



Resistance in spring wheat to the various diseases caused by *Cochliobolus sativus*  
by Aftabuddin Ahmed

A thesis submitted in partial fulfillment of the requirements for the degree of Doctor of Philosophy in  
Plant Pathology  
Montana State University  
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**Abstract:**

Sixteen spring wheat cultivars were tested for their resistance to the various diseases caused by *Cochliobolus sativus* (Ito and Kurib.) Drechsl. ex Dastur. Five Montana isolates and four Bangladesh isolates of the fungus were used in whole plant inoculations in the Plant Growth Center and in laboratory tests using detached leaves. Sources of resistance were detected and identified for different phases of the disease. Ten cultivars were resistant to root rot, eight cultivars were resistant to foliar spot blotch, and six cultivars were resistant to head blight or black point. A number of cultivars showed differential reaction to various phases of the disease, e.g., were resistant to root rot but were susceptible to foliar spot blotch. Six cultivars, namely Marberg, GP248, GP253, GP254 and GP255, were resistant to all phases of the disease.

The isolates tested differed significantly in pathogenicity, but considerable shifting in ranking occurred between experiments. Isolates obtained from roots were able to attack foliage/heads and vice versa. The isolates from Bangladesh did not have a higher temperature requirement than the USA isolates. Some cultivars were resistant to all isolates from both Bangladesh and the USA.

The maximum disease development for root rot, foliar and head blight/black point phases occurred at 30°C with a 72 hour exposure to moist conditions. The disease reactions on detached leaves were not consistent with those on intact leaves.

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CAUSED BY COCHLILOBOLUS SATIVUS

by

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This thesis has been read by each member of the thesis committee and has been found to be satisfactory regarding content, English usage, format, citations, bibliographic style, and consistency, and is ready for submission to the College of Graduate Studies.

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## ABSTRACT

Sixteen spring wheat cultivars were tested for their resistance to the various diseases caused by Cochliobolus sativus (Ito and Kurib.) Drechsl. ex Dastur. Five Montana isolates and four Bangladesh isolates of the fungus were used in whole plant inoculations in the Plant Growth Center and in laboratory tests using detached leaves. Sources of resistance were detected and identified for different phases of the disease. Ten cultivars were resistant to root rot, eight cultivars were resistant to foliar spot blotch, and six cultivars were resistant to head blight or black point. A number of cultivars showed differential reaction to various phases of the disease, e.g., were resistant to root rot but were susceptible to foliar spot blotch. Six cultivars, namely Marberg, GP248, GP253, GP254 and GP255, were resistant to all phases of the disease.

The isolates tested differed significantly in pathogenicity, but considerable shifting in ranking occurred between experiments. Isolates obtained from roots were able to attack foliage/heads and vice versa. The isolates from Bangladesh did not have a higher temperature requirement than the USA isolates. Some cultivars were resistant to all isolates from both Bangladesh and the USA.

The maximum disease development for root rot, foliar and head blight/black point phases occurred at 30°C with a 72 hour exposure to moist conditions. The disease reactions on detached leaves were not consistent with those on intact leaves.

## INTRODUCTION

Cochliobolus sativus (Ito and Kurib.) Drechsl. ex Dastur, anamorph Bipolaris sorokiniana (Sacc. in Sorok.) Shoemaker [Syn. Helminthosporium sativum (Pamm., King and Bakke)] is one of the most aggressive and virulent pathogens of wheat and barley worldwide, especially in tropical climates. It is known to cause seedling blight, common root rot, spot blotch, head blight and/or black point. These phases of the disease together are responsible for a steady annual loss in crop yield.

One of the most common diseases of dryland spring wheat in central North America is common root rot. On the Canadian prairies, common root rot caused an estimated annual yield loss of about 5.7% for the years 1969-1971. Although immune or fully resistant cultivars to common root rot are not known, marked differences have been found in the resistance of wheat cultivars. Lesions on the subcrown internodes have been correlated with yield reduction.

Severe epidemics of spot blotch on wheat are frequently reported in some tropical countries, including India and neighboring Bangladesh and Thailand. It is also considered one of the important diseases of small grains in Africa. In the United States, on the other hand, spot blotch is considered of secondary importance, although its generalized occurrence was recorded in Minnesota in 1979 and 1980. In Latin America the importance of spot blotch is restricted to Brazil, Paraguay and certain areas of Bolivia.

Spot blotch is considered an extremely destructive disease in years with warm and wet conditions during the growing season. Temperature and moisture appear to be the most important factors that influence the severity of the disease in wheat. Resistance to spot blotch has been described as being ineffective when there are long post-inoculation moist periods. The effects of temperature on growth, sporulation and germination of C. sativus, as well as on penetration of wheat leaves and disease development, have been described. However, the combined effect of post-inoculation moist period and temperature on the effectiveness of spot blotch resistance in spring wheat has not been reported.

Seedling blight, spot blotch and head blight and/or black point have become widespread in many tropical countries. Spot blotch has been managed by use of cultivars with moderate resistance but still greater resistance is needed and should be identified. There is the possibility that a correlation exists between resistance to a specific phase of the disease with resistance to other phases. However, no work has so far been reported on such a possibility.

For many diseases, inoculation of seedlings is a useful method to assess resistance. However, for common root rot, an index based on the reaction of mature plants grown in the field has proven to be consistent over several locations and years, whereas seedling reaction has not been indicative of the adult plant reaction. It appears, therefore, that a careful study of the relationship between seedling and adult plant reaction is warranted.

This study was undertaken with the following objectives:

- 1) To evaluate spring wheat cultivars as a source of resistance to the various phases of the disease caused by Cochliobolus sativus.
- 2) To evaluate the influence of post-inoculation moist period and temperature on disease reaction.
- 3) To determine whether resistance to a specific phase of the disease is correlated with the resistance to any of the other phases of the disease.
- 4) To determine whether resistance to the various phases of this disease is effective against isolates of C. sativus from the United States and Bangladesh.

## REVIEW OF LITERATURE

The Disease

Common root rot (CRR) of spring wheat (Triticum aestivum L.) and barley (Hordeum vulgare L.) caused principally by Cochliobolus sativus is a widespread and important disease (Ledingham et al. 1973; Piening et al. 1976; Sallans 1965). The pathogen can infect any of the below-ground parts of the plant, roots, crown, or stem. Infections occur as discrete lesions, although these may enlarge or coalesce (Huang and Tinline 1976). This disease has been recognized for many years on small grain cereals (Bolley 1909, 1913; Dickson 1956; Henry 1924; Simmonds 1941; Stakman 1920). It has also been known by many names such as "foot rot," "root rot," "black foot," "black stem," "white heads" and "dryland root rot" (Stevens 1919). Common root rot is found in various cereal growing areas of the world (Abbott 1931; Butler 1961; Dastur 1942; Ito and Kuribayashi 1931; McDonald 1930; McRae 1924; Pittman 1929; Sprague 1944). The voluminous available literature on the disease bears convincing testimony about its importance (Bolley 1913; Butler 1961; Christensen 1922; Dodsall 1923; Henry 1924; McKinney 1923; Mitra et al. 1935; Russel 1932; Simmonds 1939, 1941; Stakman 1920; Stevens 1922; Weniger 1932). Several comprehensive reviews have been published over the years on the disease and its pathogen (Butler 1961; Sallans 1965; Simmonds 1939, 1941). Recently, Harding has published a bibliography of more than 2400 research papers about this "prosaic but challenging pathogen" and its disease complex.

The first reports of CRR were made in the United States and Canada (Bolley 1913; Pammel et al. 1910; Simmonds 1939). Bolley (1913) was one of the first to recognize the importance of root diseases, describing the conspicuous necrosis of the crown and crown root tissues of sickly wheat plants, the yellowing and death of leaves and tillers, all of which resulted in reduced yields. Hynes (1938) in Australia described the symptoms of root rot afflicted plants, mentioning their premature ripening and bleaching, reduced growth and severe necrosis of crowns, crown roots and discolored internodes.

Spot blotch is a serious disease of spring wheat in the warm humid tropics (Prescott 1984). Initial leaf infection results from airborne conidia produced on wild grasses or from sporulation on infested residue (Wiese 1977). Rain-splash dispersal of conidia allows infection of the upper leaves and heads (Agrios 1978). Warm temperatures and high humidity increase the severity of infection and maximum host susceptibility occurs after flowering (Khanna and Shukla 1981; Nema and Joshi 1973). Field observations indicate that spot blotch is more severe on short wheat cultivars (Raemaekers 1984) as with infections by Septoria nodorum Berk. (Fried and Bronniman 1982; Scott 1973; Scharen 1964). Flag leaf size may also contribute to a higher disease pressure by permitting greater production of inoculum on large flat leaves (Raemaekers 1984). Though the pathogen can cause root and foot rots, it is the foliar phase which is mainly instrumental in lowering yields (Mitra 1930, 1934). Under suitable environmental conditions spot blotch can reach epidemic proportions and cause yield losses up to 60% (Prabhu and Singh, 1974).

The importance of C. sativus as a head blight pathogen is reflected in the losses to wheat and other cereals through reduction in the quality of the grain due to discoloration of kernels (Greaney and Wallace 1943; Machacek and Greaney 1938). This disease, called black point, is one of the principal reasons for downgrading of irrigated soft white spring wheat in western Canada (Canada Grains Council 1982). It can also be a problem in durum wheat (Machacek and Greaney 1938; Greaney and Wallace 1943; Brentzel 1944; Hanson and Christensen 1953) and in hard red spring wheat (Machacek and Greaney 1938; Greaney and Wallace 1943) in areas receiving heavy rainfall during the early stages of seed development. Black point is a concern because it lowers the quality and interferes with the milling and baking properties of grain (Canada Grains Council 1982). Recently, this disease has become important in different parts of India (Rana and Sengupta 1982).

#### Symptoms

Common root rot symptoms on cereals consist of discoloration and rotting of the lower leaf sheath, crown, culms, subcrown internodes and roots (Butler 1961; Simmonds 1941; Sallans 1965). The first symptom of the disease appears as brown or dark brown lesions on the lower leaf sheaths at about soil level and on subcrown internodes where the lesion may elongate and/or coalesce (Simmonds 1941; Huang and Tinline 1976). Infection may occur at any stage of development of the host plant originating from conidia in soil and residue in the soil and from seed borne conidia (Christensen 1922; Henry 1924; Ledingham et al. 1973; Simmonds 1959). In the soil, conidia remain viable for a long time

(Harding 1972; Simmonds et al. 1950). If infection occurs during the seedling stage, the lower leaves may become wilted and dark lesions can be seen near the soil surface without definite root lesions (Simmonds 1941). Lesions may also be found on the roots, crown and basal stems (Dickson 1956; Simmonds 1941). When severe infection occurs in the seedling stage, many plants may be killed, resulting in seedling blight or post emergence damping off (Stack 1982). Other symptoms of the disease become more distinct after the seedling stage. As the infected plants approach heading time, they may be dwarfed and lighter in color (Simmonds 1941). Occasionally, prematurely ripened plants with bleached heads containing shriveled grains may develop after heading (Sallans and Ledingham 1943; Simmonds 1941; Stack 1982). The symptoms may vary greatly with environment changes (Butler 1961; Simmonds 1941; Sprague 1950).

In the spot blotch phase of the disease a wide range of symptoms from small brown spots to streaks with chlorosis and necrosis are observed on leaves. Neema and Joshi (1971) described the symptoms as follows:

The symptoms of the disease become visible 72 hours after inoculation as water-soaked or chlorotic, more or less oval spots, ranging from pin-prick dots to about 1.5 mm in diameter. The lesions turn reddish brown with diffused margins as they age. Later the spots enlarge parallel to the veins and become more or less elliptical with tapering ends about 2-15 mm long. The lesions coalesce and form large dead patches with a slight yellow halo. Under humid conditions, the blotches usually have brown or red coloration in the center with pronounced chlorosis. Severely infected leaves become shrunken, the tips become discolored and blighted. At this stage, the affected portions of the leaves show the presence of conidiophores and conidia.

Cochliobolus sativus produces distinct, elongated, brown-black lesions that rarely exceed 1 cm in length (Wiese, 1977). The blotchy lesions contrast sharply with green or straw-colored leaf tissues even after leaf senescence. Narain et al. (1973) in India observed that the earliest symptoms appear on the older leaves prior to or about the time of heading. Later there is a blackening of the nodes and internodal areas, the glumes turn greyish brown in color as the sporulation increases, and ultimately most of the spikes become affected. Infected wheat plants ripen prematurely, grain formation is reduced and developed grains are often shriveled.

Black point is characterized by a brown diffuse discoloration of varying intensity, limited to the embryo region of the grain (El-Helaly 1947). It often has a narrow brown line extending along the groove, in some cases extending to the brush end of the kernel. The prominent symptom of black point is the dark brown to blackish discoloration near the embryonic end of the seeds (Adlakha and Joshi 1973; Rana and Sengupta 1982).

#### Economic Importance

Common root rot of wheat has been widely studied as one of the most important diseases in the cereal growing countries (Abbott 1931; Butler 1961; McDonald 1930; McRae 1924; Pittman 1929; Sprague 1944). In 1941 and 1953, Simmonds reviewed the estimates of losses in various regions of the world. The disease accounts for an annual loss in both the quantity and the quality of the crop (Machacek 1943; Wood et al. 1954). Simmonds (1935) studied healthy and diseased plants of wheat and barley

in the field, and found that moderate lesioning of the subcrown internodes resulted in yield losses of 15-20% and severe lesioning resulted in losses of 30-40%. Machacek (1943) developed a formula for yield loss assessment and estimated a 12.1% yield loss for wheat in Manitoba for the years 1939-1941. Ledingham et al. (1973) estimated that 5.7% of wheat was lost annually in the Prairie Provinces of Canada in the years 1969-1971. Tinline and Ledingham (1979) reported a 4.1-8.2% loss in yield of wheat and barley in several field tests depending on varieties. In North Dakota, Stack found losses similar to those of Ledingham and Piening (Stack and McMullen 1979; Stack 1983). The fungus reduces the yield as a result of reduced stands, reduced number of fertile tillers, number of kernels per spike and kernel weight. The symptoms are not always easy to detect in the field and infected plants often are unnoticed (Ledingham et al. 1973; Tinline et al. 1975). Verma et al. (1976) studied the effects of common root rot on components of grain yield in Manitou spring wheat during the years 1969-1971. Plants were divided into severe, moderate, slight and clean categories based mainly on the extent of lesions on the subcrown internodes. The number of tillers per plant, the number and weight of grains per head, the weight per head and 1000-kernel weight in each category were determined. Increasing values of all five components were consistently associated with decreasing disease severity.

#### The Pathogen

Cochliobolus sativus has long been considered a highly variable phytopathogenic fungus. Christensen (1925) reported that the variation

was caused primarily by mutation rather than heterokaryosis. Esser and Kuenen (1967) described the fungus as a monoecious species with a bipolar, homogenic incompatibility reproductive system. To date, the telomorph has not been reported in nature, but its anamorph Bipolaris sorokinianum occurs worldwide. It has a large host range among gramineous species (Sprague 1950). It has also been reported on dicotyledonous crops including beans (Graham et al. 1964; Gourley 1968), alfalfa, red and yellow clovers (Renfro 1963) and buckwheat (Zimmer 1974), but it is considered unimportant on them.

#### Physiological Specialization

It is well known that C. sativus is an extremely variable fungus. Its variability in culture and in pathogenicity has been recognized for many years. Christensen (1922) described four strains that differed in growth rate and morphological characters. He also showed that these isolates varied in pathogenicity when tested on various cereals and grasses. In 1923, Dodsall observed a difference in the degree of disease severity caused by two strains of C. sativus on wheat and barley. Christensen (1925, 1926) used 37 isolates of C. sativus to study their differences in virulence or pathogenicity and their degree of stability. He observed distinct differences between isolates on two cultivars of barley and two cultivars of wheat with respect to cultural characteristics and parasitic capabilities.

Mitra (1931) reported that some isolates tested were less pathogenic while others were more pathogenic than the parental isolates. In another study, Sallans and Tinline (1950) evaluated the interaction of eight

wheat cultivars to 56 isolates. Cultivar differences in common root rot reaction were quite evident and the isolates possessed varying degrees of pathogenicity. However, the cultivars tended to react similarly to the various isolates. Wood et al. (1954) found that C. sativus consisted of many parasitic strains that differed greatly in their parasitic ability, not only on wheat; rye, oats and corn, but also on barley. Ashworth et al. (1960) tested wheat, oats and sorghum for differential susceptibility to single conidium isolates of C. sativus. They classified eleven strains of C. sativus into six pathogenic groups. Wheat and sorghum were very susceptible to some strains and appeared resistant to others. Nelson and Kline (1966) investigated the pathogenicity of 91 morphologically similar isolates of C. sativus to 30 gramineous species and reported a marked variation in pathogenicity on most of the hosts utilized. There was an absence of host specificity among the isolates. Piening (1973) reported differential yield responses of ten barley cultivars to common root rot under naturally infested field conditions. Stack and McMullen (1979) noted differential susceptibility of durum wheats to common root rot. Similarly, variable pathogenicity of C. sativus isolates has been reported on wheat (El-Nashaar and Stack 1982). The first attempts to identify races of C. sativus were made as early as 1922 (Christensen 1925). Christensen (1925) reported that there existed at least 37 races of this fungus based on morphological and pathological characters. Clark and Dickson (1958) reported that isolates of C. sativus differed significantly in pathogenicity on barley. Wood (1962) strongly believed in the existence of physiologic races within C. sativus. In Brazil, Mehta (1981) established a set of differential wheat

cultivars. Based on their reaction pattern, he identified a total of 32 races. He also reported that the reaction pattern of the races on the various cultivars was altered in most cases after the isolates had been stored for eight to ten months.

### Epidemiology

Cochliobolus sativus exists wherever there is cultivation of its primary hosts, wheat and barley. Although published information on the disease over the past 50 years is extensive (Butler 1961, Sallans 1965, Simmonds 1953), further studies are needed to determine more precisely how environmental factors affect the disease intensity. Multiple infections are common and new infections occur throughout the growing season (Tinline 1977; Verma et al. 1974). Secondary infection for common root rot development throughout the growing period has been reported (Stack 1980; Tinline 1977). Once secondary conidia are formed on the above-ground plant parts, they are disseminated by wind, causing lesions on foliar parts.

The severity of the disease, however, is greatly affected by environment (Verma et al. 1974). Disease development is considered to be greatest at warm temperatures or at temperatures unfavorable for the growth of the host. Dosedall (1923) found that mycelial penetration of coleoptile and leaf tissues occurred over a range of 12-34°C and that extensive infection and rapid disease development occurred between 22-30°C. McKinney (1923) studied the effects of soil temperature on root rot on seedlings of barley, spring wheat and winter wheat in the greenhouse and in the field. In greenhouse tests, root rot developed at

all temperatures studied between 8° and 35°C, but infection was much less towards the extremes than near the middle of this range. The optimum temperature for disease development was found to be 28°C with Hanna and Hanchen barleys and Marquis wheat and 32°C for Harvest Queen wheat. Field experiments showed that roots of early sown winter wheat were more severely attacked by C. sativus than late sown winter wheats.

Mitra (1930) studied the effect of temperature on some Indian wheat and barley varieties and indicated that infection was intensified as the temperature was raised from 20 to 30°C. In field studies Sallans (1933) and Greaney (1946) noted that seedling blight was more severe when wheat was sown late in the spring than when sown early. Low temperatures were associated with low disease incidence. In contrast, Hynes (1938) found that certain isolates of C. sativus produced severe disease on wheat seedlings at relatively low temperatures. Clark and Dickson (1958) investigated the influence of temperature on spot blotch, root rot and seedling blight development on six barley varieties. The maximum spot blotch development occurred at 28°C with rapid development at 24°C and considerably less at 20° and 16°C. Root rot and seedling blight development was severe at all temperatures that ranged from 8-28°C, but maximum development occurred at 20°C. Greaney (1946) studied the effect of soil temperature on common root rot ratings of mature spring wheat plants in the field. Different temperature regimes were provided by different dates of sowing. When soil temperatures were high at planting, more disease was observed on the plant.

Cook (1960) reported significant differences under greenhouse conditions in the reactions of barley varieties to C. sativus consistent

with typical field reactions, but the results were reproducible only when temperature, rate of moisture application and incubation period in moist chambers were rigidly controlled. Morton (1962) studied the effects of temperature and humidity on the development of C. sativus on excised barley leaves. Leaf specimens at 30° became more chlorotic and senesced much more rapidly than those held at lower temperatures. Leaf pieces kept in closed tubes developed disease more quickly than those kept in open tubes, thus demonstrating the need for moisture for leaf infection.

Nema and Joshi (1973) studied the effect of temperature and moisture on spot blotch disease development on detached wheat leaves. Infection of detached leaves was quicker and more intense at 28°C than at 20°C or 25°C, but development of lesions was faster at 22°C than at 25°C or 28°C. It was concluded that a higher temperature is necessary for infection than for development of symptoms after infection. A period of 24-48 hours exposure to saturated atmosphere was enough to allow infection and subsequent disease development. Prolonged incubation under high humidity resulted in severe symptoms on the leaves.

McKinney (1923) investigated the effects of soil moisture on common root rot. He noted that as water availability increased, the percentage of diseased plants increased. Coupled with soil temperature, McKinney found that soil moisture influenced the temperature at which maximum disease development occurred. High soil temperature favored more disease at high soil moisture levels than at low soil moisture levels, while low soil temperature favored more disease at low moisture levels but not at high moisture levels.

Raemaekers and Tinline (1981) investigated the epidemic of spot blotch and head blights caused by C. sativus in rainfed summer wheat plots in Zambia and concluded that the abundant moisture with prolonged periods of leaf wetness due to numerous rainfalls and favorable temperatures (19.6-26.4°C) were the important conducive factors for the development of the disease.

Khanna and Shukla (1981) in India found that leaf blight infection occurred at 23-30°C, optimum being at 28°C and relative humidity significantly affected disease development only when the temperature was favorable.

Luz (1982) investigated the effect of moist period on the reaction of wheat cultivars to leaf spot. Five cultivars were inoculated and kept moist for periods up to 54 hours. Leaf spots developed after a minimum moist period of 6 hours. Symptom severity increased with a prolonged moist period.

Stockwell and Sherwood (1982) investigated the effect of temperature on penetration of wheat leaves by C. sativus. They reported that increased temperature resulted in increased frequency of appressoria development, increased frequency of successful penetration and enlarged areas of induced autofluorescence.

Luz and Bergstrom (1986) studied the development of spot blotch in spring wheat differing in resistance. They inoculated wheat plants and maintained them at post-inoculation temperatures ranging from 12-28°C. Generally, the minimum incubation periods for infection were shorter at temperatures higher than 18°C but were similar at temperatures between 20°C and 28°C. Lesions per unit area of leaf tissue increased with

increasing temperature. At 28°C all plants developed severe symptoms regardless of genotype.

Temperatures from 25-30°C were the most favorable for infection and head blight development. At these temperatures, exposure to continued wetness for 12 hours resulted in plant infection, whereas a 36-hour exposure was required for death of the spikelets (Andersen, 1952).

#### Resistance in Spring Wheat

The use of resistant cultivars is probably the most effective method of combating diseases and pests (Russell 1978). Christensen (1926), Greaney et al. (1938), Tyner and Broadfoot (1943) tested many wheat cultivars for resistance, with or without artificial inoculation, in the field. High degrees of resistance were not found, although the cultivars tended to fall into different resistance classifications. Sallans and Tinline (1965) reported that resistance in wheat to common root rot was a heritable character. They made crosses between wheat cultivars which had shown some resistance. The crosses gave rise to both highly resistant and highly susceptible lines that were consistent in their root rot reaction at four or five locations over a period of two years. Srivastava et al. (1971) studied the genetics of resistance of spring wheat to C. sativus in four intravarietal crosses. Resistance in Sharbati Sonora and E4853 was found to be conditioned by two complementary genes which seemed to be distinct from each other. Harding (1974) screened 5500 lines for resistance to common root rot and selected 112 lines. Joshi and Adlakha (1974) screened 430 spring wheat cultivars for resistance to two virulent strains of C. sativus under artificial

conditions and found 24 as resistant or moderately resistant. Srivastava (1982) investigated the mode of inheritance of seedling resistance to C. sativus in six crosses involving six resistant cultivars with a susceptible one. The F<sub>1</sub> plants in all six crosses exhibited a high degree of resistance, indicating that the resistance was dominant. The experiment also showed that the resistance of all six parents was conditioned by two dominant complementary genes. Adlakha et al. (1983, 1984) also reported that resistance in wheat to C. sativus was conditioned by one or two dominant factors. Bailey et al. (1984) screened 38 lines of different Triticum and Aegilops species in the greenhouse and found that 3 lines (2 Aegilops and 1 Triticum) possessed significantly higher resistance than the standard cultivar.

In a 2-year field study conducted at six locations on the Canadian Prairies, Conner and Davidson (1988) identified seven spring wheat cultivars that consistently had low incidence of black point. In separate tests the inoculation of resistant wheat cultivars with Alternaria alternata and C. sativus under controlled environmental conditions revealed significant differences in black point incidence. All cultivars were more resistant to A. alternata than the susceptible cultivar Fielder, but only three cultivars were more resistant to C. sativus than Fielder.

The above reviews clearly indicate that sources of resistance in spring wheat to C. sativus are available for common root rot, spot blotch and head blight phases of the disease. Widespread use of resistance should help reduce the chance of an epidemic and enhance the efficacy of fungicides that might be used in a disease management program.

## MATERIALS AND METHODS

Sources of Germplasm

Nine Canadian spring wheat lines GP248-GP256 received from Dr. R. Loiselle, Agriculture Canada, four Brazilian cultivars (BR4, BR8, CNT8 and BH1146) and three local cultivars (Fortuna, Marberg and Rescue) were included for detailed study of their reaction to different isolates of Cochliobolus sativus. The Canadian lines were developed for resistance to common root rot primarily caused by C. sativus (De Pauw et al. 1984). All the Brazilian lines are resistant to moderately resistant to spot blotch caused by C. sativus (personal communication with Dr. W. C. de Luz). These lines were grown in the greenhouse to produce additional seeds for the experimental studies. The local cultivars were chosen as susceptible checks (Rescue and Fortuna), while Marberg is a well adapted cultivar. Fortuna and Rescue are moderately susceptible and susceptible to root rot caused by C. sativus, respectively, and Marberg is moderately resistant.

Isolation and Maintenance of Cochliobolus sativus

Five Montana isolates of C. sativus (195(1), 214(1), 215(1), 369(1) and 370(1)) and four Bangladesh isolates (A<sub>1</sub>, B<sub>1</sub>, C<sub>1</sub> and D<sub>1</sub>) were collected. The Montana isolates originated from infected subcrown internodes of wheat. The Bangladesh isolates were obtained from leaves

of different wheat cultivars showing spot blotch symptoms collected from various locations in the country.

The diseased leaves were cut into small pieces, disinfected with 0.5% NaOCl for 3 minutes, then rinsed in sterile distilled water, dried and plated on potato dextrose agar (PDA). The Petri dishes were incubated for 2-5 days at room temperature (22-26°C). All cultures showing growth typical of C. sativus were single spored and grown on PDA plates for 10-15 days. The cultures were then transferred to PDA slants and stored at 2-4°C as stock cultures. The latter were maintained as source cultures for inoculum preparation.

#### Preparation of Inoculum

Method #1. Isolates transferred from stock cultures were grown on PDA in Petri plates for 10-15 days at room temperature (22°-24°C). A spore suspension was prepared by scraping the spores from the agar surface with a clean microscope slide into distilled water. The spore suspension was then filtered through several layers of cheese cloth to remove mycelial fragments and lumps of agar. The spore suspension was then standardized at  $10^6$  spores/ml using a hemacytometer. One drop of detergent "Tween 20" was added to each 100 ml of spore suspension.

Method #2. Infested oat kernels were prepared by adding 90 ml of distilled water to 140 g of oat kernels in a one liter canning jar. A 7 cm diameter disk of #4 Whatman filter paper was placed on top of a canning jar metal lid which had a 2 cm diameter hole in the center. The assembly was subsequently held in place with the screw top lid of the

jar. This arrangement allowed gas exchange with the outside environment while limiting outside contamination. The jars with oats were autoclaved at 121°C for 60 minutes. Plugs of mycelia of isolates of C. sativus were added to the cooled oat kernels under aseptic conditions and allowed to grow for 4-5 weeks. The infested oat kernels were spread to air dry and subsequently blended in a Waring blender. For a control, blended autoclaved oat kernels were used.

#### Root/Subcrown Internode Inoculation

Seeds of test lines and cultivars were sown in 3.5 cm X 16.5 cm cone containers with pasteurized "MSU mix" consisting of top soil (Bozeman Silt Loam), sand and peat moss (1:1:1 by volume). One seed per container was planted 8 cm deep to stimulate formation of a long subcrown internode. Each cultivar was replicated three times in a complete randomized design. The plants were grown on a greenhouse bench with a 12 hour light and temperature cycle. High Intensity Discharge metal halide lamps were used to maintain a 12 hour photoperiod. The temperature ranged from 22 ± 3°C during the light to 16 ± 3°C during the dark periods. Plants were watered whenever necessary with 20-20-20 (N:P:K) Peter's solution that also contained a few trace elements. Inoculations of subcrown internodes were done at the 2-3 leaf stage of the plants (about 2 weeks after germination) with a spore suspension (10<sup>6</sup> spores/ml) using a syringe with long blunt needle. Approximately 5 mls of spore suspension were injected on or very close to the subcrown internode. Five to six weeks following inoculation, plants were carefully removed from the containers and the roots were thoroughly washed with running tap water. Disease severity

on each plant was rated according to the extent of lesioning on the subcrown internode using a 1 to 4 scale (Tinline et al. 1975), where 1 = clean, 2 = slight, 3 = moderate and 4 = severe disease. The scale is described below:

1. Healthy: No discoloration of subcrown internode.
2. Slight: Pinpoint lesions on the subcrown internode.
3. Moderate: Extended linear lesions but without going around the entire circumference of the subcrown internode.
4. Severe: Complete discoloration of the circumference involving more than 50% of the length of subcrown internode.

Analysis of variance was performed for disease severity as it was affected by cultivar, treatment, and cultivar by treatment interaction.

In another test, plants of the same 16 spring wheat cultivars were grown in medium sized (35 x 25 x 9 cm) flats. Ten seeds were planted in one 31 cm furrow containing 2.0 gm of oat kernel inoculum of each isolate. The seeds were sown 6 cm deep to encourage long subcrown internodes. This experiment was designed as a randomized complete block with three replications. Ten isolates including a control were used. The flats were kept on the greenhouse bench under the same growth conditions as described above. At the firm dough stage (scale 87, Zadoks et al.) of plant development, the plants were uprooted and the roots washed under running tap water. All plants in each row were individually scored for disease severity on the basis of subcrown internode lesioning. The plants were grouped into healthy, slight, moderate and severe classes. The number of plants in each class was recorded. Using numerical values of 0, 2, 5 and 10 for healthy, slight, moderate and

severe classes, respectively, the disease ratings (DR) were determined. The numerical values are ratios based on past yield losses incurred by the respective classes (Tyner and Broadfoot, 1943).

$$DR(\%) = \frac{[(\text{number of plants in class} \times \text{numerical value})]}{\text{total number of plants} \times \text{maximum class value}}$$

Analysis of variance was performed for disease severity as described earlier.

In a third test seeds were coated with a 5% solution of carboxymethyl cellulose (10 ml 100<sup>-1</sup> seeds) to act as sticker. After the seeds had dried, they were atomized with a spore suspension at a concentration of 10<sup>6</sup> spores/ml until runoff. A portion of the spore suspension was heat sterilized and was used for treating control seeds. Ten seeds of each of the 16 cultivars were planted 6 cm deep in one row in a 35 x 25 x 9 cm flat. Growth, harvest, disease evaluation techniques, and analysis were the same as described above.

#### Foliar Inoculation

Five plants of each of the 16 spring wheat cultivars were grown in 20 cm plastic pots containing pasteurized "MSU mix" under greenhouse conditions at 22°C day/16°C night temperature and 14 hour day length period. The experiment was done in a randomized complete block design with three replications. The plants were watered with 20-20-20 Peter's solution whenever necessary. At the 2-3 leaf stage (growth stage 12-13, Zadoks et al. 1974), plants were inoculated using an atomizer attached to compressed air with a spore suspension (10<sup>6</sup> spores/ml) of each of the three isolates of C. sativus (214(1), A<sub>1</sub> and B<sub>1</sub>). The leaves of the

plants were sprayed until runoff occurred. One drop of "Tween 20" was added for each 100 ml spore suspension to facilitate wetting of the leaf surface. Immediately after inoculation, the plants were moved to a plastic mist chamber and held for 24 hours to maintain leaf wetness. The mist was provided by a cool water atomizer. The temperature inside the mist chamber was  $22 \pm 4^{\circ}\text{C}$ . The plants were then transferred to a growth chamber at room temperature ( $24^{\circ}\text{C}$ ) programmed for 14 hours light and 10 hours darkness with a minimum of 85% relative humidity. Twenty-four fluorescent tubes and 8 incandescent bulbs inside the growth chamber provided light. Disease severity was estimated on inoculated leaves seven days after inoculation, when lesions appeared to have reached maximum size. The severity of the disease was recorded on a 0-5 scale following the method of Large and Dolings (1962):

- 0 = Free from spotting.
- 1 = Necrotic spot without chlorosis, up to 5% of leaf area involved.
- 2 = Necrotic spots with light chlorosis, 6-20% of leaf area involved.
- 3 = Necrotic spots with pronounced chlorosis, 21-40% leaf area involved.
- 4 = 41-60% of the leaf area involved.
- 5 = Spots merging, more than 60% of the leaf area involved.

The reactions under category 0 and 1 were considered as resistant; 2 as moderately resistant; 3 as moderately susceptible; 4 as susceptible; and 5 as highly susceptible.

Analysis of variance was done for disease severity as it was affected by cultivar, isolates and cultivar x isolate interactions.

Screening for Resistance to Black Point

All sixteen spring wheat cultivars were grown in 20 cm plastic pots containing "MSU mix." Seven seeds were sown per pot. Two weeks after germination, the seedlings were thinned to four per pot. Each pot with each cultivar was replicated three times.

A spore suspension was prepared from the single spored isolates 214(1), A<sub>1</sub> and B<sub>1</sub> of C. sativus as described previously. Inoculations were carried out at anthesis using the vacuum infiltration method as described by Conner and Thomas (1985). The vacuum infiltration apparatus was constructed in the laboratory by using the diagram prepared by Moore (1936). The apparatus consisted of a iron support stand, a flask for inoculum and an inoculating chamber. The iron support stand was provided with clamps to support the inoculum flask and the inoculating chamber. The inoculating chamber was made with a large test tube with both ends open. The lower and larger end was fitted with soft split rubber stopper and was connected through a length of plastic tube and a pinch cock with the inoculum flask. Small-bore plastic tubing was used to connect the top of the inoculating chamber with a vacuum pump. The procedure of inoculation was simple. The inoculum flask was filled with spore suspension ( $10^6$  spores/ml). Wheat heads to be inoculated were pushed inside the inoculating chamber individually and submerged in the spore suspension for 30-40 seconds under vacuum pressure. Following inoculation, the remaining uninoculated tillers were removed and the pots were transferred to a mist chamber and kept for 24 hours at 22°C. The plants were then removed to the greenhouse bench and grown to maturity under

conditions described previously. At maturity the plants were harvested individually and percentage of black point kernels for each pot was calculated. Analysis of variance for percent blackened kernels was calculated and cultivar means were compared using the LSD test.

#### Evaluation of Resistance Using Detached Leaves

##### Generation of Inoculum

Inoculum of nine isolates, five from Montana and four from Bangladesh, was produced on PDA in Petri dishes for 10-15 days at 22°-26°C. A spore suspension with a concentration of  $10^6$  spores/ml was prepared as described previously. One drop of "Tween 20" was added to each 100 ml. of spore suspension.

##### Generation of Leaf Material

Ten seeds of each of the 16 spring wheat cultivars were sown in 20 cm diameter plastic pots containing pasteurized "MSU mix." After emergence, seedlings were thinned to five plants per pot. Six pots per cultivar were planted to provide sufficient leaf material for the experiment. Benzimidazole agar was prepared as described by Benedikz et al. (1981). The 0.5% water agar containing 150  $\mu\text{g}/\text{l}$  of benzimidazole was dispensed into sterile Petri dishes. When the plants were two weeks old, the leaves were harvested and three 5 cm long leaf sections were placed in each Petri dish with the adaxial side up. An automatic pipette dispensing 5  $\mu\text{l}$  of spore suspension was used to inoculate the center of each leaf segment with each of the isolates. Three replicates of inoculated leaves were incubated at 22°, 26° and 30°C for 7 days under

continuous illumination from cool-white fluorescent tubes. Control leaves were inoculated with distilled sterile water. Seven days after inoculation, the leaves were rated for length of lesion (cm), area of necrosis (cm<sup>2</sup>) and percentage of necrosis. Data were subjected to analysis of variance through the SAS ANOVA procedure following a complete randomized block design.

A separate experiment using the same procedures as described above was conducted to evaluate the reaction of mature detached wheat leaves to C. sativus. The plants of each of the 16 wheat cultivars were grown to flowering stage in the greenhouse as described previously. The flag leaves were collected and used in this experiment.

Effect of Temperature and Post-Inoculation  
Moist Period on the Pathogenicity of C. sativus

Sixteen spring wheat cultivars were grown in 20 cm diameter plastic pots containing pasteurized "MSU mix." Five plants of each cultivar were grown in each pot replicated three times. The isolates of C. sativus (214(1), A<sub>1</sub> and B<sub>1</sub>) were used to inoculate wheat plants with a spore suspension of each isolate at a concentration of 10<sup>6</sup> spores/ml. The spore suspension was prepared as described previously. About two weeks after germination, inoculum was placed on or close to subcrown internodes of the seedlings using a syringe with a long blunt needle. At the same seedling stage, the leaves of the plants were also inoculated with a spore suspension using an atomizer attached to compressed air. The inoculated plants were immediately moved to a growth chamber provided with a cool mister and kept for either a 24, 48 or 72 hour moist period

to maintain leaf wetness at temperatures of 22°, 26° and 30°C. The chamber was also programmed for 14 hours light and 10 hours darkness. The light was supplied from 24 fluorescent tubes and 8 incandescent bulbs. The experiment was repeated three times for each moist period at each temperature, providing three replications. The same plants were also inoculated at the flag leaf and heading stages (39 and 83 in Zadoks et al.'s scale) following the procedures mentioned above. The plants were allowed to stay inside the growth chamber for seven days at the given temperature including the moist period. Then the plants were taken out of the growth chamber and moved to regular greenhouse benches as described earlier. The plants were watered whenever required with 20-20-20 (N:P:K) Peter's solution. Disease severity was estimated on inoculated leaves 7 days after inoculation (just after removal from the growth chamber) using the 0-5 scale described previously. The severity of root infection on each plant was rated according to the extent of discoloration on the subcrown internode using the 0-5 scale, where 0 = clean and 5 = severe disease at hard dough stage (87 on Zadoks et al.'s scale). The data were analyzed separately at each temperature following a split-plot design using moisture as main plots and cultivars as subplots.

## RESULTS

Evaluation of GermplasmSubcrown Internode Inoculation

Differences in disease reaction on the subcrown internodes in 16 spring wheat cultivars were observed with three methods of inoculation (Table 1). The disease scores for the cultivars Fortuna, Rescue, BH1146, CNT8, BR4 and BR8 were 2.8, 3.2, 2.8, 2.5, 2.3 and 2.4, respectively, and were susceptible in the syringe inoculation method. All the Canadian lines GP248-GP256 were resistant to moderately resistant (disease rating  $\leq 2.00$ ). Among the local cultivars only Marberg was resistant (1.6). The analysis of variance for disease reaction showed that the effect of cultivars and isolates was highly significant but their interaction was insignificant (Appendix Table 31).

When infested oat kernels were used to infest the growth medium, the disease ratings for Fortuna, Rescue, BH1146, CNT8 and BR8 were 35.7%, 43.4%, 34.7%, 29.7% and 25.7%, respectively. Among the Canadian lines only GP256 showed a comparable disease rating (25.9%). The other cultivars tested had lower disease ratings (17.5-21.7%).

The cultivars responded similarly when tested using infested seeds as inoculum. In this case the disease ratings were higher for each of the cultivars, indicating that this method of inoculation was more effective: the cultivars Fortuna, Rescue, CNT8, BH1146 and BR8 showed disease ratings of 44.8%, 55.4%, 42.4%, 44.8% and 37.0%, respectively. Analyses of variance for disease ratings showed that the effect of

Table 1. Reaction of 16 spring wheat cultivars to subcrown internode inoculation with nine isolates of *Cochliobolus sativus* under greenhouse conditions.<sup>1</sup>

Method of inoculation <sup>2</sup>					
Syringe		Oat kernel		Infested seeds	
Cultivar	DR <sup>3</sup>	Cultivar	DR <sup>4</sup>	Cultivar	DR <sup>4</sup>
Rescue	3.2 a <sup>5</sup>	Rescue	43.4 a	Rescue	55.4 a
BH1146	2.8 ab	Fortuna	35.7 ab	Fortuna	44.8 b
Fortuna	2.8 ab	BH1146	34.7 ab	BH1146	44.8 b
CNT8	2.5 b	CNT8	29.7 b	CNT8	42.4 b
BR8	2.4 b	GP256	25.9 bc	BR8	37.0 c
BR4	2.3 b	BR8	25.7 c	GP256	35.2 c
GP255	2.0 c	BR4	25.2 c	BR4	34.6 c
GP252	2.0 c	GP252	21.7 d	GP248	31.0 d
GP256	1.9 c	GP255	21.5 d	GP252	30.5 d
GP253	1.7 d	GP254	21.2 d	GP255	28.8 e
GP254	1.6 d	GP248	20.5 de	GP254	28.6 e
GP249	1.6 d	GP249	18.8 f	GP249	28.5 e
Marberg	1.6 d	Marberg	18.5 f	GP253	26.9 f
GP248	1.5 e	GP253	18.2 f	GP251	26.8 f
GP250	1.5 e	GP251	17.5 g	GP250	26.4 f
GP251	1.3 f	GP250	17.5 g	Marberg	26.1 f
LSD	0.24		2.3		2.7

<sup>1</sup> Averaged across nine isolates: 195(1), 214(1), 215(1), 369(1), 370(1), A<sub>1</sub>, B<sub>1</sub>, C<sub>1</sub> and D<sub>1</sub>.

<sup>2</sup> Syringe - inoculum placed on or close to subcrown internodes.  
Oat kernel - infected oat kernels placed in rows in growth medium.  
Infested seeds - spores adhered to carboxyl methyl cellulose treated seeds at the rate of 10<sup>6</sup> spores/ml.

<sup>3</sup> Disease reaction on 1-4 scale  
1 = clean: no discoloration on subcrown internodes.  
2 = slight: pinpoint lesion on subcrown internodes.  
3 = moderate: linear lesions on one side of subcrown internodes.  
4 = severe: 50% discolored with lesions on the whole circumference of subcrown internodes.

<sup>4</sup> Disease rating on 0-100%, 0 = clean to 100 = severe.

<sup>5</sup> P = 0.05. Column means not followed by the same letter are significantly different by LSD test.

cultivar, isolate and isolate x cultivar were highly significant in both infested oat kernel and infested seed inoculations (Appendix Tables 32 and 33).

The difference in pathogenicity of isolates of C. sativus tested was also evident (Table 2). Isolates 214(1) and B<sub>1</sub> were most pathogenic in the syringe inoculation method (disease reaction 2.6 and 2.5, respectively) as well as in the infested oat kernel inoculation method (disease ratings 31.6% and 31.4%, respectively). The pathogenicity of isolates differed greatly with the infested seed inoculation method. The mean of disease ratings was the highest for isolate C<sub>1</sub> (41%) followed by D<sub>1</sub> (39.6%), 370(1) and 369(1) with disease ratings of 39.3% and 38.5%, respectively. The disease ratings of other isolates varied from 32.6% to 37.8%.

#### Foliar Inoculation

The same 16 spring wheat cultivars were tested for resistance to foliar infection using three isolates of C. sativus. Substantial differences in resistance among cultivars were observed (Table 3). Fortuna, Rescue, GP249, GP256, CNT8 and BH1146 were susceptible with disease reactions of 2.2, 2.8, 2.3, 2.4, 2.2 and 2.5, respectively. The other 11 cultivars tended to be more resistant with disease reactions ranging from 1.4 to 2.0. The Canadian lines GP249 and GP256 had disease ratings higher than 2.0, indicating susceptibility. However, these two cultivars were resistant to root inoculations (Table 1).

Table 2. Relative pathogenicity of nine isolates of *Cochliobolus sativus* on subcrown internodes of 16 spring wheat cultivars under greenhouse conditions.<sup>1</sup>

Method of inoculation <sup>2</sup>					
Syringe		Oat kernel		Infested seeds	
Isolate <sup>3</sup>	DR <sup>4</sup>	Isolate <sup>3</sup>	DR	Isolate <sup>3</sup>	DR
214(1)	2.6 a	214(1)	31.6 a	C <sub>1</sub>	41.0 a
B <sub>1</sub>	2.5 a	B <sub>1</sub>	31.4 a	D <sub>1</sub>	39.6 b
C <sub>1</sub>	2.2 b	370(1)	29.4 b	370(1)	39.3 b
D <sub>1</sub>	2.1 bc	215(1)	28.6 b	369(1)	38.5 bc
369(1)	2.1 bc	369(1)	27.9 b	215(1)	37.8 bc
370(1)	2.0 c	195(1)	25.2 c	195(1)	36.6 cd
215(1)	2.0 c	C <sub>1</sub>	23.0 d	B <sub>1</sub>	35.8 cde
195(1)	2.0 c	A <sub>1</sub>	23.0 d	214(1)	33.2 e
A <sub>1</sub>	1.8 d	D <sub>1</sub>	21.1 e	A <sub>1</sub>	32.6 e
LSD	0.19		1.9		2.1

<sup>1</sup> Averaged across 16 standard cultivars.

<sup>2</sup> Syringe - inoculum placed on or close to subcrown internodes.  
Oat kernel - infected oat kernels placed in rows in growth medium.  
Infested seeds - spores adhered to CMC treated seeds at the rate of 10<sup>6</sup> spores/ml.

<sup>3</sup> Isolates: 195(1), 214(1), 215(1), 369(1) and 370(1) from Montana; A<sub>1</sub>, B<sub>1</sub>, C<sub>1</sub> and D<sub>1</sub> from Bangladesh.

<sup>4</sup> Disease reaction on 1-4 scale

1 = clean: no discoloration on subcrown internodes.

2 = slight: pinpoint lesion on subcrown internodes.

3 = moderate: linear lesions on one side of subcrown internodes.

4 = severe: 50% discolored with lesions on the whole circumference of subcrown internodes.

<sup>5</sup> P = 0.05, column means not followed by the same letter are significantly different by LSD test.

Table 3. Reaction of 16 spring wheat cultivars to foliar inoculation with three isolates of Cochliobolus sativus under greenhouse conditions.<sup>1</sup>

Cultivars	Disease reaction <sup>2</sup>
Rescue	2.9 a <sup>3</sup>
BH1146	2.4 ab
GP256	2.4 ab
GP249	2.3 bc
CNT8	2.3 bc
Fortuna	2.2 bcd
GP250	2.2 bcd
GP253	2.0 cd
GP252	1.9 cd
GP251	1.9 cd
GP255	1.8 cde
GP254	1.7 def
GP248	1.7 def
BR4	1.6 dfg
Marberg	1.6 dfg
BR8	1.4 h
LSD	0.25

<sup>1</sup> Averaged across three isolates: 214(1), A<sub>1</sub> and B<sub>1</sub>.

<sup>2</sup> Disease reaction on 0-5 scale

0 = free of spots.

1 = necrotic spots without chlorosis, up to 5% leaf area involved.

2 = necrotic area with light chlorosis, 6-20% leaf area involved.

3 = necrotic area with pronounced chlorosis, 21-40% leaf area involved.

4 = 41-60% leaf area involved.

5 = spots merging, more than 60% leaf area involved.

<sup>3</sup> P = 0.05, column means not followed by the same letter are significantly different by LSD test.

The three isolates used in this experiment differed in pathogenicity (Table 4). Isolate 214(1) produced the highest disease reaction (2.9) followed by B<sub>1</sub> (2.8) and A<sub>1</sub> (2.3). The analysis of variance on disease reaction indicated that the effects of cultivar, isolate and isolate x cultivar were highly significant (Appendix Table 34).

Table 4. Relative pathogenicity of three isolates of Cochliobolus sativus on foliar infection of 16 spring wheat cultivars under greenhouse conditions.<sup>1</sup>

Isolate <sup>2</sup>	Disease reaction <sup>3</sup>
214(1)	2.9 a <sup>4</sup>
B <sub>1</sub>	2.8 b
A <sub>1</sub>	2.3 c
LSD	0.13

<sup>1</sup> Averaged across 16 standard cultivars.

<sup>2</sup> Isolates: 214(1) from Montana and A<sub>1</sub> and B<sub>1</sub> from Bangladesh.

<sup>3</sup> Disease reaction on 0-5 scale

0 = free of spots.

1 = necrotic spots without chlorosis, up to 5% leaf area involved.

2 = necrotic area with light chlorosis, 6-20% leaf area involved.

3 = necrotic area with pronounced chlorosis, 21-40% leaf area involved.

4 = 41-60% leaf area involved.

5 = spots merging, more than 60% leaf area involved.

<sup>4</sup> P = 0.05, column means not followed by the same letter are significantly different by LSD test.

Evaluation of Resistance to Black Point

The incidence of black point in seeds of 16 cultivars inoculated with Montana isolate 214(1) and Bangladesh isolates A<sub>1</sub> and B<sub>1</sub> C. sativus was variable (Table 5). The percentage of blackened seeds was rather low for all the cultivars (2.9-17.5%). Even then, the cultivars could be separated according to percent blackened seeds. The highest percentage of blackened seeds was recorded in the local susceptible cultivar Rescue (17.5%) followed by Fortuna (15.5%), BR4 (13.6%), CNT8 (13.5%) and GP256 (13.2%). The percentage of blackened seeds was low in most of the Canadian cultivars/lines except GP256 (13.2%). The other cultivars tested had 4.2-12.6 percent black pointed seeds. C. sativus caused a discoloration that extended well beyond the germ end of the seed. Isolations from surface sterilized blackened seeds from this experiment confirmed that the fungus used for inoculation was responsible for the black point symptoms on the seeds.

The effect of different isolates was also noticeable. The results are presented in Table 6. The Montana isolate B<sub>1</sub> caused the highest percentage of blackened seeds (15%) followed by the Montana isolate 214(1) that caused 14.6% seeds to be black pointed. Even though there was a difference in the incidence of black point caused by isolates 214(1) and B<sub>1</sub>, they could not be separated statistically.

The analysis of variance for percentage of blackened seeds showed significant effect due to cultivar and isolates but there was no effect of cultivar x isolates (Appendix Table 35).

Table 5. Black point incidence in 16 spring wheat cultivars inoculated with three isolates of Cochliobolus sativus under greenhouse conditions.<sup>1</sup>

Cultivars	Percentage of blackened kernels
Rescue	17.5 a <sup>2</sup>
Fortuna	15.5 a
BR4	13.6 ab
CNT8	13.5 ab
GP256	13.2 abc
GP249	12.6 abc
BH1146	10.9 bc
GP251	10.8 bc
GP252	10.5 bc
GP250	9.5 cd
GP248	7.2 cde
GP254	7.1 cde
BR8	6.8 de
Marberg	4.6 e
GP255	4.2 e
GP253	2.9 e
LSD	4.5

<sup>1</sup> Averaged across three isolates: 214(1), A<sub>1</sub> and B<sub>1</sub>.

<sup>2</sup> P = 0.05, column means not followed by the same letter are significantly different by LSD test.

Table 6. Relative pathogenicity of three isolates of Cochliobolus sativus on black point incidence of 16 spring wheat cultivars under greenhouse conditions.

Isolate <sup>2</sup>	Percentage of blackened kernels
B <sub>1</sub>	15.0 a <sup>3</sup>
214(1)	14.6 a
A <sub>1</sub>	10.1 b
LSD	2.2

<sup>1</sup> Averaged across 16 standard cultivars.

<sup>2</sup> Isolates: 214(1) from Montana and A<sub>1</sub> and B<sub>1</sub> from Bangladesh.

<sup>3</sup> P = 0.05, row means not followed by the same letter are significantly different by LSD test.

Evaluation of Resistance Using Detached Leaves

Sixteen spring wheat cultivars were evaluated through detached leaf inoculation. Spot blotch development was measured on the basis of lesion length, necrotic area and percent leaf necrosis at temperatures of 22°, 26° and 30°C. The effect of temperature on lesion length, necrotic area and percent leaf area is shown in Table 7. It is apparent that all three parameters for spot blotch development were highest at 26°C for both detached seedling and mature leaves. An increase in lesion length, necrosis area and percent leaf necrosis was observed when the temperature was raised from 22° to 26°C. At 30°C there was a decrease in all the parameters as compared to 26°C.

Table 7. Effect of temperature on disease reaction of 16 spring wheat cultivars on detached seedling and mature leaves inoculated with nine isolates of Cochliobolus sativus.<sup>1</sup>

Disease reaction	Temperature					
	Seedling leaves			Mature leaves		
	22°C	26°C	30°C	22°C	26°C	30°C
Lesion length (cm)	2.3 c <sup>2</sup>	3.1 a	2.6 b	2.3 b	3.1 a	2.3 b
Necrotic area (cm <sup>2</sup> )	1.4 b	1.7 a	1.4 b	1.9 b	2.4 a	1.9 b
Percent leaf necrosis (%)	44.5 c	62.3 a	51.5 b	38.7 b	54.2 a	36.9 c

<sup>1</sup> Averaged across 16 standard cultivars and nine isolates: 195(1), 214(1), 215(1), 369(1), 370(1), A<sub>1</sub>, B<sub>1</sub>, C<sub>1</sub> and D<sub>1</sub>.

<sup>2</sup> P = 0.05, means in each row not followed by the same letter are significantly different by LSD test.

The length of lesion which developed on detached seedling leaves of 16 spring wheat cultivars varied at different temperatures (Table 8). The cultivar GP252 had the longest lesions (2.6 cm) at 22°C. The shortest lesions were manifested by the cultivars GP254 and Marberg (2.1 cm in each). At 26°C, GP255 had the longest lesions (3.5 cm) while BR8 had the shortest lesions (2.5 cm). The longest lesions were manifested by the cultivar Rescue (3.4 cm) at 30°C with BR8 having the shortest lesions (2.1 cm).

Table 8. Lesion length on detached seedling leaves of 16 spring wheat cultivars inoculated with nine isolates of *Cochliobolus sativus* at three temperatures.<sup>1</sup>

22°C		26°C		30°C	
Cultivar	Lesion length (cm)	Cultivar	Lesion length (cm)	Cultivar	Lesion length (cm)
GP252	2.6 a <sup>2</sup>	GP255	3.5 a	Rescue	3.4 a
GP253	2.5 ab	Rescue	3.4 ab	GP253	3.2 b
Fortuna	2.4 bc	Marberg	3.4 ab	GP252	3.2 b
Rescue	2.4 bc	Fortuna	3.3 bcd	Fortuna	2.9 c
CNT8	2.3 bcd	GP256	3.3 bcd	GP254	2.7 d
BR4	2.3 bcd	GP249	3.2 cde	GP249	2.7 d
BH1146	2.3 bcd	GP252	3.2 cde	GP250	2.7 d
GP255	2.3 bcd	CNT8	3.2 cde	Marberg	2.5 ef
BR8	2.2 def	GP253	3.2 cde	CNT8	2.5 ef
GP250	2.2 def	GP254	3.2 cde	BH1146	2.4 efg
GP251	2.2 def	BR4	3.1 efg	GP248	2.4 efg
GP248	2.2 def	GP248	3.1 efg	GP255	2.4 efg
GP249	2.2 def	GP250	3.1 efg	BR4	2.3 fgh
GP256	2.1 fgh	GP251	2.9 g	GP251	2.2 hi
Marberg	2.1 fgh	BH1146	2.8 h	GP256	2.2 hi
GP254	2.1 fgh	BR8	2.5 i	BR8	2.1 i
LSD	0.12		0.16		0.18

<sup>1</sup> Averaged across nine isolates: 195(1), 214(1), 215(1), 369(1), 370(1), A<sub>1</sub>, B<sub>1</sub>, C<sub>1</sub> and D<sub>1</sub>.

<sup>2</sup> P = 0.05, means in column not followed by the same letter are significantly different by LSD test.

Data on necrotic area produced on different cultivars are presented in Table 9. The necrotic area was the greatest for GP252 (1.6 cm<sup>2</sup>), GP249 (1.8 cm<sup>2</sup>) and GP253 (1.8 cm<sup>2</sup>) at 22°, 26° and 30°C, respectively. In contrast, GP254 (1.2 cm<sup>2</sup>), BR8 (1.5 cm<sup>2</sup>) and GP256 (1.1 cm<sup>2</sup>) had the least necrotic area at temperatures of 22°, 26° and 30°C, respectively.

Table 9. Necrotic area of detached seedling leaves of 16 spring wheat cultivars inoculated with nine isolates of Cochliobolus sativus at three temperatures.<sup>1</sup>

22°C		26°C		30°C	
Cultivar	Necrotic area (cm <sup>2</sup> )	Cultivar	Necrotic area (cm <sup>2</sup> )	Cultivar	Necrotic area (cm <sup>2</sup> )
GP252	1.6 a <sup>2</sup>	GP249	1.8 a	GP253	1.8 a
BR8	1.5 ab	GP255	1.8 a	GP252	1.7 b
BH1146	1.5 bc	GP250	1.8 a	Rescue	1.7 b
CNT8	1.4 bcd	GP252	1.8 a	GP249	1.7 b
GP250	1.4 bcd	Rescue	1.8 a	Fortuna	1.5 c
GP251	1.4 bcd	Marberg	1.7 b	GP250	1.5 c
GP253	1.4 bcd	Fortuna	1.7 b	GP254	1.4 d
BR4	1.4 bcd	GP254	1.7 b	CNT8	1.3 e
Rescue	1.4 bcd	CNT8	1.7 b	BH1146	1.3 e
Fortuna	1.4 bcd	GP256	1.7 b	GP251	1.3 e
GP249	1.3 de	BR4	1.7 b	GP248	1.2 f
GP255	1.3 de	GP253	1.7 b	GP255	1.2 f
GP256	1.3 de	GP248	1.6 c	BR8	1.2 f
Marberg	1.3 de	GP251	1.6 c	Marberg	1.2 f
GP248	1.2 f	BH1146	1.6 c	BR4	1.1 g
GP254	1.2 f	BR8	1.7 c	GP256	1.1 g
LSD	0.09		0.10		0.13

<sup>1</sup> Averaged across nine isolates: 195(1), 214(1), 215(1), 369(1), 370(1), A<sub>1</sub>, B<sub>1</sub>, C<sub>1</sub> and D<sub>1</sub>.

<sup>2</sup> P = 0.05, means in each column not followed by the same letter are significantly different by LSD test.

The cultivars also reacted differently in terms of percent leaf necrosis produced on detached seedling leaves (Table 10). The percent leaf necrosis was the highest for GP252 (50.3%), GP255 (69.6%) and Rescue (67.8%) at 22°, 26° and 30°C, respectively. The cultivars GP254 (40.3%), BR8 (48.2%) and GP256 (42.3%) had the lowest percent leaf necrosis at 22°, 26° and 30°, respectively. In Table 11, the cultivars were ranked in susceptibility from high to low at the three temperatures. The analysis of variance on lesion length, necrotic area and percent leaf necrosis on detached seedling leaves showed that cultivars, isolates, and cultivar x isolate interactions were significantly different at three temperatures (Appendix Tables 36-53).

The responses of the cultivars were also variable for lesion length, necrotic area and percent leaf necrosis when the mature detached leaves (flag leaf) were inoculated (Tables 12, 13, 14). The local cultivar Marberg (3.6 cm), GP256 (5.0 cm) and Fortuna (3.5 cm) produced the longest lesions at 22°, 26° and 30°C, respectively, while GP249 (1.8 cm), GP251 (2.1 cm) and GP255 (1.8 cm) had the shortest lesions at these temperatures, respectively (Table 12).

The necrotic area was the highest for GP252 (2.7 cm<sup>2</sup>), Fortuna (3.1 cm<sup>2</sup>) and Fortuna at 22°, 26° and 30°C, respectively. The cultivar GP249 (1.4 cm<sup>2</sup>), GP251 (1.5 cm<sup>2</sup>) and GP255 (1.4 cm<sup>2</sup>) had the least necrotic area at 22°, 26° and 30°C, respectively (Table 13).

Data in Table 14 shows that the cultivar Rescue (59.1%), Fortuna (69.0%) and Fortuna (67.2%) had the highest percent leaf necrosis, whereas cultivars GP249 (27.8%), GP251 (39.4%) and GP250 (25.7%) had the lowest percent leaf necrosis at 22°, 26° and 30°C, respectively. The

Table 10. Percent necrosis of detached seedling leaves of 16 spring wheat cultivars inoculated with nine isolates of *Cochliobolus sativus* at three temperatures.

22°C		26°C		30°C	
Cultivar	Percent leaf necrosis	Cultivar	Percent leaf necrosis	Cultivar	Percent leaf necrosis
GP252	50.3 a <sup>2</sup>	GP255	69.6 a	Rescue	67.8 a
Rescue	49.4 a	Rescue	69.1 ab	GP253	65.0 a
GP253	48.9 ab	Marberg	66.3 bc	GP252	61.9 b
Fortuna	47.0 bc	Fortuna	65.9 bc	Fortuna	58.1 c
CNT8	46.2 cd	GP253	64.6 cd <sup>1</sup>	GP249	54.0 d
BR4	45.7 cde	GP249	64.3 cd	GP254	53.9 d
GP250	44.6 def	BR4	64.0 cde	Marberg	49.9 e
GP255	43.9 efg	CNT8	64.0 cde	CNT8	49.5 e
BR8	43.8 efg	GP252	63.2 cde	GP250	49.3 ef
BH1146	43.1 fg	GP256	62.7 de	BH1146	49.0 ef
GP251	42.5 fgh	GP254	61.4 de	GP255	46.7 fg
GP256	42.4 fgh	GP250	60.8 e	GP248	45.6 gh
GP248	41.9 gh	GP248	60.8 e	GP251	44.1 ghi
GP249	41.8 gh	GP251	57.1 f	BR4	43.7 hi
Marberg	40.5 h	BH1146	54.9 f	BR8	42.5 i
GP254	40.3 h	BR8	48.2 g	GP256	42.3 i
LSD	2.30		3.25		2.86

<sup>1</sup> Averaged across nine isolates: 195(1), 214(1), 215(1), 369(1), 370(1), A<sub>1</sub>, B<sub>1</sub>, C<sub>1</sub> and D<sub>1</sub>.

<sup>2</sup> P = 0.05, means in each column not followed by the same letter are significantly different by LSD test.

Table 11. Ranking of 16 spring wheat cultivars from scores obtained for lesion length, necrotic area, and percent leaf necrosis on detached seedling leaves at different temperatures.

Susceptibility to <u>Cochliobolus</u> <u>sativus</u>	Lesion length			Necrotic area			Percent leaf necrosis		
	22°	26°	30°	22°	26°	30°	22°	26°	30°
	High ↑ ↓ Low	GP252 GP253 Fortuna Rescue CNT8 BR4 BH1146 GP255 BR8 GP250 GP251 GP248 GP249 GP256 Marberg GP254	GP255 Rescue Marberg Fortuna GP256 GP249 GP252 CNT8 GP253 GP254 BR4 GP255 GP251 GP248 GP250 BH1146 BR8	Rescue GP253 GP252 Fortuna GP254 GP249 GP250 Marberg CNT8 BH1146 GP248 GP255 BR4 GP251 GP256 BR8	GP252 BR8 BH1146 CNT8 GP250 GP251 GP253 BR4 Rescue Fortuna GP249 GP255 Marberg GP248 Marberg GP248 GP254	GP249 GP255 GP250 GP252 Rescue Marberg GP250 GP254 CNT8 CNT8 BR4 GP253 GP248 BR8 Marberg GP251 BR4 BR8	GP253 GP252 Rescue GP249 Fortuna GP250 CNT8 BH1146 GP251 GP248 GP255 BR8 Marberg Marberg BR4 GP256	GP252 Rescue GP253 Fortuna CNT8 BR4 GP250 GP255 BH1146 GP251 GP256 GP248 GP248 GP249 Marberg Marberg GP254	GP255 Rescue Marberg Fortuna GP253 GP249 BR4 CNT8 GP252 GP254 GP256 GP254 GP250 GP248 GP251 BH1146 BR8

Table 12. Lesion length of detached mature leaves of 16 spring wheat cultivars inoculated with nine isolates of Cochliobolus sativus at three temperatures.<sup>1</sup>

22°C		26°C		30°C	
Cultivar	Lesion length (cm)	Cultivar	Lesion length (cm)	Cultivar	Lesion length (cm)
Marberg	3.6 a <sup>2</sup>	GP256	5.0 a	Fortuna	3.5 a
Rescue	2.8 ab	BR8	4.7 ab	GP256	3.3 a
Fortuna	2.7 ab	Rescue	3.4 abc	GP252	3.0 ab
GP252	2.6 ab	Fortuna	3.4 abc	Rescue	2.8 abc
CNT8	2.4 b	GP252	3.3 abc	Marberg	2.3 bcd
BR8	2.3 b	BR4	3.3 abc	BR8	2.3 bcd
GP253	2.3 b	Marberg	3.1 abc	CNT8	2.1 cd
BR4	2.1 b	GP255	2.9 bc	GP253	2.1 cd
GP255	2.1 b	GP254	2.8 bc	GP254	2.0 cd
GP256	2.0 b	CNT8	2.7 bc	BH1146	2.0 cd
BH1146	2.0 b	GP253	2.6 bc	GP248	1.9 d
GP248	2.0 b	GP248	2.6 bc	GP251	1.9 d
GP254	1.9 b	GP249	2.6 bc	GP250	1.8 d
GP250	1.8 b	BH1146	2.5 bc	BR4	1.8 d
GP251	1.8 b	GP250	2.4 bc	GP249	1.8 d
GP249	1.8 b	GP251	2.1 c	GP255	1.8 d
LSD	1.2		1.8		0.8

<sup>1</sup> Averaged across nine isolates: 195(1), 214(1), 215(1), 369(1), 370(1), A<sub>1</sub>, B<sub>1</sub>, C<sub>1</sub> and D<sub>1</sub>.

<sup>2</sup> P = 0.05, means in each column not followed by the same letter are significantly different by LSD test.

Table 13. Necrotic area of detached mature leaves of 16 spring wheat cultivars inoculated with nine isolates of Cochliobolus sativus at three temperatures.<sup>1</sup>

22°C		26°C		30°C	
Cultivar	Necrotic area (cm <sup>2</sup> )	Cultivar	Necrotic area (cm <sup>2</sup> )	Cultivar	Necrotic area (cm <sup>2</sup> )
GP252	2.7 a <sup>2</sup>	Fortuna	3.1 a	Fortuna	3.3 a
Fortuna	2.7 a	Rescue	3.0 a	GP252	3.2 a
Rescue	2.4 b	BR4	2.9 b	Rescue	2.9 b
GP255	2.0 c	GP252	2.8 b	BR8	2.1 c
GP253	1.9 cd	Marberg	2.5 c	Marberg	2.1 c
CNT8	1.9 cd	GP255	2.5 c	BH1146	1.7 d
GP256	1.8 cde	BR8	2.3 cd	GP254	1.6 de
GP248	1.8 cde	CNT8	2.3 cd	GP253	1.6 de
BR8	1.7 cdef	GP254	2.3 cd	GP248	1.6 de
GP254	1.7 cdef	GP249	2.3 cd	CNT8	1.5 def
BR4	1.7 cdef	BH1146	2.3 cd	GP256	1.5 def
Marberg	1.7 cdef	GP253	2.3 cd	GP251	1.5 def
BH1146	1.6 efg	GP256	2.1 de	BR4	1.5 def
GP250	1.6 efg	GP248	2.1 de	GP249	1.4 efg
GP251	1.5 gh	GP250	2.0 def	GP250	1.4 efg
GP249	1.4 h	GP251	1.8 ef	GP255	1.4 efg
LSD	0.19		0.17		0.17

<sup>1</sup> Averaged across nine isolates: 195(1), 214(1), 215(1), 369(1), 370(1), A<sub>1</sub>, B<sub>1</sub>, C<sub>1</sub> and D<sub>1</sub>.

<sup>2</sup> P = 0.05, means in each column not followed by the same letter are significantly different by LSD test.

Table 14. Percent necrosis of detached mature leaves of 16 spring wheat cultivars inoculated with nine isolates of Cochliobolus sativus at three temperatures.

22°C		26°C		30°C	
Cultivar	Percent leaf necrosis	Cultivar	Percent leaf necrosis	Cultivar	Percent leaf necrosis
Rescue	59.1 a	Fortuna	69.0 a	Fortuna	67.2 a
Fortuna	51.4 b	Rescue	68.2 ab	GP252	58.2 b
GP252	50.6 b	GP252	66.1 bc	Rescue	54.3 c
CNT8	40.3 c	BR4	63.6 c	Marberg	40.1 d
GP253	40.2 c	Marberg	60.2 d	GP253	37.8 de
GP256	37.4 d	GP255	56.7 e	BR8	36.4 e
GP255	37.4 d	GP254	53.6 f	BH1146	32.9 f
GP254	36.9 de	CNT8	51.6 fg	GP256	32.5 f
BR4	36.1 de	GP253	49.8 h	CNT8	32.4 f
BR8	35.7 def	GP248	49.6 gh	GP254	32.2 f
BH1146	34.9 def	GP256	49.2 gh	GP251	31.5 fg
Marberg	34.6 ef	GP249	49.0 gh	GP248	28.7 gh
GP248	34.2 ef	BH1146	48.3 h	GP255	27.2 hi
GP250	33.9 f	BR8	47.3 hi	GP249	27.0 hi
GP251	29.7 g	GP250	45.3 i	BR4	26.7 hi
GP249	27.8 g	GP251	39.4 j	GP250	25.7 i
LSD	2.7		2.6		2.3

<sup>1</sup> Averaged across nine isolates: 195(1), 214(1), 215(1), 369(1), 370(1), A<sub>1</sub>, B<sub>1</sub>, C<sub>1</sub> and D<sub>1</sub>.

<sup>2</sup> P = 0.05, means in each column not followed by the same letter are significantly different by LSD test.

cultivars were ranked on the basis of lesion length, necrotic area and percent leaf necrosis as shown in Table 15. Analysis of variance on necrotic area and percent leaf necrosis showed that cultivars, isolates and cultivar x isolate interactions were significantly different (Appendix Tables 39 and 40). However, in case of lesion length, the effect of isolates was significantly different but the effect of cultivars and cultivar x isolate interactions was not significantly different (Appendix Table 38).

The overall means of lesion length, necrotic area, percent leaf necrosis of detached seedling leaves were regressed on those of detached mature leaves. Only the relationship between percent leaf necrosis of detached seedling leaves with those of mature detached leaves was found significant ( $r = 0.73$ ).

The effect of isolates was significant on the manifestation of lesion length, necrotic area and percent leaf necrosis. The data on the effect of isolates on detached seedling leaves are shown in Tables 16, 17 and 18 for lesion length, necrotic area and percent leaf necrosis, respectively, for each temperature. The isolate 370(1) caused the longest lesion (3.4 cm) followed by 195(1) with 3.3 cm and the isolate A<sub>1</sub> caused the shortest lesion (0.7 cm) at 22°C. At 26°C isolate 369(1) caused the longest lesion (4.3 cm) and A<sub>1</sub> had the shortest lesion length (2.6 cm). However, at 30°C isolate 215 produced the longest lesions and 214(1) the shortest.

For detached mature leaves, isolates were also significantly different in causing lesion length (Table 19). Isolate 370(1), 195(1) and 215(1) caused the longest lesions of 3.3 cm, 4.9 cm and 3.4 cm at

Table 15. Ranking of 16 spring wheat cultivars from scores obtained for lesion length, necrotic area, and percent leaf necrosis on detached mature leaves at three temperatures.

Susceptibility to <u>Cochliobolus</u> <u>sativus</u>	Lesion length			Necrotic area			Percent leaf necrosis		
	22°	26°	30°	22°	26°	30°	22°	26°	30°
High	Marberg	GP256	Fortuna	GP252	Fortuna	Fortuna	Rescue	Fortuna	Fortuna
↑	Rescue	BR8	GP256	Fortuna	Rescue	GP252	Fortuna	Rescue	GP252
	Fortuna	Rescue	GP252	Rescue	BR4	Rescue	GP252	GP252	Rescue
	GP252	Fortuna	Rescue	GP255	GP252	BR8	CNT8	BR4	Marberg
	CNT8	GP252	Marberg	GP253	Marberg	Marberg	GP253	Marberg	GP253
	BR8	BR4	BR8	CNT8	GP255	BH1146	GP256	GP255	BR8
	GP253	Marberg	CNT8	GP256	BR8	GP254	GP255	GP254	BH1146
	BR4	GP255	GP253	GP248	CNT8	GP253	GP254	CNT8	GP256
	GP255	GP254	GP254	BR8	GP254	GP248	BR4	GP253	CNT8
	GP256	CNT8	BH1146	GP254	GP249	CNT8	BR8	GP248	GP254
	BH1146	GP253	GP248	BR4	GH1146	GP256	BH1146	GP256	GP251
	GP248	GP248	GP251	Marberg	GP253	GP251	Marberg	GP249	GP248
	GP254	GP249	GP250	BH1146	GP256	BR4	GP248	BH1146	GP255
	GP250	BH1146	BR4	GP250	GP248	GP249	GP250	BR8	GP249
	GP251	GP250	GP249	GP251	GP250	GP250	GP251	GP250	BR4
Low	GP249	GP251	GP255	GP249	GP251	GP255	GP249	GP251	GP250

Table 16. Effect of isolates of Cochliobolus sativus on lesion length development on detached seedling leaves of 16 spring wheat cultivars at three temperatures.<sup>1</sup>

22°C		26°C		30°C	
Isolate <sup>2</sup>	Lesion length (cm)	Isolate <sup>1</sup>	Lesion length (cm)	Isolate <sup>1</sup>	Lesion length (cm)
370(1)	3.4 a <sup>3</sup>	369(1)	4.3 a	215(1)	3.9 a
195(1)	3.3 a	195(1)	4.1 b	369(1)	3.4 b
C <sub>1</sub>	3.0 b	215(1)	4.0 b	B <sub>1</sub>	3.2 c
D <sub>1</sub>	3.0 b	370(1)	3.9 c	370(1)	3.2 c
B <sub>1</sub>	3.0 b	214(1)	3.8 c	C <sub>1</sub>	3.2 c
215(1)	2.2 c	D <sub>1</sub>	3.1 d	A <sub>1</sub>	2.9 d
369(1)	2.2 c	B <sub>1</sub>	3.0 e	195(1)	2.6 e
214(1)	2.0 d	C <sub>1</sub>	2.8 f	D <sub>1</sub>	1.9 f
A <sub>1</sub>	0.7 e	A <sub>1</sub>	2.6 g	214(1)	1.9 f
LSD	0.01		0.13		0.15

<sup>1</sup> Averaged across 16 standard cultivars.

<sup>2</sup> Isolates: 195(1), 214(1), 215(1), 369(1) and 370(1) from Montana and A<sub>1</sub>, B<sub>1</sub>, C<sub>1</sub> and D<sub>1</sub> from Bangladesh.

<sup>3</sup> P = 0.05, means in each column not followed by the same letter are significantly different by LSD test.

Table 17. Effect of isolates of *Cochliobolus sativus* on necrotic area development on detached seedling leaves of 16 spring wheat cultivars at three temperatures.<sup>1</sup>

22°C		26°C		30°C	
Isolate <sup>2</sup>	Necrotic area (cm <sup>2</sup> )	Isolate	Necrotic area (cm <sup>2</sup> )	Isolate	Necrotic area (cm <sup>2</sup> )
370(1)	1.9 a <sup>3</sup>	215(1)	2.4 a	215(1)	2.0 a
195(1)	1.9 a	369(1)	2.3 a	B <sub>1</sub>	1.8 b
D <sub>1</sub>	1.9 a	195(1)	2.3 a	369(1)	1.8 b
C <sub>1</sub>	1.8 b	370(1)	2.1 b	C <sub>1</sub>	1.8 b
B <sub>1</sub>	1.6 c	214(1)	2.1 b	370(1)	1.7 c
215(1)	1.6 c	D <sub>1</sub>	1.6 c	A <sub>1</sub>	1.5 d
214(1)	1.4 d	B <sub>1</sub>	1.5 d	195(1)	1.3 e
369(1)	1.4 d	A <sub>1</sub>	1.4 e	D <sub>1</sub>	1.1 f
A <sub>1</sub>	0.3 e	C <sub>1</sub>	1.4 e	214(1)	0.9 g
LSD	0.07		0.07		0.1

<sup>1</sup> Averaged across 16 standard cultivars.

<sup>2</sup> Isolates: 195(1), 214(1), 215(1), 369(1) and 370(1) from Montana and A<sub>1</sub>, B<sub>1</sub>, C<sub>1</sub> and D<sub>1</sub> from Bangladesh.

<sup>3</sup> P = 0.05, means in each column not followed by the same letter are significantly different by LSD test.

Table 18. Effect of isolates of *Cochliobolus sativus* on percent leaf necrosis development on detached seedling leaves of 16 spring wheat cultivars at three temperatures.<sup>1</sup>

22°C		26°C		30°C	
Isolate <sup>2</sup>	Percent leaf necrosis	Isolate	Percent leaf necrosis	Isolate	Percent leaf necrosis
370(1)	67.6 a <sup>3</sup>	369(1)	85.1 a	215(1)	75.8 a
195(1)	66.6 a	195(1)	80.3 b	369(1)	68.8 b
C <sub>1</sub>	61.2 b	215(1)	79.9 bc	B <sub>1</sub>	64.0 c
B <sub>1</sub>	61.1 b	370(1)	77.5 c	C <sub>1</sub>	63.1 c
D <sub>1</sub>	60.1 b	214(1)	73.5 d	370(1)	62.0 c
369(1)	43.0 c	D <sub>1</sub>	61.6 e	A <sub>1</sub>	58.0 d
215(1)	39.0 d	B <sub>1</sub>	58.7 f	195(1)	48.7 e
214(1)	36.2 e	C <sub>1</sub>	54.6 g	214(1)	37.8 f
A <sub>1</sub>	10.2 f	A <sub>1</sub>	51.9 h	D <sub>1</sub>	36.5 f
LSD	1.8		2.5		2.2

<sup>1</sup> Averaged across 16 standard cultivars.

<sup>2</sup> Isolates: 195(1), 214(1), 215(1), 369(1) and 370(1) from Montana and A<sub>1</sub>, B<sub>1</sub>, C<sub>1</sub> and D<sub>1</sub> from Bangladesh.

<sup>3</sup> P = 0.05, means in each column not followed by the same letter are significantly different by LSD test.

Table 19. Effect of isolates of *Cochliobolus sativus* on lesion length development on detached mature leaves of 16 spring wheat cultivars at three temperatures.<sup>1</sup>

22°C		26°C		30°C	
Isolate <sup>2</sup>	Lesion length (cm)	Isolate <sup>1</sup>	Lesion length (cm)	Isolate <sup>1</sup>	Lesion length (cm)
370(1)	3.3 a <sup>3</sup>	195(1)	4.9 a	215(1)	3.4 a
369(1)	3.1 a	214(1)	4.7 a	B <sub>1</sub>	3.2 ab
C <sub>1</sub>	2.8 ab	215(1)	3.9 ab	370(1)	3.1 ab
B <sub>1</sub>	2.8 ab	370(1)	3.8 ab	369(1)	2.9 ab
215(1)	2.7 ab	D <sub>1</sub>	3.2 abc	D <sub>1</sub>	2.7 ab
195(1)	2.6 ab	369(1)	3.1 abc	C <sub>1</sub>	2.5 bc
214(1)	1.9 bc	B <sub>1</sub>	3.1 abc	195(1)	2.0 c
A <sub>1</sub>	1.9 bc	A <sub>1</sub>	2.5 bc	214(1)	1.9 c
D <sub>1</sub>	1.6 c	C <sub>1</sub>	1.9 c	A <sub>1</sub>	1.1 d
LSD	0.9		1.8		0.7

<sup>1</sup> Averaged across 16 standard cultivars.

<sup>2</sup> Isolates: 195(1), 214(1), 215(1), 369(1) and 370(1) from Montana and A<sub>1</sub>, B<sub>1</sub>, C<sub>1</sub> and D<sub>1</sub> from Bangladesh.

<sup>3</sup> P = 0.05, means in each column not followed by the same letter are significantly different by LSD test.

22°, 26° and 30°C, respectively, while isolates D<sub>1</sub> (1.6 cm), C<sub>1</sub> (1.9 cm) and A<sub>1</sub> (1.1 cm) produced the shortest lesions at these temperatures. Isolates also behaved differently for producing necrotic areas on detached leaves. For detached seedling leaves isolate 370(1) produced the largest necrotic area (1.9 cm<sup>2</sup>) at 22°C, while isolate 215(1) was responsible for the largest necrotic areas at 26°C (2.4 cm<sup>2</sup>) and 30°C (2.0 cm<sup>2</sup>), respectively (Table 17).

On mature detached leaves isolate 370(1) produced the largest necrotic area (3.3 cm<sup>2</sup>) at 22°C, while isolate 215(1) produced the largest necrotic area at both 26°C (3.7 cm<sup>2</sup>) and 30°C (3.1 cm<sup>2</sup>). Isolates D<sub>1</sub> (1.1 cm<sup>2</sup>), C<sub>1</sub> (1.2 cm<sup>2</sup>) and A<sub>1</sub> produced the smallest necrotic area at 22°, 26° and 30°C, respectively (Table 20).

The data in Tables 18 and 21 show the effect of isolates on percent leaf necrosis on detached seedling and mature leaves. The highest percent leaf necrosis was caused by isolate 370(1), 369(1) and 215(1) at 22° (67.6%), 26° (85.1%) and 30°C (75.8%), respectively, on detached seedling leaves (Table 18). On the other hand, isolate A<sub>1</sub> produced the lowest percent necrosis both at 22°C (10.2%) and 26° (51.9%) and D<sub>1</sub> at 30°C (36.5%).

On mature detached leaves, 370(1) produced the highest percent leaf necrosis (59.1%) and A<sub>1</sub> the lowest at 22°C. The isolate 215(1) produced the highest percent leaf necrosis at 26° (74.8%) and 30° (63.1%) (Table 21). On the contrary, isolate C<sub>1</sub> and A<sub>1</sub> had lowest percent leaf necrosis at 26° (32.7%) and 30°C (13.3%).

Table 20. Effect of isolates of *Cochliobolus sativus* on necrotic area development on detached mature leaves of 16 spring wheat cultivars at three temperatures.<sup>1</sup>

22°C		26°C		30°C	
Isolate <sup>2</sup>	Necrotic area (cm <sup>2</sup> )	Isolate	Necrotic area (cm <sup>2</sup> )	Isolate	Necrotic area (cm <sup>2</sup> )
370(1)	3.3 a <sup>3</sup>	215(1)	3.7 a	215(1)	3.1 a
C <sub>1</sub>	2.9 b	370(1)	3.3 b	370(1)	2.9 b
195(1)	2.7 c	214(1)	3.3 b	B <sub>1</sub>	2.9 b
B <sub>1</sub>	2.3 d	195(1)	3.1 c	369(1)	2.5 c
369(1)	2.1 e	369(1)	2.8 d	C <sub>1</sub>	2.0 d
215(1)	1.8 f	D <sub>1</sub>	2.6 e	195(1)	1.7 e
A <sub>1</sub>	1.4 g	B <sub>1</sub>	2.3 f	D <sub>1</sub>	1.6 e
214(1)	1.2 h	A <sub>1</sub>	1.8 g	214(1)	1.4 f
D <sub>1</sub>	1.1 h	C <sub>1</sub>	1.2 h	A <sub>1</sub>	0.7 g
LSD	0.15		0.13		0.13

<sup>1</sup> Averaged across 16 standard cultivars.

<sup>2</sup> Isolates: 195(1), 214(1), 215(1), 369(1) and 370(1) from Montana and A<sub>1</sub>, B<sub>1</sub>, C<sub>1</sub> and D<sub>1</sub> from Bangladesh.

<sup>3</sup> P = 0.05, means in each column not followed by the same letter are significantly different by LSD test.

Table 21. Effect of isolates of *Cochliobolus sativus* on percent leaf necrosis development on detached mature leaves of 16 spring wheat cultivars at three temperatures.<sup>1</sup>

22°C		26°C		30°C	
Isolate <sup>2</sup>	Percent leaf necrosis	Isolate	Percent leaf necrosis	Isolate	Percent leaf necrosis
370(1)	59.1 a <sup>3</sup>	215(1)	74.8 a	215(1)	63.1 a
B <sub>1</sub>	54.3 b	370(1)	72.6 b	370(1)	57.0 b
C <sub>1</sub>	54.0 b	214(1)	65.4 c	B <sub>1</sub>	55.0 b
215(1)	51.5 c	D <sub>1</sub>	64.9 cd	369(1)	51.7 c
195(1)	49.8 c	369(1)	63.1 de	C <sub>1</sub>	40.7 d
214(1)	35.3 d	195(1)	61.8 ef	D <sub>1</sub>	31.0 e
369(1)	34.9 d	B <sub>1</sub>	60.5 f	195(1)	30.4 e
D <sub>1</sub>	24.5 e	A <sub>1</sub>	45.9 g	214(1)	27.0 f
A <sub>1</sub>	24.2 e	C <sub>1</sub>	32.7 h	A <sub>1</sub>	13.3 g
LSD	2.1		2.0		2.3

<sup>1</sup> Averaged across 16 standard cultivars.

<sup>2</sup> Isolates: 195(1), 214(1), 215(1), 369(1) and 370(1) from Montana and A<sub>1</sub>, B<sub>1</sub>, C<sub>1</sub> and D<sub>1</sub> from Bangladesh.

<sup>3</sup> P = 0.05, means in each column not followed by the same letter are significantly different by LSD test.

The pathogenicity of the isolates also varied considerably as determined by their ability to attack root and leaves (Tables 2, 4, 16, 17, 18, 19, 20 and 21). Isolate 214(1) was the most pathogenic in causing root rot using the syringe and oat kernel methods of inoculation, but it was less pathogenic using the infested seed inoculation method. On foliar inoculation of seedlings it was also found to be most pathogenic. However, with detached leaf inoculation this isolate caused only moderate infection. Although the isolate 370(1) caused low root infection, it was the most pathogenic on detached leaves. Isolates 195(1) and 215(1) were found to cause slight to moderate root infection whereas these were very pathogenic on detached leaves. Isolate A<sub>1</sub> was the least pathogenic in causing either root or leaf infection. The remainder of the isolates tested were variable in causing root rot and leaf infection.

Effect of Post-Inoculation Temperature and Moisture  
Period on the Pathogenicity of *C. sativus*

The effect of post-inoculation temperature on disease development was significant. The data on root rot, spot blotch at the seedling, flag leaf and heading stages at different temperatures are shown in Table 22. Maximum disease development occurred at 30°C in most stages of plant growth. For root rot and spot blotch phases, less disease developed at 26°C than at either 22° or 30°C. At the flag leaf and heading stages, the disease was temperature dependent, i.e., the higher the temperature, the more severe was the disease.

Table 22. Effect of post-inoculation temperature on the reaction of 16 spring wheat cultivars at different stages of host development.<sup>1</sup>

Temp (C)	Disease reaction <sup>2</sup>			
	Roots	Seedling	Flag leaf	Heading
22°	1.7 a <sup>3</sup>	1.6 a	1.4 b	1.1 b
26°	1.6 b	1.4 b	1.6 a	1.3 a
30°	1.7 a	1.7 a	1.7 a	1.5 a
LSD	0.09	0.15	0.18	0.19

<sup>1</sup> Averaged across 16 standard cultivars, three isolates (214(1), A<sub>1</sub> and B<sub>1</sub>) and three moisture periods (24 hr, 48 hr and 72 hr).

<sup>2</sup> Disease reaction:

Roots - 0-5 scale

- 0 = No discoloration on subcrown internodes.
- 1 = Pinpoint lesions on subcrown internodes.
- 2 = Linear lesions on one side of subcrown internodes.
- 3 = Up to 25% of subcrown internodes discolored with lesions over entire circumference.
- 4 = Up to 50% of subcrown internodes discolored with lesions over entire circumference.
- 5 = More than 50% of subcrown internodes discolored with lesions over entire circumference.

Seedling, flag leaf and heading - 0-5 scale

- 0 = Free from spotting.
- 1 = Necrotic spot without chlorosis, up to 5% leaf area involved.
- 2 = Necrotic spot with light chlorosis, 6-20% leaf area involved.
- 3 = Necrotic spots with pronounced chlorosis, 21-40% leaf area involved.
- 4 = 41-60% leaf area involved.
- 5 = Spots merging, more than 60% leaf area involved.

<sup>3</sup> P = 0.05, means in each column not followed by the same letters are significantly different by LSD test.

The cultivar response to disease development was significantly different at different temperatures (Tables 23, 24, 25 and 26). The reaction of cultivars to root infection is presented in Table 23. The cultivar Rescue had the highest level of root infection (2.8), followed by Fortuna (2.4), CNT8 (2.2), BR8 (2.2) and BH1146 (2.1) at 22°C. The ranking of cultivars was consistent in their disease reaction at 26° and 30°C, but the severity of the disease was less. The Canadian lines GP248-GP256 had comparatively less root infection than other cultivars at all temperatures.

The cultivars also varied in response to foliar infection at the seedling stage at different temperatures (Table 24). Fortuna and Rescue had a disease incidence of 1.8 and 2.5 at 22°C. The susceptible cultivar Rescue had the highest disease reaction at 26°C. Rescue and Fortuna produced a disease reaction of 2.7 and 2.1, respectively, at 30°C. The mean of disease reactions for Fortuna and Rescue were 1.9 and 2.5, respectively, across all temperatures. The Canadian lines had less disease at those temperatures.

The disease reaction of cultivars for the flag leaf stage (Table 25) showed that the cultivar Rescue was susceptible to the disease at all three temperatures (2.3-2.6). Fortuna had a high level of disease at 26° (2.1) and 30°C (2.2). All other cultivars showed some resistance (disease score less than 2.0) at 22° and 26°C. The average disease reactions of Fortuna and Rescue were 2.1 and 2.5, respectively.

The spring wheat cultivars were also temperature sensitive to foliar infection at the heading stage. All cultivars showed an increased level of disease development with an increase in temperature (Table 26). The

Table 23. Reaction of 16 spring wheat cultivars to root inoculation with three isolates of *Cochliobolus sativus* at three temperatures.<sup>1</sup>

Cultivar	Disease reaction <sup>2</sup>			Mean
	22°C	26°C	30°C	
Fortuna	2.4 b <sup>3</sup>	2.1 b	2.1 b	2.2 ab
Marberg	1.6 e	1.4 ef	1.5 h	1.5 bcd
Rescue	2.8 a	2.8 a	2.7 a	2.8 a
GP248	1.4 f	1.5 e	1.5 h	1.5 bcd
GP249	1.6 e	1.2 gh	1.5 h	1.4 cd
GP250	1.2 fg	1.2 gh	1.3 i	1.2 de
GP251	1.1 g	1.0 i	1.2 i	1.1 e
GP252	1.7 c	1.7 d	1.8 de	1.7 abcd
GP253	1.2 fg	1.1 hi	1.2 i	1.2 de
GP254	1.2 fg	1.1 hi	1.5 h	1.3 cd
GP255	1.7 e	1.4 ef	1.6 fgh	1.6 abcd
GP256	1.4 f	1.4 ef	1.6 fgh	1.5 bcd
BR4	1.9 d	1.9 c	2.0 bc	1.9 abc
BR8	2.2 c	1.8 cd	1.9 cd	2.0 abc
CNT8	2.2 c	1.9 c	1.7 ef	1.9 abc
BH1146	2.1 cd	1.9 c	1.9 cd	2.0 abc
LSD	0.18	0.17	0.14	0.16

<sup>1</sup> Averaged over three isolates (214(1), A<sub>1</sub> and B<sub>1</sub>) and three moisture periods (24 hr, 48 hr and 72 hr).

<sup>2</sup> Disease reaction, 0-5 scale

- 0 = No discoloration on subcrown internodes.
- 1 = Pinpoint lesions on subcrown internodes.
- 2 = Linear lesions on one side of subcrown internodes.
- 3 = Up to 25% of subcrown internodes discolored with lesions on whole circumference.
- 4 = Up to 50% of subcrown internodes discolored with lesions on whole circumference.
- 5 = More than 50% of subcrown internodes discolored with lesions on whole circumference.

<sup>3</sup> P = 0.05, means in each column not followed by the same letters are significantly different by LSD test.

Table 24. Reaction of 16 spring wheat cultivars to foliar inoculation with three isolates of *Cochliobolus sativus* at seedling stage at three temperatures.<sup>1</sup>

Cultivar	Disease reaction <sup>2</sup>			Mean
	22°C	26°C	30°C	
Fortuna	1.8 b <sup>3</sup>	1.7 b	2.1 b	1.9 ab
Marberg	1.4 gh	0.9 f	1.3 gh	1.2 cd
Rescue	2.5 e	2.3 a	2.7 a	2.5 a
GP248	1.6 bcd	1.4 cd	1.6 d	1.5 abc
GP249	1.6 bcd	1.4 cd	1.6 d	1.5 abc
GP250	1.4 cde	1.2 de	1.3 gh	1.3 abc
GP251	1.4 cde	1.1 e	1.5 de	1.3 abc
GP252	1.2 h	1.2 de	1.4 efg	1.3 abc
GP253	1.6 bcd	1.2 de	1.3 gh	1.4 abc
GP254	1.6 bcd	1.4 cd	1.5 de	1.5 abc
GP255	1.3 def	1.7 b	1.8 c	1.6 abc
GP256	1.8 b	1.7 b	1.8 c	1.8 abc
BR4	1.8 b	1.3 de	1.5 de	1.5 abc
BR8	1.2 h	0.9 f	1.2 h	1.1 d
CNT8	1.5 cde	1.6 bc	1.7 c	1.6 abc
BH1146	1.7 bcd	1.4 cd	1.9 c	1.7 abc
LSD	0.22	0.22	0.14	0.22

<sup>1</sup> Averaged over three isolates (214(1), A<sub>1</sub> and B<sub>1</sub>) and three moisture periods (24 hr, 48 hr and 72 hr).

<sup>2</sup> Disease reaction, 0-5 scale

0 = Free from spotting.

1 = Necrotic spot without chlorosis, up to 5% leaf area involved.

2 = Necrotic spot with light chlorosis, 6-20% leaf area involved.

3 = Necrotic spots with pronounced chlorosis, 21-40% leaf area involved.

4 = 41-60% leaf area involved.

5 = Spots merging, more than 60% leaf area involved.

<sup>3</sup> P = 0.05, means in each column not followed by the same letters are significantly different by LSD test.

Table 25. Reaction of 16 spring wheat cultivars to foliar inoculation with three isolates of *Cochliobolus sativus* at flag leaf stage at three temperatures.<sup>1</sup>

Cultivar	Disease reaction <sup>2</sup>			Mean
	22°C	26°C	30°C	
Fortuna	1.9 b <sup>3</sup>	2.1 b	2.2 b	2.1 ab
Marberg	0.7 k	0.9 g	0.8 g	0.8 fg
Rescue	2.3 a	2.5 a	2.6 a	2.5 a
GP248	1.2 hi	1.5 e	1.6 d	1.4 def
GP249	1.8 bc	1.7 de	1.6 d	1.7 cde
GP250	0.8 k	0.6 h	0.8 g	0.7 g
GP251	1.1 ij	1.1 f	1.1 f	1.1 efg
GP252	1.9 b	1.6 e	2.2 b	1.9 abc
GP253	1.3 gh	1.2 f	1.3 e	1.3 ef
GP254	1.6 de	1.7 de	1.7 d	1.7 cde
GP255	1.5 ef	1.9 cd	1.9 c	1.8 bcd
GP256	1.7 cd	1.9 cd	2.0 bc	1.5 abc
BR4	0.7 k	1.6 e	1.5 d	1.3 ef
BR8	1.0 j	1.1 f	0.9 g	1.0 efg
CNT8	1.4 fg	1.7 de	2.0 bc	1.7 cde
BH1146	1.9 b	2.0 bc	1.9 c	1.9 abc
LSD	0.15	0.18	0.16	0.16

<sup>1</sup> Averaged over three isolates (214(1), A<sub>i</sub> and B<sub>i</sub>) and three moisture periods (24 hr, 48 hr and 72 hr).

<sup>2</sup> Disease reaction, 0-5 scale

0 = Free from spotting.

1 = Necrotic spot without chlorosis, up to 5% leaf area involved.

2 = Necrotic spot with light chlorosis, 6-20% leaf area involved.

3 = Necrotic spots with pronounced chlorosis, 21-40% leaf area involved.

4 = 41-60% leaf area involved.

5 = Spots merging, more than 60% leaf area involved.

<sup>3</sup> P = 0.05, means in each column not followed by the same letters are significantly different by LSD test.

Table 26. Reaction of 16 spring wheat cultivars to foliar inoculation with three isolates of *Cochliobolus sativus* at heading stage at three temperatures.<sup>1</sup>

Cultivar	Disease reaction <sup>2</sup>			Mean
	22°C	26°C	30°C	
Fortuna	1.7 b <sup>3</sup>	1.8 b	2.1 b	1.9 ab
Marberg	0.3 g	0.8 gh	1.3 fg	0.8 fg
Rescue	2.2 a	2.4 a	2.5 a	2.4 a
GP248	1.2 cd	1.8 b	1.8 cd	1.6 abc
GP249	1.0 de	1.3 e	1.5 e	1.3 cde
GP250	0.6 f	0.7 h	0.3 i	0.5 g
GP251g	1.2 cd	0.9 g	1.3 fg	1.1 def
GP252	1.8 b	1.3 e	1.2 g	1.4 bcde
GP253	0.5 fg	0.7 h	1.3 fg	0.8 fg
GP254	0.6 f	1.0 fg	1.5 e	1.0 efg
GP255	0.9 e	1.6 cd	1.8 cd	1.4 bcde
GP256	1.4 c	1.6 cd	1.9 cd	1.6 abc
BR4	1.1 de	1.2 ef	1.4 ef	1.2 de
BR8	0.5 f	0.8 gh	0.8 h	0.7 fg
CNT8	1.2 cd	1.6 cd	1.7 d	1.5 abcd
BH1146	1.7 b	1.8 b	1.9 cd	1.8 abc
LSD	0.20	0.21	0.16	0.22

<sup>1</sup> Averaged over three isolates (214(1), A<sub>1</sub> and B<sub>1</sub>) and three moisture periods (24 hr, 48 hr and 72 hr).

<sup>2</sup> Disease reaction, 0-5 scale

0 = Free from spotting.

1 = Necrotic spot without chlorosis, up to 5% leaf area involved.

2 = Necrotic spot with light chlorosis, 6-20% leaf area involved.

3 = Necrotic spots with pronounced chlorosis, 21-40% leaf area involved.

4 = 41-60% leaf area involved.

5 = Spots merging, more than 60% leaf area involved.

<sup>3</sup> P = 0.05, means in each column not followed by the same letters are significantly different by LSD test.

cultivar Rescue was susceptible at all temperatures and Fortuna was susceptible only at 30°C. Other cultivars tested were resistant to moderately resistant at all three temperatures.

Correlation analyses were done for disease reaction between different stages of plant growth at different temperatures. There was a good relationship for disease development between flag leaf and heading stages at 22°C ( $r = 0.76$ ), 26°C ( $r = 0.91$ ) and 30°C ( $r = 0.83$ ) as shown in Figure 1.

The effect of post-inoculation moisture period was substantial on disease reaction at different host development stages (Table 27). The disease severity varied with different moisture periods. The differences were accentuated by extending periods of leaf wetness after inoculation. The disease severity was higher when moisture periods increased from 24 hours to 48 hours and 72 hours at root, seedling, flag leaf and heading stages. Correlations for disease reaction were calculated between root, seedling, flag leaf and heading stages at different moisture period. A high correlation ( $r = .82$ ) was obtained for disease development between flag leaf and heading stages for the 72 hour moisture period. The combined effect of temperature and duration of moisture period at different stages of host development is shown in Table 28. The severity of root infection at 30°C increased with increase in moisture period, but it was inconsistent at 22° and 26°C. However, the severity of foliar infection at seedling and flag leaf stages and infection on heads increased with the increase in temperature and moisture period.

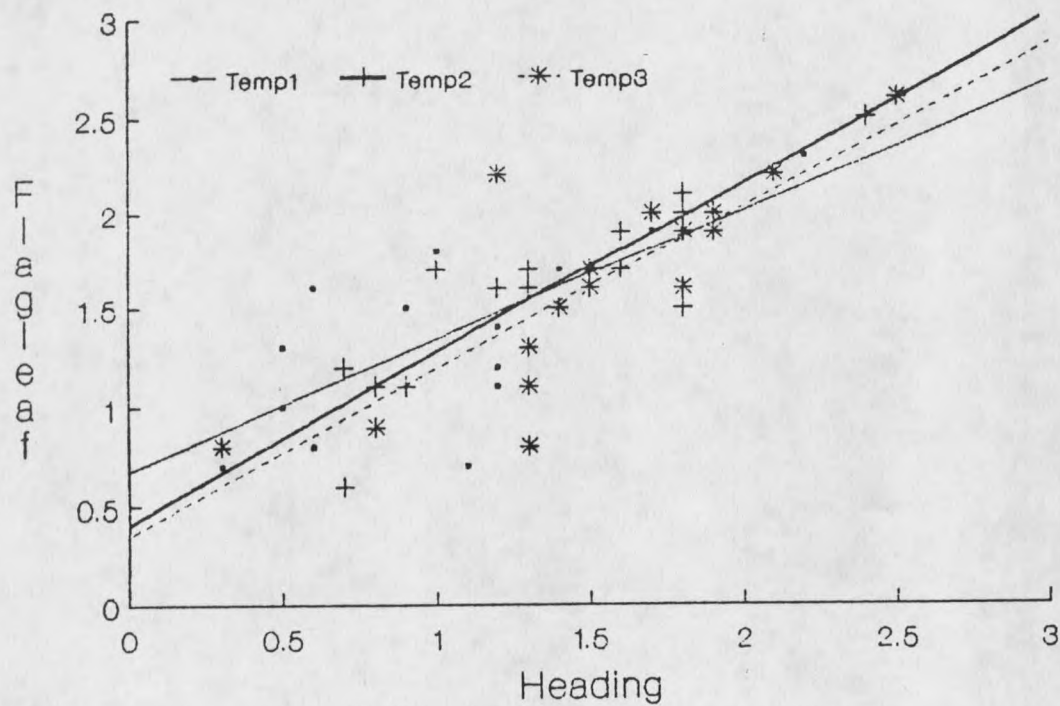


Figure 1. Relationship between flag leaf and heading phases for disease reaction at three temperatures.

Temperature 1 = 22°C ( $r = 0.76$ ).

Temperature 2 = 26°C ( $r = 0.91$ ).

Temperature 3 = 30°C ( $r = 0.83$ ).

Table 27. Effect of post-inoculation moisture on the reaction of 16 spring wheat cultivars inoculated with three isolates of *Cochliobolus sativus* at different stages of host development.<sup>1</sup>

Moisture period	Disease reaction <sup>2</sup>			
	Roots	Seedling	Flag leaf	Heading
24 hour	1.6 b <sup>3</sup>	1.1 c	1.2 c	0.9 c
48 hour	1.7 a	1.6 b	1.5 b	1.4 b
72 hour	1.7 a	1.9 a	1.8 a	1.7 a
LSD	0.09	0.2	0.2	0.2

<sup>1</sup> Averaged across 16 standard cultivars, three isolates (214(1), A<sub>1</sub> and B<sub>1</sub>) and three temperatures (22°, 26° and 30°C).

<sup>2</sup> Disease reaction:

Roots - 0-5 scale

- 0 = No discoloration on subcrown internodes.
- 1 = Pinpoint lesions on subcrown internodes.
- 2 = Linear lesions on one side of subcrown internodes.
- 3 = Up to 25% of subcrown internodes discolored with lesions over entire circumference.
- 4 = Up to 50% of subcrown internodes discolored with lesions over entire circumference.
- 5 = More than 50% of subcrown internodes discolored with lesions over entire circumference.

Seedling, flag leaf and heading - 0-5 scale

- 0 = Free from spotting.
- 1 = Necrotic spot without chlorosis, up to 5% leaf area involved.
- 2 = Necrotic spot with light chlorosis, 6-20% leaf area involved.
- 3 = Necrotic spots with pronounced chlorosis, 21-40% leaf area involved.
- 4 = 41-60% leaf area involved.
- 5 = Spots merging, more than 60% leaf area involved.

<sup>3</sup> P = 0.05, means in each column not followed by the same letters are significantly different by LSD test.

Table 28. Effect of temperature and moisture period on disease reaction of 16 spring wheat cultivars inoculated with three isolates of *Cochliobolus sativus* at different stages of growth.<sup>1</sup>

Growth stages	Disease reaction <sup>2</sup>								
	Temperature								
	22°C			26°C			30°C		
	Moisture period (hr)								
	24	48	72	24	48	72	24	48	72
Roots	1.6 b <sup>3</sup>	1.9 a	1.6 b	1.6 a	1.5 a	1.6 a	1.4 b	1.7 ab	1.9 a
Seedling	0.9 c	1.7 b	2.1 a	1.0 b	1.5 a	1.7 a	1.3 b	1.7 a	1.9 a
Flag leaf	1.0 c	1.4 b	1.9 a	1.3 b	1.5 b	1.9 a	1.4 b	1.7 ab	1.8 a
Heading	0.7 c	1.2 b	1.5 a	1.0 b	1.3 b	1.8 a	1.1 b	1.6 a	1.9 a

<sup>1</sup> Averaged across 16 standard cultivars and three isolates (214(1), A<sub>1</sub> and B<sub>1</sub>).

<sup>2</sup> Disease reaction:

Roots - 0-5 scale

- 0 = No discoloration on subcrown internodes.
- 1 = Pinpoint lesions on subcrown internodes.
- 2 = Linear lesions on one side of subcrown internodes.
- 3 = Up to 25% of subcrown internodes discolored with lesions over entire circumference.
- 4 = Up to 50% of subcrown internodes discolored with lesions over entire circumference.
- 5 = More than 50% of subcrown internodes discolored with lesions over entire circumference.

Seedling, flag leaf and heading - 0-5 scale

- 0 = Free from spotting.
- 1 = Necrotic spot without chlorosis, up to 5% leaf area involved.
- 2 = Necrotic spot with light chlorosis, 6-20% leaf area involved.
- 3 = Necrotic spots with pronounced chlorosis, 21-40% leaf area involved.
- 4 = 41-60% leaf area involved.
- 5 = Spots merging, more than 60% leaf area involved.

<sup>3</sup> P = 0.05, means in each row under each temperature not followed by the same letter are significantly different by the LSD test.

The isolates of C. sativus tested also differed noticeably in their pathogenicity as affected by moisture duration (Table 29). The Bangladeshi isolate B<sub>1</sub> produced the highest disease incidence level of 2.4, 2.2 and 2.2, compared to 2.2, 2.1 and 2.1 for Montana isolate 214(1) at root, seedling and flag leaf stages, respectively. Statistically there was no significant difference in their pathogenicity at heading stage.

The cultivars' response to C. sativus over all temperatures, moisture periods and isolates was variable (Table 30). The cultivar Rescue was the most susceptible and consistent in disease reaction at root, seedling, flag leaf and heading stages. Fortuna showed a disease reaction of 2.2 and 2.0 at root and flag leaf stages. The rest of the cultivars had disease severity levels below 2.0 indicating that these were resistant to moderately resistant to C. sativus.

Table 29. Effect of three isolates of *Cochliobolus sativus* on the reaction of 16 spring wheat cultivars at different stages of host development.<sup>1</sup>

Isolate	Disease reaction <sup>2</sup>			
	Roots	Seedling	Flag leaf	Heading
A <sub>1</sub>	2.0 c <sup>3</sup>	1.9 c	1.9 c	1.6 b
B <sub>1</sub>	2.4 a	2.2 a	2.2 a	1.8 a
214(1)	2.2 b	2.1 b	2.1 b	1.8 a
LSD	0.05	0.06	0.06	0.06

<sup>1</sup> Averaged across 16 standard cultivars, three temperatures (22°, 26° and 30°C) and three moisture periods (24 hr, 48 hr and 72 hr).

<sup>2</sup> Disease reaction:

Roots - 0-5 scale

- 0 = No discoloration on subcrown internodes.
- 1 = Pinpoint lesions on subcrown internodes.
- 2 = Linear lesions on one side of subcrown internodes.
- 3 = Up to 25% of subcrown internodes discolored with lesions over entire circumference.
- 4 = Up to 50% of subcrown internodes discolored with lesions over entire circumference.
- 5 = More than 50% of subcrown internodes discolored with lesions over entire circumference.

Seedling, flag leaf and heading - 0-5 scale

- 0 = Free from spotting.
- 1 = Necrotic spot without chlorosis, up to 5% leaf area involved.
- 2 = Necrotic spot with light chlorosis, 6-20% leaf area involved.
- 3 = Necrotic spots with pronounced chlorosis, 21-40% leaf area involved.
- 4 = 41-60% leaf area involved.
- 5 = Spots merging, more than 60% leaf area involved.

<sup>3</sup> P = 0.05, means in each column not followed by the same letters are significantly different by LSD test.

Table 30. Reaction of 16 spring wheat cultivars to three isolates of Cochliobolus sativus at different stages of host development.<sup>1</sup>

Roots		Seedling		Flag leaf		Heading	
Cultivar	DR <sup>2</sup>	Cultivar	DR <sup>2</sup>	Cultivar	DR <sup>2</sup>	Cultivar	DR <sup>2</sup>
Rescue	2.7 a	Rescue	2.5 a	Rescue	2.5 a	Rescue	2.4 a
Fortuna	2.2 b	Fortuna	1.9 b	Fortuna	2.0 b	Fortuna	1.9 b
BH1146	2.0 c	GP256	1.8 bc	BH1146	2.0 b	BH1146	1.8 b
BR8	1.9 c	BH1146	1.7 cd	GP252	1.9 c	GH256	1.6 c
BR4	1.9 c	CNT8	1.6 de	GP256	1.9 c	GP248	1.6 c
CNT8	1.9 c	GP255	1.6 de	GP255	1.8 d	CNT8	1.5 cd
GP252	1.7 d	GP248	1.6 de	GP249	1.7 de	GP252	1.4 d
GP255	1.5 e	GP249	1.6 de	CNT8	1.7 de	GP255	1.4 d
Marberg	1.5 e	GP254	1.5 e	GP254	1.6 f	GP249	1.3 e
GP256	1.5 e	BR4	1.5 e	GP248	1.5 g	BR4	1.2 e
GP248	1.4 ef	GP253	1.4 f	BR4	1.3 h	GP251	1.2 e
GP249	1.4 ef	GP251	1.3 fg	GP253	1.2 h	GP254	1.0 f
GP254	1.3 g	GP250	1.3 fg	GP251	1.1 i	Marberg	0.8 g
GP250	1.2 g	GP252	1.3 fg	BR8	1.0 i	GP253	0.8 g
GP253	1.2 g	Marberg	1.2 h	Marberg	0.8 j	BR8	0.7 g
GP251	1.1 h	BR8	1.1 h	GP250	0.7 j	GP250	0.5 h
LSD	0.11		0.13		0.12		0.13

<sup>1</sup> Averaged across three temperatures (22°, 26° and 30°C), three moisture periods (24 hr, 48 hr and 72 hr) and three isolates (214(1), A<sub>1</sub> and B<sub>1</sub>).

<sup>2</sup> Disease reaction:

Roots - 0-5 scale

- 0 = No discoloration on subcrown internodes.
- 1 = Pinpoint lesions on subcrown internodes.
- 2 = Linear lesions on one side of subcrown internodes.
- 3 = Up to 25% of subcrown internodes discolored with lesions over entire circumference.
- 4 = Up to 50% of subcrown internodes discolored with lesions over entire circumference.
- 5 = More than 50% of subcrown internodes discolored with lesions over entire circumference.

Seedling, flag leaf and heading - 0-5 scale

- 0 = Free from spotting.
- 1 = Necrotic spot without chlorosis, up to 5% leaf area involved.
- 2 = Necrotic spot with light chlorosis, 6-20% leaf area involved.
- 3 = Necrotic spots with pronounced chlorosis, 21-40% leaf area involved.
- 4 = 41-60% leaf area involved.
- 5 = Spots merging, more than 60% leaf area involved.

<sup>3</sup> P = 0.05, means in each column not followed by the same letters are significantly different by LSD test.

## DISCUSSION

Cochliobolus sativus is a versatile pathogen of wheat and other cereals throughout the world. It is the main causal organism of four important diseases: seedling blight, common root rot/crown rot, foliar spot blotch and head blight/black point. The pathogen is adapted to all environments where wheat grows from high, dry, cold plains to moist, hot, subtropical/tropical lowlands. In tropical climates, it is one of the most aggressive and virulent pathogens attacking all plant parts. The pathogen is widespread and can survive in soil as a saprophyte on crop residue for a long period of time. It may also be seed borne.

Common root rot of spring wheat is an important disease in the northern great plains of North America. Drought and warm temperatures are the most important predisposing factors. Plants under nutritional stress or injured by Hessian fly are also subject to attack (Wiese 1977). Moisture, or at least high relative humidity, is required for root infection but thereafter disease development is highly dependent on warm temperatures and moisture availability. Seedling blight, spot blotch, and head blight are the serious diseases caused by C. sativus in warm, humid climates. Temperature and moisture appear to be the most important factors influencing the severity of these diseases. Prolonged periods of leaf-surface wetness and high temperature favor infection and severity of these diseases.

Cultivars or land races of spring wheat, regardless of their origin, could become useful sources of resistance to these diseases. In this study an attempt was undertaken to understand the reaction of 16 spring wheat cultivars to each separate phase of the disease. Three methods of inoculation for screening cultivars for resistance to common root rot were examined. The infested seed inoculation method was the most effective in inducing disease reaction in the roots and subcrown internodes. It produced higher disease ratings in comparison to the infested oat kernel inoculation method. The use of CMC as a sticker might have helped the inoculum to adhere firmly to the seeds.

The Canadian germplasm lines were consistently resistant to common root rot except GP256, which was resistant to intermediate in reaction. It is not surprising that the Canadian lines were resistant since these lines were developed for resistance to common root rot under Canadian conditions. Among other lines tested, Marberg was also resistant to common root rot. Sources of resistance of wheat and barley to CRR have been reported by many workers (Harding 1972, 1974; Loiselle 1962, 1964; Tyner and Broadfoot 1943). It appears from the results of this study that there are useful sources of resistance in spring wheat to common root rot that can be exploited to develop resistant cultivars. The local cultivar Rescue was consistently the most vulnerable to the disease, followed by Fortuna. Both of these lines are solid stem and provide resistance to the wheat stem sawfly. Researchers have shown that there is a strong correlation between stem solidness and susceptibility to common root rot (McKenzie and Atkinson 1968). The Brazilian lines exhibited susceptibility to common root rot. These lines were reported

to be resistant to moderately resistant to the leaf spot/blotch phase of the disease, but their reaction to CRR had never been evaluated.

Leaf spot/blotch is a serious constraint to wheat production in the warm humid tropics (Prescott, 1984). This disease has been managed by use of cultivars with moderate resistance, but greater resistance is needed (Adlakha et al. 1984). In this investigation, an attempt was made to study the sources of resistance in spring wheat cultivars to the foliar phase of this disease. It was observed that sources of resistance are available in the spring wheat cultivars tested. Two Brazilian lines (BR4 and BR8), six Canadian lines (GP248, GP251, GP252, GP253, GP254 and GP255) and the local cultivar Marberg were resistant. It is interesting to note that these six Canadian cultivars and Marberg were also resistant to root infection. These lines should be a valuable source of resistance in a breeding program to develop resistant and well adapted, high yielding cultivars. Sources of resistance to leaf blotch were also reported by other workers (Joshi et al. 1974; Kararah et al. 1981; Adlakha et al. 1984). Widespread use of resistance should help reduce the chance of an epidemic and enhance the efficacy of fungicides that might be employed in a disease management program.

The detached leaf technique was used to evaluate spring wheat cultivars for resistance to the foliar phase of the disease and was found to be an effective and rapid method for comparing isolates of C. sativus. With this method isolates with different virulence levels can be selected easily for comparison with whole plant inoculations which are time consuming to conduct. The cultivars Marberg, GP248, GP254, GP256, BR4 and BR8 were resistant to detached seedling leaf inoculation, whereas

GP249, GP250, GP251, GP255 and BR4 were resistant to detached mature leaf inoculations. However, GP252 and GP253 were susceptible in detached leaf inoculations even though these were resistant to whole plant inoculations. This indicates that detached leaf reactions do not always reflect reaction of intact leaf inoculations. The utilization of detached leaves was found useful in studying resistance of wheat cultivars to Septoria nodorum (Baker et al. 1978; Benedikz et al. 1981; Karjalainen 1984) and to Pyrenophora tritici-repentis (Krupinsky 1987).

Sources of resistance to foliar infection were also observed in cultivars when screened at three temperatures and moisture periods. Favorable environment was provided to ensure infection. Foliar inoculation at the seedling stage revealed that cultivars Marberg, GP250, GP251, GP252, GP253, GP254, BR4 and BR8 were resistant. All of these cultivars except GP252 and GP254 were also resistant when inoculated at the flag leaf stage.

Sources of resistance of wheat to black point have been reported by many workers (Adlakha and Joshi 1973; Rana and Sengupta 1982; Conner and Thomas 1985; Conner and Davidson 1988; Statler et al. 1975). In this study an attempt was made to screen for reaction of cultivars to black point. The greenhouse tests demonstrated that there were a number of sources of resistance to black point in the spring wheat cultivars tested. Marberg, GP253, GP255 and BR8 were resistant, while Fortuna, Rescue, BR4 and CNT8 were susceptible. The Canadian lines resistant to black point also had resistance against root rot. The vacuum infiltration technique used provided a rapid and reliable means of inoculation and permitted screening of a large number of plants in a short period of

time. Inoculations were done at anthesis and plants were frequently watered. This might have provided enough moisture at milk or mid-dough stages to significantly increase black point incidence in susceptible cultivars. Similar results were reported by Conner (1987). Inoculations under controlled conditions placed cultivars under heavy selection pressure for resistance. The black pointed kernels were highly shriveled in most of the cultivars. This observation is also in agreement with several other reports (Andersen 1952; Huguelet and Kiesling 1973).

The difference in pathogenicity of various isolates in different phases of the disease was significant. In subcrown internode inoculations, the isolates showed inconsistency in virulence with the different methods of inoculation (Table 2). The Montana isolates (from infected roots) as well as the Bangladeshi isolates (from diseased leaves) were equally inconsistent in causing disease reaction. The Bangladeshi isolates were also capable of infecting roots. In subcrown internode inoculation at three temperatures and moisture periods, the Bangladeshi isolate B<sub>1</sub> was the most pathogenic in causing root infection even though it was obtained from infected leaves (Table 28).

In foliar inoculation of whole seedlings, the Montana isolate 214(1) was the most pathogenic in foliar infection. Isolates were found to differ in their ability to cause foliar infection on detached seedling and mature leaves. The Montana isolates were more virulent at 26°C than at 22°C or 30°C on both seedling and mature leaves. The Bangladeshi isolates were inconsistent in affecting lesion length, necrotic area and percent leaf necrosis at these three temperatures.

In foliar inoculations of whole plants at seedling and flag leaf stages across three temperatures and three moist periods, significant differences in pathogenicity were also observed. The Bangladeshi isolate B<sub>1</sub> caused the most severe infection at both stages. The Montana isolate 214(1), though primarily a root rotter, was the second most pathogenic isolate. The Bangladesh isolate B<sub>1</sub> was the most pathogenic in causing black pointed seeds followed by the Montana isolate 214(1). From the results obtained through different experiments it was observed that each isolate tested was able to infect roots, foliage and heads of spring wheat cultivars.

Environment plays an important role in disease development. The effect of post-inoculation temperature and/or moist period on infection and disease development has been studied in several host-parasite relationships (Alderman et al. 1983; Allen et al. 1983; Luz and Bergstrom 1986). The effect of temperature was studied on the reaction of 16 spring wheat cultivars using the detached leaf inoculation technique. Temperature played an important role in disease reaction of these cultivars on detached leaves. Lesions were longer, necrotic area was larger, and percent leaf necrosis was more at 26°C than at 22°C or 30°C. Nema and Joshi (1973) also observed on detached leaves similar increase in lesion size at 22°, 25° and 28°C, but it was negligible at 35°C.

Variation in post-inoculation temperature and moisture period affected disease development on spring wheat inoculated with C. sativus. Temperature has been reported to influence the root phase of the disease, but its effect on the resistance to the organism in this phase is still unknown and warrants further investigation. Results of this

investigation on the effect of temperature were not consistent for root rot and foliar infection at seedling stage. Disease severity for the root rot phase was less at 26°C than at 22° or 30°C. The disease was most severe at 30°C at root, seedling, flag leaf and heading stages of the host development. Similar effects of temperature on development of spot blotch on barley (Clark and Dickson 1958; Morton 1962), as well as on spot blotch (Nema and Joshi 1973) and head blight (Andersen 1952) phases on wheat, have been reported previously. Luz and Bergstrom (1986) observed that spot blotch development increased with increasing temperature, and at 28°C all plants developed severe symptoms regardless of genotype. The results obtained in this investigation differ from their investigation. Marberg, GP248, GP250, GP251, GP253, GP254 were resistant at the highest temperature (30°C). Luz and Bergstrom (1986) used only three cultivars to test the effect of temperature on spot blotch development. In this investigation 16 cultivars were evaluated, which possibly could be the reason for detecting a number of sources for resistance at the higher temperature.

Post-inoculation moist period is also a critical factor for disease development on wheat leaves inoculated with C. sativus (Luz 1982). The results showed that the disease severity on roots, foliage at seedling and flag leaf stages, and on heads increased significantly when post-inoculation period increased from 24 hours through 72 hours.

The relationship between the reaction of cultivar to various phases of the disease was examined. All the Canadian cultivars and Marberg were resistant to the root rot phase of the disease. Marberg, GP248, GP251, GP252, GP253, GP254, GP255, BR4 and BR8 were resistant to the foliar

phase, while Marberg, GP248, GP253, GP254, GP255 and BR8 were resistant to the head blight/black point phase of the disease (Appendix Table 66). The reactions of cultivars to detached leaf inoculation were inconsistent with those of intact leaves. The cultivars GP252 and GP253 were susceptible in detached leaf inoculation whereas they were resistant to whole plant inoculations.

This investigation shows that the isolates from Montana and Bangladesh, irrespective of their origin, were found to cause infection to various parts of the plants, including subcrown internodes, leaves and heads. Each of the isolates was capable of producing symptoms of the disease at all phases in varying degrees of severity (Appendix Tables 67-72). They seem to be mutually inclusive and together form a "group of pathogens" instead of being highly specialized cultures capable of producing a particular phase of the disease.

The resistance in some of the cultivars such as Marberg, GP248, GP253, GP254 and GP255 was effective against all phases of the disease. It will be of major importance to use these valuable sources of resistance in wheat breeding programs to develop high yielding, resistant and well adapted cultivars.

## SUMMARY

1. Sources of resistance were identified for different phases of the disease. In the root rot phase the cultivars Marberg, GP248, GP249, GP250, GP251, GP252, GP253, GP254, GP255 and GP256 were resistant. The cultivars Marberg, GP248, GP251, GP252, GP253, GP254, GP255, BR4 and B8 exhibited resistance in the foliar phase, while Marberg, GP248, GP253, GP254, GP255 and BR8 were resistant in the head blight and/or black point phase of the disease.
2. Several cultivars/lines, such as Marberg, GP248, GP253, GP254 and GP255, have resistance to all phases of the disease.
3. Some cultivars/lines, such as GP249, GP256 and BR4, showed differential reaction to various phases of the disease.
4. There were differences in pathogenicity of isolates tested. Isolates from roots were able to attack foliage/heads and vice versa.
5. Isolates of Bangladesh do not have higher temperature optima than the USA isolates.
6. Detached leaf reaction does not always reflect intact leaf reaction.
7. Some cultivars show resistance to isolates of both Bangladesh and the USA:

Root rot phase:	GP248, GP250 and GP251 (Appendix Table 67)
Foliar phase:	BR4 and BR8 (Appendix Table 70)
Head blight/black point phase:	Marberg, GP253 and GP255 (Appendix Table 72)

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APPENDIX

Table 31. Analysis of variance for disease reaction of 16 spring wheat cultivars to subcrown internode inoculation with syringe.

Source	df	SS	MS	F-value	P-value
Blocks	2	2.079	1.04		
Isolate	9	65.50	7.28	32.79	0.0000
Cultivar	15	145.00	9.70	43.70	0.0000
Isolate x cultivar	135	35.50	0.26	1.18	0.1156
Error	318	70.59	0.22		

Table 32. Analysis of variance for disease reaction of 16 spring wheat cultivars to subcrown internode inoculation with infested oat kernels.

Source	df	SS	MS	F-value	P-value
Blocks	2	16.99	8.49		
Isolate	9	23996.00	2666.30	125.69	0.0000
Cultivar	15	25932.00	1728.80	81.50	0.0000
Isolate x cultivar	135	13962.00	103.42	4.88	0.0000
Error	318	6745.90	21.21		

Table 33. Analysis of variance for disease rating of 16 spring wheat cultivars to subcrown internode inoculation with infested seeds.

Source	df	SS	MS	F-value	P-value
Blocks	2	29.50	14.75		
Isolate	9	39978.00	4442.0	155.66	0.0000
Cultivar	15	33037.00	2202.50	77.18	0.0000
Isolate x cultivar	135	14242.00	105.49	3.70	0.0000
Error	318	9074.90	28.54		

Table 34. Analysis of variance for disease reaction of 16 spring wheat cultivars to foliar inoculation of whole plants at seedling stage.

Source	df	SS	MS	F-value	P-value
Blocks	2	0.1354	0.0677	882.56	0.0000
Isolate	3	264.44	88.15	882.56	0.0000
Cultivar	15	28.01	1.87	18.69	0.0000
Isolate x cultivar	45	13.82	0.307	3.07	0.0000
Error	126	12.59	0.0999		

Table 35. Analysis of variance for percent black pointed seeds of 16 spring wheat cultivars to inoculated with isolates of G. sativus.

Source	df	SS	MS	F-value	P-value
Blocks	2	207.19	103.60		
Isolate	3	6644.0	2214.7	70.27	0.0000
Cultivar	15	3241.7	216.12	6.86	0.0000
Isolate x cultivar	45	1607.3	35.72	1.13	0.2905
Error	126	3971.0	31.52		

Table 36. Analysis of variance for lesion length development on inoculated detached seedling leaves of 16 spring wheat cultivars at temperature of 22°C.

Source	df	SS	MS	F-value	P-value
Cultivar	15	8.3414	0.5560	9.85	0.0001
Isolate	9	561.8202	62.4244	1106.01	0.0001
Cultivar x isolate	135	25.9900	0.1925	3.41	0.0001
Error	320	18.0611	0.05644		

Table 37. Analysis of variance for necrotic area development on inoculated detached seedling leaves of 16 spring wheat cultivars at temperature of 22°C.

Source	df	SS	MS	F-value	P-value
Cultivar	15	5.4886	0.3659	10.58	0.0001
Isolate	9	200.7518	22.3057	644.73	0.0001
Cultivar x isolate	135	23.6179	0.1749	5.06	0.0001
Error	320	11.0710	0.0345		

Table 38. Analysis of variance for percent leaf necrosis on inoculated detached seedling leaves of 16 spring wheat cultivars at temperature of 22°C.

Source	df	SS	MS	F-value	P-value
Cultivar	15	4423.1304	294.8754	14.32	0.0001
Isolate	9	243664.3960	27073.8218	1315.02	0.0001
Cultivar x isolate	135	13975.4078	103.5215	5.03	0.0001
Error	320	6588.2084	20.5882		

Table 39. Analysis of variance for lesion length development on inoculated detached mature leaves of 16 spring wheat cultivars at temperature of 22°C:

Source	df	SS	MS	F-value	P-value
Cultivar	15	101.0586	6.7372	1.23	0.2489
Isolate	9	407.4829	45.2758	8.25	0.0001
Cultivar x isolate	135	833.9183	6.1771	1.13	0.2002
Error	320	1755.9531	5.4873		

Table 40. Analysis of variance for necrotic area development on inoculated detached mature leaves of 16 spring wheat cultivars at temperature of 22°C.

Source	df	SS	MS	F-value	P-value
Cultivar	15	63.9720	4.2648	29.79	0.0001
Isolate	9	425.7569	47.3063	330.42	0.0001
Cultivar x isolate	135	183.9009	1.3622	9.51	0.0001
Error	320	45.8140	0.1431		

Table 41. Analysis of variance for percent leaf necrosis on inoculated detached mature leaves of 16 spring wheat cultivars at temperature of 22°C.

Source	df	SS	MS	F-value	P-value
Cultivar	15	30737.0899	2049.1393	70.43	0.0001
Isolate	9	149630.6587	16625.6287	571.40	0.0001
Cultivar x isolate	135	70020.4101	518.6697	17.83	0.0001
Error	320	9310.8521	29.0964		

Table 42. Analysis of variance for lesion length development on inoculated detached seedling leaves of 16 spring wheat cultivars at temperature of 26°C.

Source	df	SS	MS	F-value	P-value
Cultivar	15	28.4929	1.8995	18.31	0.0001
Isolate	9	683.4107	75.9345	731.86	0.0001
Cultivar x isolate	135	88.0522	0.6522	6.29	0.0001
Error	320	33.2016	0.1037		

Table 43. Analysis of variance for necrotic area development on inoculated detached seedling leaves of 16 spring wheat cultivars at temperature of 26°C.

Source	df	SS	MS	F-value	P-value
Cultivar	15	3.7757	0.2517	6.38	0.0001
Isolate	9	223.1790	24.7976	628.79	0.0001
Cultivar x isolate	135	36.2822	0.2687	6.81	0.0001
Error	320	12.6198	0.0394		

Table 44. Analysis of variance for percent leaf necrosis on inoculated detached seedling leaves of 16 spring wheat cultivars at temperature of 26°C.

Source	df	SS	MS	F-value	P-value
Cultivar	15	12850.8626	856.7242	20.92	0.0001
Isolate	9	267321.8434	29702.4270	725.21	0.0001
Cultivar x isolate	135	41942.6019	310.6859	7.59	0.0001
Error	320	13106.2941	40.9572		

Table 45. Analysis of variance for lesion length development on inoculated detached mature leaves of 16 spring wheat cultivars at temperature of 26°C.

Source	df	SS	MS	F-value	P-value
Cultivar	15	320.5893	21.7326	0.98	0.4715
Isolate	9	889.8683	98.8742	4.55	0.0001
Cultivar x isolate	135	2589.1767	19.1790	0.88	0.7961
Error	320	6948.9124	21.7153		

Table 46. Analysis of variance for necrotic area development on inoculated detached mature leaves of 16 spring wheat cultivars at temperature of 26°C.

Source	df	SS	MS	F-value	P-value
Cultivar	15	59.1695	3.9446	35.24	0.0001
Isolate	9	564.4512	62.7168	560.24	0.0001
Cultivar x isolate	135	90.1566	0.6678	5.97	0.0001
Error	320	35.8228	0.1119		

Table 47. Analysis of variance for percent leaf necrosis on inoculated detached mature leaves of 16 spring wheat cultivars at temperature of 26°C.

Source	df	SS	MS	F-value	P-value
Cultivar	15	35129.3806	2341.9587	88.97	0.0001
Isolate	9	223240.1469	24804.4608	942.28	0.0001
Cultivar x isolate	135	40965.8127	303.4505	11.53	0.0001
Error	320	8423.6602	26.3239		

Table 48. Analysis of variance for lesion length development on inoculated detached seedling leaves of 16 spring wheat cultivars at temperature of 30°C.

Source	df	SS	MS	F-value	P-value
Cultivar	15	68.4629	4.5641	33.28	0.0001
Isolate	9	540.0213	60.0023	437.48	0.0001
Cultivar x isolate	135	197.7639	1.4649	10.68	0.0001
Error	320	43.8892	0.1371		

Table 49. Analysis of variance for necrotic area development on inoculated detached seedling leaves of 16 spring wheat cultivars at temperature of 30°C.

Source	df	SS	MS	F-value	P-value
Cultivar	15	23.8353	1.5890	24.89	0.0001
Isolate	9	157.7115	17.5235	274.52	0.0001
Cultivar x isolate	135	68.3726	0.5064	7.93	0.0001
Error	320	20.4264	0.0638		

Table 50. Analysis of variance for percent leaf necrosis on inoculated detached seedling leaves of 16 spring wheat cultivars at temperature of 30°C.

Source	df	SS	MS	F-value	P-value
Cultivar	15	29027.9203	1935.1947	61.07	0.0001
Isolate	9	211466.5488	23496.2832	741.54	0.0001
Cultivar x isolate	135	75987.0421	562.8670	17.76	0.0001
Error	320	10139.5118	31.6860		

Table 51. Analysis of variance for lesion length development on inoculated detached mature leaves of 16 spring wheat cultivars at temperature of 30°C.

Source	df	SS	MS	F-value	P-value
Cultivar	15	137.9615	9.1974	3.36	0.0001
Isolate	9	474.7378	52.7486	19.27	0.0001
Cultivar x isolate	135	442.0877	3.2747	1.20	0.1024
Error	320	875.9020	2.7371		

Table 52. Analysis of variance for necrotic area development on inoculated detached mature leaves of 16 spring wheat cultivars at temperature of 30°C.

Source	df	SS	MS	F-value	P-value
Cultivar	15	191.2328	12.7488	116.09	0.0001
Isolate	9	438.1232	48.6803	443.30	0.0001
Cultivar x isolate	135	106.2556	0.7870	7.17	0.0001
Error	320	35.1407	0.1098		

Table 53. Analysis of variance for percent leaf necrosis on inoculated detached mature leaves of 16 spring wheat cultivars at temperature of 30°C.

Source	df	SS	MS	F-value	P-value
Cultivar	15	68477.1081	4565.1405	143.18	0.0001
Isolate	9	179684.6310	19964.9590	626.17	0.0001
Cultivar x isolate	135	38837.4207	287.6846	9.02	0.0001
Error	320	10202.9734	31.8843		

Table 54. Analysis of variance for root infection of 16 spring wheat cultivars inoculated with different isolates of Cochliobolus sativus at 22°C across three moisture periods.

Source	df	SS	MS	F-value	P-value
Moisture	2	7.0751	3.5375	21.96	0.0001
Replications (moisture)	6	2.1016	0.3502	2.17	0.0447
Cultivar	15	130.9869	8.7324	54.21	0.0001
Moisture x cultivar	30	21.2293	0.7076	4.39	0.0001
Isolate	3	495.6093	165.2031	1025.65	0.0001
Moisture x isolate	6	11.1190	1.8531	11.51	0.0001
Isolate x cultivar	45	60.7723	1.3504	8.38	0.0001
Moisture x isolate x cultivar	90	40.7143	0.4523	2.81	0.0001
Error	378	60.885	0.1610		

Table 55. Analysis of variance for foliar infection at seedling stage of 16 spring wheat cultivars inoculated with different isolates of Cochliobolus sativus at 22°C across three moisture periods.

Source	df	SS	MS	F-value	P-value
Moisture	2	141.7951	70.8975	303.33	0.0001
Replications (moisture)	6	10.8561	1.8093	7.74	0.0001
Cultivar	15	57.6802	3.8453	16.45	0.0001
Moisture x cultivar	30	16.1365	0.5378	2.30	0.0001
Isolate	3	489.4733	163.1577	698.06	0.0001
Moisture x isolate	6	51.5219	8.5869	36.74	0.0001
Isolate x cultivar	45	34.7474	0.7721	3.30	0.0001
Moisture x isolate x cultivar	90	46.7396	0.5193	2.22	0.0001
Error	378	88.3505	0.2337		

Table 56. Analysis of variance for foliar infection at flag leaf stage of 16 spring wheat cultivars inoculated with different isolates of Cochliobolus sativus at 22°C across three moisture periods.

Source	df	SS	MS	F-value	P-value
Moisture	2	64.5266	32.2633	303.71	0.0001
Replications (moisture)	6	18.3245	3.0540	28.75	0.0001
Cultivar	15	126.2027	8.4135	79.20	0.0001
Moisture x cultivar	30	68.4466	2.2815	21.48	0.0001
Isolate	3	397.4874	132.4958	1247.24	0.0001
Moisture x isolate	6	31.6444	5.2740	49.65	0.0001
Isolate x cultivar	45	67.5559	1.5012	14.13	0.0001
Moisture x isolate x cultivar	90	77.5155	0.8612	8.11	0.0001
Error	378	40.1554	0.1062		

Table 57. Analysis of variance for head infection of 16 spring wheat cultivars inoculated with different isolates of Cochliobolus sativus at 22°C across three moisture periods.

Source	df	SS	MS	F-value	P-value
Moisture	2	72.6485	36.3242	189.77	0.0001
Replications (moisture)	6	11.0380	1.8396	9.61	0.0001
Cultivar	15	161.9566	10.7971	56.41	0.0001
Moisture x cultivar	30	68.4466	2.2815	21.48	0.0001
Isolate	3	240.7414	80.2471	419.23	0.0001
Moisture x isolate	6	31.6444	5.2740	49.65	0.0001
Isolate x cultivar	45	113.4716	2.5215	13.17	0.0001
Moisture x isolate x cultivar	90	91.4359	1.0159	5.31	0.0001
Error	378	72.3553	0.1914		

Table 58. Analysis of variance for root infection of 16 spring wheat cultivars inoculated with different isolates of Cochliobolus sativus at 26°C across three moisture periods.

Source	df	SS	MS	F-value	P-value
Moisture	2	0.6600	0.3300	2.23	0.1088
Replications (moisture)	6	2.8344	0.4724	3.19	0.0045
Cultivar	15	115.1465	7.6764	51.90	0.0001
Moisture x cultivar	30	9.9805	0.3326	2.25	0.0003
Isolate	3	472.3279	157.4426	1064.53	0.0001
Moisture x isolate	6	3.9542	0.6590	4.46	0.0002
Isolate x cultivar	45	73.4539	1.6323	11.04	0.0001
Moisture x isolate x cultivar	90	25.9229	0.2880	1.95	0.0001
Error	378	55.9055	0.1478		

Table 59. Analysis of variance for foliar infection at seedling stage of 16 spring wheat cultivars inoculated with different isolates of Cochliobolus sativus at 26°C across three moisture periods.

Source	df	SS	MS	F-value	P-value
Moisture	2	50.9779	25.4889	111.07	0.0001
Replications (moisture)	6	12.4795	2.0799	9.06	0.0001
Cultivar	15	65.9671	4.3978	19.16	0.0001
Moisture x cultivar	30	21.5509	0.7183	3.13	0.0001
Isolate	3	391.5918	130.5306	568.79	0.0001
Moisture x isolate	6	46.4737	7.7456	33.75	0.0001
Isolate x cultivar	45	62.3381	1.3852	6.04	0.0001
Moisture x isolate x cultivar	90	75.4329	0.8381	3.65	0.0001
Error	378	86.7470			

Table 60. Analysis of variance for foliar infection at flag leaf stage of 16 spring wheat cultivars inoculated with different isolates of Cochliobolus sativus at 26°C across three moisture periods.

Source	df	SS	MS	F-value	P-value
Moisture	2	33.5098	16.7549	109.90	0.0001
Replications (moisture)	6	7.7234	1.2872	8.44	0.0001
Cultivar	15	126.3083	8.4205	55.23	0.0001
Moisture x cultivar	30	53.2451	1.7748	11.64	0.0001
Isolate	3	475.3486	158.4495	1039.29	0.0001
Moisture x isolate	6	19.4267	3.2377	21.24	0.0001
Isolate x cultivar	45	88.0510	1.9566	12.83	0.0001
Moisture x isolate x cultivar	90	57.5894	0.6398	4.20	0.0001
Error	378	57.6298	0.1524		

Table 61. Analysis of variance for head infection of 16 spring wheat cultivars inoculated with different isolates of Cochliobolus sativus at 26°C across three moisture periods.

Source	df	SS	MS	F-value	P-value
Moisture	2	60.8051	30.4025	145.08	0.0001
Replications (moisture)	6	18,3329	3.0554	14.58	0.0001
Cultivar	15	130.5955	8.7063	41.55	0.0001
Moisture x cultivar	30	30.4415	1.0147	4.84	0.0001
Isolate	3	347.4227	115.8075	552.62	0.0001
Moisture x isolate	6	25.8359	4.3059	20.55	0.0001
Isolate x cultivar	45	60.2838	1.3396	6.39	0.0001
Moisture x isolate x cultivar	90	47.5040	0.5278	2.52	0.0001
Error	378	79.2137			

Table 62. Analysis of variance for root infection of 16 spring wheat cultivars inoculated with different isolates of Cochliobolus sativus at 30°C across three moisture periods.

Source	df	SS	MS	F-value	P-value
Moisture	2	21.7001	10.8500	106.69	0.0001
Replications (moisture)	6	6.3183	1.0530	10.35	0.0001
Cultivar	15	77.0199	5.1346	50.49	0.0001
Moisture x cultivar	30	14.8931	0.4964	4.88	0.0001
Isolate	3	552.0700	184.0233	1809.52	0.0001
Moisture x isolate	6	8.2876	1.3812	13.58	0.0001
Isolate x cultivar	45	38.5927	0.8576	8.43	0.0001
Moisture x isolate x cultivar	90	35.7279	0.3969	3.90	0.0001
Error	378	38.4416	0.1016		

Table 63. Analysis of variance for foliar infection at seedling stage of 16 spring wheat cultivars inoculated with different isolates of Cochliobolus sativus at 30°C across three moisture periods.

Source	df	SS	MS	F-value	P-value
Moisture	2	30.0476	15.023	156.05	0.0001
Replications (moisture)	6	4.5133	0.7522	7.81	0.0001
Cultivar	15	74.6171	4.9744	51.67	0.0001
Moisture x cultivar	30	26.6968	0.8898	9.24	0.0001
Isolate	3	550.0840	183.3613	1904.49	0.0001
Moisture x isolate	6	20.7673	3.4612	35.95	0.0001
Isolate x cultivar	45	57.6570	1.2812	13.31	0.0001
Moisture x isolate x cultivar	90	73.1548	0.8128	8.44	0.0001
Error	378	36.3933	0.0962		

Table 64. Analysis of variance for foliar infection at flag leaf stage of 16 spring wheat cultivars inoculated with different isolates of Cochliobolus sativus at 30°C across three moisture periods.

Source	df	SS	MS	F-value	P-value
Moisture	2	19.6859	9.84	78.90	0.0001
Replications (moisture)	6	13.9908	2.3318	18.69	0.0001
Cultivar	15	158.3354	10.5556	84.61	0.0001
Moisture x cultivar	30	21.4318	0.7143	5.73	0.0001
Isolate	3	506.9113	168.9704	1354.46	0.0001
Moisture x isolate	6	9.0334	1.5055	12.07	0.0001
Isolate x cultivar	45	134.2920	2.9842	23.92	0.0001
Moisture x isolate x cultivar	90	53.8931	0.5988	4.80	0.0001
Error	378	47.1558	0.1247		

Table 65. Analysis of variance for head infection of 16 spring wheat cultivars inoculated with different isolates of Cochliobolus sativus at 30°C across three moisture periods.

Source	df	SS	MS	F-value	P-value
Moisture	2	60.4019	30.2009	239.27	0.0001
Replications (moisture)	6	17.0411	2.8401	22.50	0.0001
Cultivar	15	139.8355	9.3223	73.86	0.0001
Moisture x cultivar	30	15.7352	0.5245	4.16	0.0001
Isolate	3	445.0897	148.3632	1175.41	0.0001
Moisture x isolate	6	24.2112	4.0352	31.97	0.0001
Isolate x cultivar	45	67.2155	1.4936	11.83	0.0001
Moisture x isolate x cultivar	90	38.3560	0.4261	3.38	0.0001
Error	378	47.7121	0.1262		

Table 66. Resistance of 16 spring wheat cultivars at different phases of the disease.

	SCI <sup>1</sup>			Foliar <sup>2</sup>		Black point
	Syr <sup>a</sup>	Oats <sup>b</sup>	CMC <sup>c</sup>	Whole plants <sup>a</sup>	Detached leaves <sup>b</sup>	
Fortuna	I	S	S	I	S	S
Marberg	R	R	R	R	I	R
Rescue	S	S	S	S	S	S
GP248	R	R	R	R	R	R
GP249	R	R	R	I	I	S
GP250	R	R	R	I	R	I
GP251	R	R	R	R	R	I
GP252	I	R	R	R	S	I
GP253	R	R	R	R	S	R
GP254	R	R	R	R	I	R
GP255	I	R	R	R	R	R
GP256	R	I	R	I	R	S
BR4	I	R	R	R	R	S
BR8	I	I	I	R	R	R
CNT8	I	I	I	I	R-I	S
BH1146	S	S	S	I	R	I

<sup>1</sup> SCI = Subcrown internode phase<sup>2</sup> Foliar phase<sup>a</sup> Syr = Syringe inoculation

R = resistant (DR=1.3-1.9)

I = intermediate (DR=2.0-2.8)

S = susceptible (DR=2.8-3.2)

<sup>a</sup> Whole plants

R = resistant (DR=1.4-1.9)

I = intermediate (DR=2.0-2.5)

S = susceptible (DR=2.6-2.9)

<sup>b</sup> Oats = infested oat kernels

R = resistant (DR=17.0-25.0%)

I = intermediate (DR=26.0-34.0%)

S = susceptible (DR=35.0-43.0%)

<sup>b</sup> Detached leaves

R = resistant (DR=42.0-49.0%)

I = intermediate (DR=50.0-57.0%)

S = susceptible (DR=58.0-68.4)

<sup>c</sup> CMC = infested seeds

R = resistant (DR=26.0-35.0%)

I = intermediate (DR=36.0-44.0%)

S = susceptible (DR=45.0-55.0%)

<sup>3</sup> Black point phase

R = resistant (DR=4.0-7.0%)

I = intermediate (DR = 8.0-11.0%)

S = susceptible (DR=12.0-17.0%)

Table 67. Effect of isolates of *Cochliobolus sativus* on resistance of 16 spring wheat cultivars to subcrown internode inoculation with syringe.

Cultivar	Isolate <sup>1</sup>								
	195(1)	214(1)	215(1)	369(1)	370(1)	A <sub>1</sub>	B <sub>1</sub>	C <sub>1</sub>	D <sub>1</sub>
Fortuna	S <sup>2</sup>	S	S	I	I	S	S	S	I
Marberg	R	I	R	R	R	R	I	R	R
Rescue	S	S	S	S	S	S	S	S	S
GP248	R	R	R	R	R	R	R	R	R
GP249	I	R	R	R	R	I	R	R	R
GP250	R	R	R	R	R	R	R	R	R
GP251	R	R	R	R	R	R	R	R	R
GP252	R	I	I	I	R	R	I	R	I
GP253	I	R	R	R	R	I	R	R	R
GP254	R	R	I	R	R	I	R	R	R
GP255	I	I	I	R	R	I	I	R	R
GP256	I	I	I	R	R	I	R	R	R
BR4	I	I	I	I	I	I	I	I	I
BR8	I	S	I	I	I	I	S	S	I
CNT8	S	I	I	I	I	S	R	I	I
BH1146	S	S	S	S	S	S	S	S	S

<sup>1</sup> Isolates: 195(1), 214(1), 215(1), 369(1) and 370(1) from Montana, USA, and A<sub>1</sub>, B<sub>1</sub>, C<sub>1</sub> and D<sub>1</sub> from Bangladesh.

<sup>2</sup> Reaction of cultivars

R = Resistant

I = Intermediate

S = Susceptible

Table 68. Effect of isolates of *Cochliobolus sativus* on resistance of 16 spring wheat cultivars to subcrown internode inoculation with infested oat kernels.

Cultivar	Isolate <sup>1</sup>								
	195(1)	214(1)	215(1)	369(1)	370(1)	A <sub>1</sub>	B <sub>1</sub>	C <sub>1</sub>	D <sub>1</sub>
Fortuna	I <sup>2</sup>	S	S	S	S	I	S	I	S
Marberg	I	R	R	R	R	R	R	I	R
Rescue	S	S	S	S	S	S	S	S	S
GP248	I	R	I	I	I	R	I	R	R
GP249	I	R	R	R	R	R	R	R	R
GP250	R	R	R	R	R	R	R	R	I
GP251	R	R	R	I	R	R	R	R	R
GP252	R	R	R	I	R	R	R	I	R
GP253	R	R	R	R	R	R	R	I	R
GP254	R	R	R	R	R	R	I	R	I
GP255	I	R	R	I	R	R	R	R	R
GP256	I	R	I	R	I	I	R	R	I
BR4	R	R	R	I	I	R	R	S	S
BR8	R	R	R	I	I	I	R	S	S
CNT8	S	I	I	I	I	I	I	S	S
BH1146	S	S	S	I	S	S	I	S	R

<sup>1</sup> Isolates: 195(1), 214(1), 215(1), 369(1) and 370(1) from Montana, USA, and A<sub>1</sub>, B<sub>1</sub>, C<sub>1</sub> and D<sub>1</sub> from Bangladesh.

<sup>2</sup> Reaction of cultivars

R = Resistant

I = Intermediate

S = Susceptible

Table 69. Effect of isolates of *Cochliobolus sativus* on resistance of 16 spring wheat cultivars to subcrown internode inoculation with infested seeds.

Cultivar	Isolate <sup>1</sup>								
	195(1)	214(1)	215(1)	369(1)	370(1)	A <sub>1</sub>	B <sub>1</sub>	C <sub>1</sub>	D <sub>1</sub>
Fortuna	I <sup>2</sup>	S	S	I	S	I	S	I	S
Marberg	I	R	R	R	R	R	R	R	R
Rescue	S	S	S	S	S	S	S	S	S
GP248	I	I	R	I	I	I	R	R	R
GP249	I	R	R	R	R	R	R	R	R
GP250	R	R	R	R	R	R	R	R	I
GP251	R	R	R	I	R	R	R	R	R
GP252	I	R	R	R	I	R	R	R	I
GP253	R	R	R	R	R	R	R	I	R
GP254	R	R	R	R	R	R	R	R	I
GP255	I	R	R	I	R	R	R	R	R
GP256	I	R	I	R	I	I	R	R	I
BR4	I	R	R	I	I	R	R	S	S
BR8	R	R	I	I	I	I	R	S	S
CNT8	S	I	S	I	I	I	I	S	S
BH1146	S	S	S	I	S	S	I	S	R

<sup>1</sup> Isolates: 195(1), 214(1), 215(1), 369(1) and 370(1) from Montana, USA, and A<sub>1</sub>, B<sub>1</sub>, C<sub>1</sub> and D<sub>1</sub> from Bangladesh.

<sup>2</sup> Reaction of cultivars

R = Resistant

I = Intermediate

S = Susceptible

Table 70. Effect of isolates of Cochliobolus sativus on resistance of 16 spring wheat cultivars on foliar inoculation at seedling stage.

Cultivar	Isolate <sup>1</sup>		
	214(1)	A <sub>1</sub>	B <sub>1</sub>
Fortuna	I <sup>2</sup>	I	I
Marberg	I	R	R
Rescue	S	S	S
GP248	I	R	R
GP249	S	I	S
GP250	I	I	R
GP251	I	R	R
GP252	I	R	I
GP253	I	R	I
GP254	I	R	R
GP255	I	R	I
GP256	S	I	I
BR4	R	R	R
BR8	R	R	R
CNT8	I	I	I
BH1146	S	S	I

<sup>1</sup> Isolates: 214(1) from Montana, USA, and A<sub>1</sub> and B<sub>1</sub> from Bangladesh.

<sup>2</sup> Reaction of cultivars

R = Resistant

I = Intermediate

S = Susceptible

Table 71. Effect of isolates of *Cochliobolus sativus* on resistance of 16 spring wheat cultivars on inoculated detached seedling leaves.

Cultivar	Isolate <sup>1</sup>								
	195(1)	214(1)	215(1)	369(1)	370(1)	A <sub>1</sub>	B <sub>1</sub>	C <sub>1</sub>	D <sub>1</sub>
Fortuna	S <sup>2</sup>	S	I	S	I	S	R	S	S
Marberg	S	S	R	I	R	S	R	R	I
Rescue	S	S	S	S	S	S	S	I	S
GP248	I	R	R	R	R	S	S	I	R
GP249	I	I	S	I	R	S	S	I	S
GP250	S	R	R	I	I	R	S	I	I
GP251	R	R	R	I	I	R	S	I	I
GP252	S	S	I	S	S	I	S	S	S
GP253	S	S	S	S	S	I	S	S	S
GP254	S	R	S	I	I	I	S	S	R
GP255	I	R	R	I	R	R	S	S	I
GP256	R	R	R	I	S	R	R	I	I
BR4	R	R	I	I	I	R	I	I	I
BR8	R	R	I	I	R	R	S	I	R
CNT8	I	S	R	R	I	I	S	R	S
BH1146	R	I	I	I	I	R	S	I	I

<sup>1</sup> Isolates: 195(1), 214(1), 215(1), 369(1) and 370(1) from Montana, USA, and A<sub>1</sub>, B<sub>1</sub>, C<sub>1</sub> and D<sub>1</sub> from Bangladesh.

<sup>2</sup> Reaction of cultivars

R = Resistant

I = Intermediate

S = Susceptible

Table 72. Effect of isolates of Cochliobolus sativus on resistance of 16 spring wheat cultivars to black point.

Cultivar	Isolate <sup>1</sup>		
	214(1)	A <sub>1</sub>	B <sub>1</sub>
Fortuna	S <sup>2</sup>	S	S
Marberg	R	R	R
Rescue	S	S	S
GP248	R	I	R
GP249	S	I	R
GP250	I	I	I
GP251	I	S	R
GP252	S	I	I
GP253	R	R	R
GP254	R	I	R
GP255	R	R	R
GP256	S	S	I
BR4	I	S	I
BR8	I	R	R
CNT8	S	S	S
BH1146	I	I	I

<sup>1</sup> Isolates: 214(1) from Montana, USA, and A<sub>1</sub> and B<sub>1</sub> from Bangladesh.

<sup>2</sup> Reaction of cultivars

R = Resistant

I = Intermediate

S = Susceptible

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