

***Botrytis* STALK AND HEAD ROT OF SUNFLOWER IN PAKISTAN**

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SUMMARY

Botrytis stalk and head rot was observed on three sunflower inbred lines under greenhouse and field conditions at NARC, Islamabad. The disease was characterized by rotting of the stalk and head with profuse production of conidiophores and conidia on grey to dark brown lesions. Based on the morphological and cultural characteristics, the causal organism was identified as *Botrytis cinerea* Pers. ex. Fr., which appears to be a new record on sunflower from Pakistan. The incidence of stalk rot was 25 and 30 percent in inbred lines HA-89 and R HA-296, respectively and 30 percent in CMS HA-89 line for head rot under greenhouse while in traces under field conditions.

Key words: Sunflower, *Botrytis cinerea*, determining pathogenicity.

INTRODUCTION

Stalk and head rot of sunflower (*Helianthus annuus* L.) caused by *Botrytis cinerea* Pers. ex. Fr. has been reported under natural conditions from Canada (Sackston, 1960), Egypt (Tarabein and Michail, 1978), France (Botton, 1976), Germany (Schicht, 1955), Hungary (Bakos et al., 1968), India (Mathur et al., 1981), Romania (Iliescu et al., 1980) USSR (Novinskii, 1979), Turkey (Ozkutlu, 1976), and the United States (Yakutkin, 1979). The disease is reported to be quite serious in certain parts of Europe. It occurred in epiphytotic form in Hungary during 1965-66 (Bakos et al., 1968). Serious losses in yield of the crop have been reported in the Ukraine region of the Soviet Union by Novinskii (1979). The disease was also reported to occur in Yugoslavia (Aćimović, 1983).

Three exotic sunflower inbred lines, HA-89, CMS HA-89 and HA-296, planted in pots under greenhouse in 1986 and under field conditions in March, 1989 at the National Agricultural Research Centre (NARC), Islamabad, were found infected by a disease of unknown etiology characterized by the rotting of stalk and head of sunflower. Isolations from the infected parts of the plants consistently yielded the fungus *Botrytis cinerea* (Pers.) ex. Fr.

The purpose of this study was to report the occurrence and identification and to fulfil Koch's postulate for determining the pathogenicity of *B. cinerea* on sunflower.

MATERIALS AND METHODS

Isolation and identification

From the naturally infected plants, small pieces (2-3 mm) from the advancing margins of the stalk and head rot lesions were aseptically cut and washed under running water, surface sterilized for 1 minute in 1.0 % solution of sodium hypochlorite, rinsed twice in sterile distilled water (SDW) and plated on Difco potato-dextrose agar (PDA) in

petri-plates. Then, these isolation plates were incubated at 22°C for 7 to 10 days. Six isolates of *B. cinerea* established from different lines were compared for their cultural and morphological characteristics. No variation was noted and thus, a single isolate was used for pathogenicity tests.

Pathogenicity

Pathogenicity tests were conducted on 6 to 7 week old plants of susceptible inbred lines RHA-296 and CMS H-89 grown in pots under greenhouse conditions at NARC, Islamabad. Culture of *B. cinerea* was prepared from a single isolate grown on PDA for 7 to 10 days at the temperature of 22°C.

For inoculation, stalk of highly susceptible inbred line RHA-296 was wounded and cut with a sterile cork-bore 4 mm in diameter. A plug of agar (3 mm) with mycelium and conidia was placed on the cut portion of the stalk and secured with tape. The experiment was repeated three times by inoculating 5 plants in each replication. The same numbers of stalks were also cut and inoculated with sterile PDA plugs and treated as control.

For head inoculation, conidia of *B. cinerea* were harvested from 10 to 15 days old culture grown on PDA. Conidial suspension in SDW was prepared and adjusted to 6×10^6 conidia per ml and then applied with a camel hair brush to 5 flowers of CMS HA-89 inbred line wounded with a sterile needle, 3 days after the onset of flowering. Similarly, 5 heads were also inoculated with SDW to serve as control.

All the plants with inoculated stalks and heads alongwith the control were irrigated and covered with clear polyethylene bags for 48 hours to provide humidity for infection. Then, the plants so treated were kept in the greenhouse at 23°C and examined daily for the development of disease symptoms.

RESULTS AND DISCUSSIONS

Natural symptoms

On the infected stalk the fungus produced symptoms as grey to dark brown lesions, 3 to 10 cm in length and often zonated with profuse production of conidiophores and conidia (Figures 1 and 2). Some plants with stalk rot symptoms were also found wilted. Petiole infection was also observed which resulted in death of individual leaves. Incidence of stalk rot was 25 and 30 percent in inbred lines HA-89 and RHA-296, respectively, and 30 percent in CMS HA-89 for head rot in 1986 under greenhouse while in traces under field condition during March, 1989.

Isolation and identification

B. cinerea was consistently isolated from naturally infected stalks and heads of sunflower inbred lines. Identification was based on the morphological and cultural characteristics of the causal organism as described by Ellis and Waller (1974). Conidia were hyaline, single celled, ellipsoid to ovoid, and measuring 10 to 12 x 6-10 μm . Black sclerotia of irregular shape were also developed near the margin of petri-plates within 20 to 25 days at 22°C.

Pathogenicity

First on the stalk and head of the inoculated plants, soft watery rot lesions were observed which later on turned dark brown and within 10 to 15 days after inoculations light brown or cream coloured mycelial growth with abundant sporulation developed on

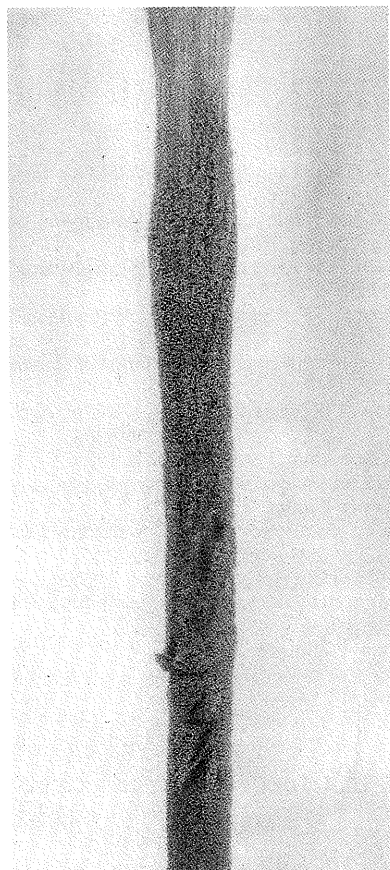


Fig.1 Profuse production of conidia of *Botrytis cinerea* on sunflower stalk.

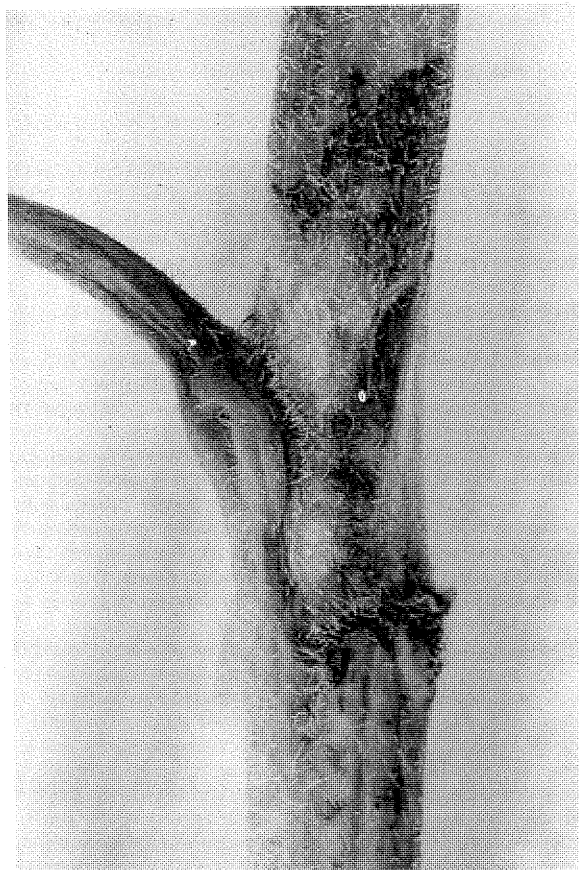


Fig.2 Grey to brown lesions of *B. cinerea* on sunflower stalk and leaf petiole.

the lesions similar to those observed in naturally infected plants. Lesions on the stalk extended up to 3 to 10 cm in both direction from the point of inoculation, with zonated margins, while the control remained symptomless. Reisolation from artificially inoculated plants yielded the fungal isolates morphologically similar to the original isolates.

The stalk and head rot of sunflower was found to be caused by *B. cinerea* Pers. ex. Fr. The occurrence of this pathogen was observed under greenhouse as well as field conditions during March 1986 and 1989. The onset of the disease on the inbred lines of sunflower appeared to coincide with temperature and humidity. It is generally agreed that infection of *B. cinerea* requires a period of high humidity or rain and low to moderate temperature (Baker, 1946 and Jarvis, 1980). The incidence of stalk rot was 25 and 30 percent on HA-89 and RHA-296, respectively, and 30 percent in CMS HA-89 for head rot under greenhouse while in traces under field conditions.

Although the pathogen has been reported from many parts of the world, however, this appears to be a new record of *B. cinerea* on sunflower in Pakistan as it is not included in the Fungi of Pakistan, compiled by Mirza and Qureshi (1978).

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BOTRITIS DE TALLO Y PODREDUMBRE DE CAPITULO DEL GIRASOL EN PAKISTAN

RESUMEN

La botritis de tallo y podredumbre de capítulo de tres líneas puras en condiciones de invernadero y campo fueron observadas en NARC Islamabad. La enfermedad se caracterizó por lesiones con pudrimientos en base a características morfológicas y culturales el organismo causal fué identificado como *Botrytis cinerea* pers. ex. fr. que fué identificado por primera vez en Pakistan. La incidencia de la podredumbre de tallo fué 25 y 30% en líneas puras HA-89, RHA-296 respectivamente y 30% en la línea HA-89 para podredumbre de capítulo en invernadero y solo trasae on condiciones de campo.

X ATTAQUES DE BOTRYTIS SUR TIGE ET CAPITULE DU TOURNESOL AU PAKISTAN

RESUMÉ

La pourriture de la tige et du capitule due au botrytis a été observée sur trois lignées de tournesol en serre et en conditions de champ, au NARC d'Islamabad. La maladie a été caractérisée par la pourriture de la tige et du capitule avec une production importante de conidiophores et de conidies sur des lésions grises à brun foncé. Compte tenu des caractéristiques morphologiques et culturelles, l'organisme incriminé a été identifié comme *Botrytis cinerea* Pers. ex. Fr., ce qui apparait comme une nouveauté chez le tournesol au Pakistan. L'incidence de la pourriture de tige a été de 25 et 30% sur la lignées HA89 et RH296, respectivement, et celle de la pourriture de capitule, de 30% sur la lignée Cms HA89 en conditions de serre avec des traces en conditions de plein champ.