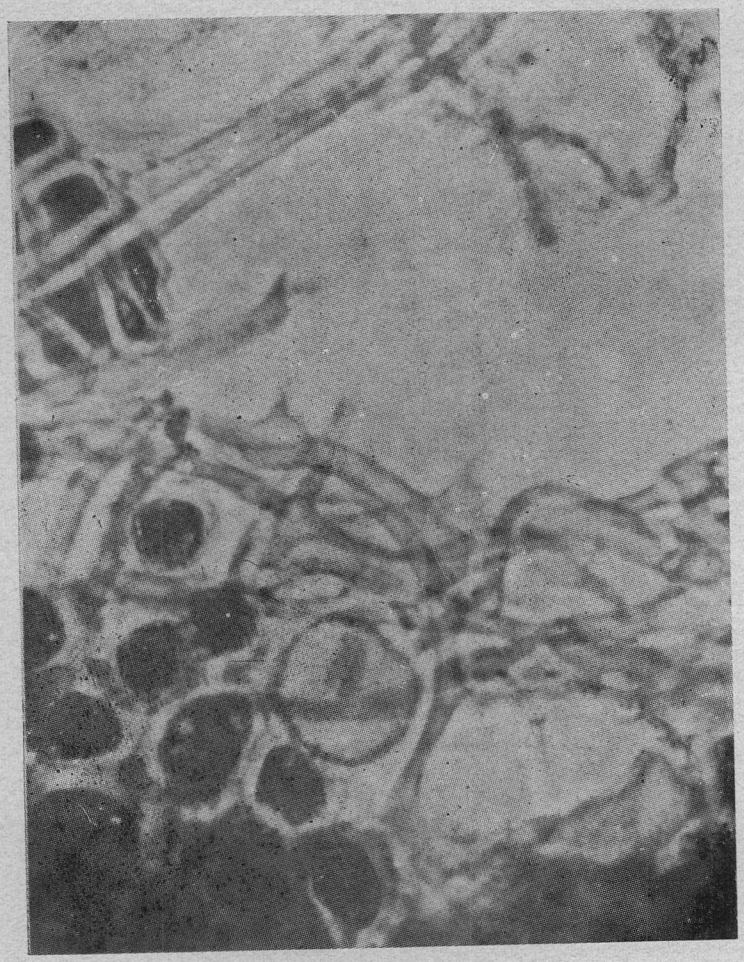

Fig. 4. Plumular but region with stained host embryonal nuclei and mycalieim (x750)

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