COMPONENTS OF PARTIAL RESISTANCE, MODE OF INHERITANCE OF RESISTANCE TO SEPTORIA TRITICI BLOTCH, AND STATUS OF SEPTORIA DISEASES IN MOROCCO

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CHAPTER I

INTRODUCTION

Septoria is the name commonly applied to over 1,000 form-species of fungi, most of which are plant parasites. Approximately 100 species are parasitic on cereals and grasses (Scharen and Sanderson, 1985). Twenty-five years ago, little research had been conducted on septoria diseases of cereals. However, this has changed since then with regular publication of research from most of the wheat growing areas of the world.

Two major pathogens of the septoria group have had the greatest impact on wheat production: *Mycosphaerella graminicola* (Fuckel) Schroeter (Anamorph: *Septoria tritici* Rob.ex Desm.) and *Leptosphaeria nodorum* Muller [Anamorph: *S. nodorum* (Berk.) Berk.]. *M. graminicola* incites the disease septoria tritici blotch (STB) of wheat and causes losses even where conditions are only marginally conducive to development of the disease (Eyal et al., 1987). STB can be pernicious and yield losses from slight to 60% have been attributed to natural infection (Brownell and Gilchrist, 1979; Eyal, 1972; Eyal et al., 1987; King et al., 1983; Shipton et al., 1971). Increased occurrence and severity of STB have elevated *S. tritici* to a pathogen of worldwide importance, and has resulted in numerous research and crop

improvement programs worldwide (Brown and Rosielle, 1980; Djerbi etal., 1974; Eyal, 1972; Forni and Zitelli, 1979; Gough and Smith, 1985; Mann et al., 1985; Tyagi et al., 1969; Van Ginkel, 1986; Van Ginkel and Scharen, 1988).

STB, formerly a minor disease in North Africa, is now a major disease problem of wheat in countries such as Morocco and Tunisia. This change is due in part to large-scale plantings of introduced, high-yielding wheats, and to the occurrence of weather conditions favorable for development of septoria diseases (Djerbi et al., 1974; Saari and Wilcoxson, 1974; Stewart et al., 1972).

The destructiveness of STB in Morocco has been recognized sporadically since the late 1960's. A severe epidemic occurred in 1968-1969 when the growing season was unusually wet and cool. All high-yielding varieties were susceptible to the disease, particularly Siete Cerros, which was severely infected by *Septoria* (Santiago, 1970). Saâdaoui (1975) found *S. tritici* to be the predominant species, and Schluter and Janati (1976) reported losses up to 18% in fungicide experiments. The perfect state of *S. tritici* has not yet been reported, although the disease is becoming widely distributed in Morocco (Saâdaoui, 1987). Developing resistance to STB has become an important component of the Moroccan Cereal Breeding Program. Recently, a number of bread wheat cultivars with diverse sources of resistance (mainly from winter wheat germplasm) were introduced, and their resistance was further increased by

selection (Jlıbene, 1990).

Therefore the objectıves of my research were to:

1. Investigate the components of partial resistance to *S. tritici* in selected bread wheat cultıvars wıth different sources of resistance.
2. Study the mode of ınherıtance of seedlıng plant resistance to septorıa tritici blotch ın sprıng wheats.
3. Determine the occurrence, distribution and severıty of septorıa dıseases in Morocco.

CHAPTER II

LITERATURE REVIEW

Pathogen

Within the form-class Fungi Imperfecti, *S. tritici* is classified in the order Sphaeropsidales. Slender, elongated pycnidiospores (35-98 μ x 1-3 μ) are produced within pycnidia, which are embedded in the epidermal and mesophyll tissues on both sides of the leaf. When exposed to moisture, the spores often exude from ostioles in worm-like masses or cirrhi. The teleomorph state, *M. graminicola* is associated with the loculoascomycetes, order Dothideales, in the family Dothideaceae. This family has a perithecioid pseudothecium as a fruiting body that contains the asci. Each ascus contains eight ascospores with two cells of unequal size (Eyal et al., 1987; Scharen and Sanderson, 1985). The pseudothecial structure has two layers with the outer layer pigmented (Sanderson et al., 1985).

Prior to 1983, confusion existed in the literature and among workers regarding the nomenclature of the organisms causing the septoria diseases of cereals. In 1983, at a workshop on the Septoria Diseases of Cereals, participants agreed that the taxonomic names of the fungi involved in the septoria disease complex would be based on their perfect

state, namely *Leptosphaeria nodorum* E. Muller, *L. avenaria* Web. f.sp. *triticea* and *Mycosphaerella graminicola* (Fuckel) Schroeter. This agreement included the common names of the disease as septoria nodorum blotch, septoria avenae blotch, and septoria tritici blotch, that the lower case "s" be used for septoria in common names, etc, and that italics would not be used in these common names (Scharen, 1985).

Environmental Conditions Required for Germination, Penetration, & Infection

Pycnidiospores are exuded through the ostiole of a pycnidium and are embedded in a mucilaginous cirrhus that prevents germination *in situ* but promotes germination when water is available. Germination occurs either by elongation of the apical cell or by budding. In the laboratory, spores germinate on moist leaves within 12 hr and penetrate either through stomata or directly through the walls of the epidermis after 24 hr (Renfro and Young, 1956). Moisture is important at all stages of infection (Hess and Shaner, 1985 & 1987; Hilu and Bever, 1957; King et al., 1983; Shaner and Finney, 1976; Shipton et al., 1971). Plants subjected to a 96 hr-moist period after inoculation develop a more severe infection type than plants subjected to shorter moist periods (Shipton et al., 1971). A moist period of only 24 hr is generally insufficient to produce disease symptoms (Hess and Shaner, 1985).

Temperatures reported for germination of *S. tritici* conidia are a minimum of 2-3 C and a maximum of 33-37 C with

an optimum of 20-25 (Eyal et al., 1987; Shipton et al., 1971). Two consecutive days with a minimum temperature of 7 C or less inhibited infection (Renfro and Young, 1956; Shaner and Finney, 1976). Low temperatures affect spore germination, mycelial growth, and lesion and pycnidia development by lengthening the time required for each (Eyal et al., 1987).

The time from infection to production of pycnidia depends on environmental conditions (moisture, temperature, light), cultivar, and the *Septoria* isolate. A compensation between moisture duration and temperature appears to exist in susceptible wheat cultivars with severe disease resulting from either long moist periods followed by cool incubation conditions or short moist periods followed by warm incubation conditions (Hess and Shaner, 1985 and 1987). The optimum light intensity for spore germination in *S. tritici* and mycelial growth on leaf surfaces is 8,000-12,000 Lux (Benedict, 1971), while pycnidial formation is most rapid at 2,000 Lux. It may be concluded that the infection process occurs best on rainy, cloudy days with temperatures between 20-25 C.

Symptom Expression and Disease Development

The host range of *S. tritici* is limited to the cereals primarily with the main economic host being *Triticum* spp. On wheat, symptoms develop seven to 21 days after fungal penetration. All above-ground parts may be infected and

develop characteristic lesions, although the symptoms and signs are usually confined to the foliage. Root development may be reduced (Gough and Merkle, 1977). After penetration the hyphae enlarge slightly and grow in all directions. The hyphae are intercellular with few exceptions and do not produce haustoria (Shipton et al., 1971). Initial symptoms are chlorotic flecks that are usually found on the lowermost leaves. The flecks expand into irregular necrotic lesions. Lesions tend to be restricted laterally and assume parallel sides. Initially they appear sunken and grayish-green. The pathogen advances within killed tissues, but necrosis extends beyond colonized cells, apparently because of diffusible toxins. A phytotoxin produced in cultures of *S. tritici* reproduced all the symptoms of the pathogen when applied to leaves (Malcolm, 1978). The coalescing lesions may occupy most of the leaf tissue and abundant pycnidia are produced within 13 days according to Weber (1922). Pycnidia range in color from light to dark brown, are scattered within the lesion and can occur on both the upper and lower surfaces of the leaf. The size of pycnidia may vary among cultivars and is affected by the number of pycnidia present (Eyal and Brown, 1976). Pycnidiospore production may be related to cultivar response, with lower production occurring on resistant cultivars (Gough, 1978).

Pycnidiospores can remain viable in pycnidia on infected stubble for several months (Hilu and Bever, 1957). Although *S. tritici* can colonize wild grasses, which could

act as a reservoir of primary inoculum (Brokenshire, 1975), crop residues are considered the most likely source of primary inoculum (Holmes and Colhoun, 1975; King et al., 1983; Sanderson et al., 1985). Whether this inoculum is in the form of pycnidiospores or ascospores depends on the weather conditions to which the crop debris are exposed. Pycnidiospores are responsible for short-range dispersal within crops while ascospores account for long-range dispersal between crops (Sanderson et al., 1985).

Spores are produced in a thick, sticky matrix containing a high concentration of preserving sugars and proteins (Fournet, 1969). This preserving medium permits spores to remain viable during periods of dry weather. Release of pycnidiospores occurs when free water is present on infected host tissue or when the ambient air is almost saturated (King et al., 1983). An oozing drop, or cirrhus, containing pycnidiospores is exuded through the ostiole at the top of a pycnidium following leaf wetting. After drying, part of the oozing drop may return into the pycnidium or remain on the top of the ostiole (Eyal et al., 1987).

Septoria tritici reportedly is incapable of forming pycnidia in dead host tissue (Djerbi et al., 1977), and pycnidia are not capable of regenerating new pycnidiospores after release of spores (Eyal et al., 1987). The bulk of the spores in a pycnidium are released on the first wetting (Eyal, 1971). After each wetting, fewer pycnidiospores are released. Due to the difficulty in detecting the fruiting

bodies of *M. graminicola*, the occurrence of the perfect state has only been reported in Australia, Brazil, the United Kingdom, New Zeland, and the United States (Eyal et al., 1987; Madariaga et al., 1989; Metha, 1989; Sanderson et al., 1985).

Sources of Resistance

There are numerous reports on the existence of sources of resistance to *S. tritici*. Amongst the hexaploid wheats, resistance was first reported by Beach (1919) who found that within the ten varieties he tested, six were susceptible, two intermediate, and two resistant. In 1957, Narvaez and Caldwell reported resistance in three wheat varieties following artificial inoculation; namely Nabob, Lerma 52, and P 14. Later, Sewell and Caldwell (1960) demonstrated high resistance in the varieties Kanqueen and P 14-3H-15H-2H using excised seedling leaves maintained on benzimidazole solution. Following artificial inoculation, Rillo and Caldwell (1966) reported high resistance in Bulgaria 88. In an extensive screening program in Western Australia, Rosielle (1972) listed 34 resistant common bread wheat accessions. Numerous resistant varieties including PV-18, Lerma Rojo, and Sonora 63 were found in North-Western Punjab by Tyagi et al. (1969). In Great Britain, Baker (1970) observed large varietal differences in commercial wheat varieties using a detached leaf technique. Forni and Zetelli (1979) reported that 57 of 108 F5 plants from a cross

between Altas 66 and Super X appeared to be resistant. In contrast, Weber (1922) found that all 250 wınter wheats he tested using artıficial inoculation were susceptible. Rosen (1947) was able to transfer resistance by hybridızatıon ın a cross between (Red Rock x Hope) with CI 12017, and to increase resistance levels by selectıon.

Resistance to septoria has been reported in other *Triticum* spp. Beach (1919) found that *T. vulgare* varieties differed in susceptibility and that the fungus failed to infect *T. dicoccum*, *T. durum*, and *T. polonicum*. In 1922, Weber reported that two accessions of *T. dicoccum* were susceptible, as was one accession of each of the following: *T. compactum*, *T. durum*, *T. monococcum*, *T. polonıcum*, *T. spelta*, and *T. turgidum*. Weber (1922) also obtaıned infection on rye (*Secale cereale*) and *Poa pratensıs*. Mackıe (1929) could not obtaın immune varıetıes ın *T. vulgare* but found many immune varieties in other *Tritıcum* specıes. No indication was given as to the manner of testing. In Punjab, India, nearly ımmune varietıes of *T. durum* were found by Luthra et al. (1938). Resistance was also found ın two varıeties of *T. durum* with a longer period of ıncubatıon ın comparison to *T. vulgare* (Hilu and Bever, 1957). Hıgh resistance to *S. tritici*, was observed in *T. carthlıcum*, *T. dicoccum*, *T. polonicum*, *T. pyramidale* and many cultivars of *T. durum*, and *T. aestivum* (Rosielle, 1972). Brokenshire (1976) reported that the hexaploid wheats *T. aestıvum*, *T. spelta*, and *T. compactum* were more susceptıble than the

tetraploid species, except for a highly susceptible selection of *T. dicoccum*. Pycnidia were absent from leaves and heads of triticale, *T. timopheevi*, and *T. polonicum*. Resistant entries of *T. dicoccum*, *T. timopheevi*, and *T. spelta* were identified after seedling inoculation by Krupinsky et al. (1977).

Populations and accession lines of diploid and tetraploid wild *Triticum* spp. with different genomes were evaluated for seedling resistance to seven *S. tritici* isolates (Yechilevich-Auster et al., 1983). Of 22 *T. monococcum boeoticum* lines (genome AA), only two were susceptible while 25 of 47 wild emmer lines (*T. turgidum dicoccoides*) were resistant. A high level of resistance to *S. tritici* has been detected among populations and accessions of *T. longissimum*, *T. speltoides*, and *T. tauschii*.

Resistance to *S. tritici* has been transferred from *Agropyron elongatum* (Gough and Tuleen, 1979; Rillo et al., 1970), and from triticale (May, 1983) to bread wheat. In many countries (Djerbi et al., 1976; Eyal, 1981; Eyal et al., 1987; Scharen and Eyal, 1980 and 1983), durum wheats show a higher frequency of resistance to *S. tritici* than spring bread wheats. However, in Tunisia several bread wheat lines and cultivars were highly resistant to *S. tritici* whereas few durum wheat cultivars showed good resistance (Djerbi et al., 1976). The International Maize and Wheat Improvement Center (CIMMYT) requires resistance to all

prevalent mexican isolates of *S. tritici* in bread wheat distributed internationally from their wheat program, which should effectively increase resistant germplasm on a worldwide basis (Van Ginkel and Scharen, 1988).

Resistance to STB appears to be more widely distributed among *T. aestivum* cultivars with winter growth habit than among those with spring growth habit (Eyal, 1981). The utilization of resistant winter wheat germplasm of diverse sources in spring wheat breeding programs may enhance genetic protection against *S. tritici* (Eyal., 1981).

Inheritance of Resistance to *S. tritici*

Although incorporation of resistance to *S. tritici* is an objective in many breeding programs and several sources of resistance have been reported, little is known about the number and the nature of the genes controlling resistance and how they relate to each other.

Simple inheritance has been demonstrated in some cultivars. Mackie (1929) showed that resistance in an unnamed variety was inherited as a single recessive gene. In the spring wheat cultivars Lerma 50 and P 14 resistance was conditioned by a single dominant gene (Narvaez and Caldwell, 1957). Resistance of the winter wheat cultivar Bulgaria 88, governed by a single dominant gene expressed in the adult plant (Rillo and Caldwell, 1966), was incorporated into the soft red winter wheat cultivar Oasis (Patterson et al., 1974). It was reported that inheritance of resistance

in cultivars Veranopolis and IAS-20 might be due to a single gene and that the inheritance pattern and the breeding behavior of resistance were similar in both cultivars. This suggested that these cultivars may have the same gene for resistance (Rosielle and Brown, 1979). Inheritance of a single dominant gene also was reported by Wilson (1985) in Veranopolis AUS 1553 and AUS 16144 wheat cultivars; however, the gene for resistance in Veranopolis appeared to be different than the one in IAS-20.

Other modes of inheritance have been reported including duplicate dominant genes (Wilson, 1985) and single incomplete dominance (Lee and Gough, 1984; Wilson, 1985). Resistance in the tall winter wheat cultivar Nabob appeared to be conditioned by two independent genes, each lacking dominance, but with additive effects (Narvaez and Caldwell, 1957). In contrast, the inheritance of resistance in the cultivar Seabreeze seemed to be determined by recessive genes in at least three loci (Rosielle and Brown, 1979). Resistance of the four winter wheats tested by Danon and Eyal (1986), to isolate ATCC 48507 of *M. graminicola*, was controlled by one or two dominant genes.

Using the Kampmeijer (1981) analysis of host-parasite matrices, which is based on the assumption of a gene-for-gene relationship, Yechilevich-Auster et al. (1983) estimated the presence of nine interacting genes between seven *S. tritici* isolates and 44 accessions of bread and durum wheats, wild *Triticum* spp. and triticale. An estimate

of seven resistant components were attributed to the spring wheat line H 574 and a similar number of virulence components was found in some isolates of *S. tritici*. Utilizing the same analysis, Eyal et al. (1985) estimated the presence of 28 different genes in 97 *S. tritici* isolates secured from 22 countries and tested on 35 wheat and triticale cultivars. Four hypothetical genes were indicated in the spring wheat Bobwhite "S". Eyal and Levy (1987) hypothesized 12 complementary genes in 42 *M. graminicola* isolates tested on 16 bread and durum wheat cultivars. The spring bread wheat cultivar Titan with seven hypothesized genes exhibited a high level of resistance. The effectiveness of IAS-20 was much lower (Eyal and Levy, 1987) than that reported from Australia (Rosielle and Brown, 1979).

Most studies of wheat resistance were conducted on seedlings or mature plants in greenhouse under controlled conditions (Brokenshire, 1976; Eyal et al., 1985; Gough and Smith, 1985; Krupinsky et al., 1977; Rillo and Caldwell, 1966; Van Ginkel and Scharen, 1988), or in fields on mature plants (Brokenshire, 1976; Eyal et al., 1983; Rosielle and Brown, 1979; Tyagi et al., 1969; Wilson, 1985). High positive correlations existed between seedling and mature plant reactions (Rillo and Caldwell, 1966), and significant correlations were obtained between field and glasshouse tests (Brokenshire, 1976).

Numerous scales describing percentage leaf area

infected, pycnidial density, or symptom severity have been used to evaluate host plant reaction to *S. tritici* (Eyal and Brown, 1976; Rosielle, 1972; Saari and Prescott, 1975; Shearer and Smith, 1978). The Saari-Prescott 0-9 scale and its modification, the double digit 00-09 scoring scale, describes the vertical progression of disease and severity. The 6-point scale of Rosielle is based on symptom description, and plants with values of two or lower are considered resistant. Eyal et al.(1983) studied the relation between disease severity [percent pycnidia (PCD) of *S. tritici* on the uppermost leaves] and the Septoria Progress Coefficient (SPC), expressed as the ratio between disease height/plant height, in which four distinct cultivar-response classes were categorized. Cultivars with a PCD of 15% combined with a low SPC were considered resistant. Yechilevich-Auster et al. (1983) suggested that a 30% pycnidial coverage be used as the point of differentiation between resistant and susceptible classes. Eyal et al. (1985) used 16.6% necrotic leaf area to differentiate between resistant and susceptible classes, while 28.8 % pycnidial coverage was used to separate between the two intermediate response classes (moderate resistance and moderate susceptible classes) (Eyal and Levy, 1987).

Most field and greenhouse work on inheritance of resistance to *S. tritici* has concentrated on genes presumed to condition major effects. When studying the host-pathogen relationships of durum wheat and *S. tritici*, Van Ginkel and

Scharen (1988) did not observe complete host resistance. Additive gene action was of prime importance, although dominance effects and transgressive segregation also were observed. Epistasis seemed negligible in the durum wheat studied (Van Ginkel, 1986 ;Van Ginkel and Scharen, 1987). Differences due to cultivars and isolates were highly significant while the cultivar x isolate interaction was relatively small and not significant. Thus pathogen-host species specialization, as opposed to pathogen-host cultivar specialization, was proposed to account for the apparent absence of differential gene-for-gene relationships, and variation among different isolate-cultivar combinations results from differences in aggressiveness of pathogen isolates and horizontal resistance of cultivars (Van Ginkel, 1986; Van Ginkel and Scharen, 1988).

While complete immunity to septoria tritici blotch has not been found in bread wheats (*T. aestivum*), many wheats possess effective levels of resistance, frequently referred to as partial resistance. The expression of partial resistance is complex, as is its measurement, because the reaction of a host cultivar to a pathogen isolate may occur in some or in all phases of the infection cycle. Thus differences between cultivars tested with one isolate, or differences between isolates tested on one cultivar, may involve spore germination, penetration, colonization, sporulation, incubation and latent periods, infectious period, and so forth. As resistance can be expressed in any

of these phases, overall resistance is thought to reflect the sum of active components in an isolate-cultivar combination (Zadoks and Schein, 1979).

Over the past few years, there has been an increasing awareness of the importance of the components of partial resistance to disease as resistance mechanisms and as impediments to the development of epidemics (Cunfer et al., 1988; Jeger et al., 1983; Jones, 1985; Parlevliet, 1979). Some authors have suggested that the most precise approach to an evaluation of disease resistance is the measurement of individual components of partial resistance (Cunfer et al., 1988; Jeger et al., 1983; Jones, 1985). These components have been evaluated occasionally as parameters to screen for wheat with resistance to STB (Brokenshire, 1976; Gough, 1978; Shearer and Smith, 1978).

Literature Cited

Baker, C. J. 1970. Varietal reaction of wheat to leaf infection by *Leptosphaeria nodorum* and *Septoria tritici*. Trans. Br. Mycol. Soc. 54: 500-504.

Beach, W. S. 1919. Biologic specialization in the genus *Septoria*. Am. J. Botany 6: 1-33.

Benedict, W. G. 1971. Differential effect of light intensity on the infection of wheat by *S. tritici* Desm. under controlled environment conditions. Physiol. Pl. Path. 1: 55-66.

Brokenshire, T. 1975. The role of graminaceous species in the epidemiology of *Septoria tritici* on wheat. Pl. Path. 24: 33-38.

Brokenshire, T. 1976. The reaction of wheat genotypes to *S. tritici*. Ann. Appl. Biol. 82: 415-423.

Brown, A. G. P., and Rosielle, A. A. 1980. Prospects for control of *Septoria*. J. Agric. 21: 8-11.

Brownell, K. H., and Gilchrist, D. G. 1979. Assessment of yield loss in wheat cultivars infected by *Septoria tritici* in California. Phytopathology 69: 1022-1023.

Cunfer, B. M., Stooksbury, D. E., and Johnson, J. W. 1988. Components of partial resistnce to *Leptosphaeria nodorum* among seven soft red winter wheats. Euphytica 37: 129-140.

Danon, T., and Eyal, Z. 1986. The inheritance of resistance in spring and winter bread wheats to two isolates of *Mycosphaerella graminicola*. Phytopathology 76: 1098 (Abstr.).

Djerbi, M., Ghodbane, A. G., Daaloul, A., and Varughese, G. 1974. Identification des sources de résistance à la septoriose du blé (*S. tritici* Rob. et Desm.). Ann. Phytopathol. 6: 495-496.

Djerbi, M., Ghodbane, A. G., Daaloul, A., and Varughese, G. 1976. Studies on the septoria leaf blotch disease of wheat: search for resistance germplasm to *S. tritici* Rob. and Desm. Poljopr. Znan. Smotra 39: 137-142.

Djerbi, M., Hdidar, S., Hafsa, M., et Ben Brahim, M. 1977. Epidémiologie du *Septoria tritici* Rob. et Desm. Conservation et mode de formation de l'inoculum primaire. Société Française de Phytopathologie pp: 91-101.

Eyal, Z. 1971. The kinetics of pycnidiospore liberation in *S. tritici*. Can. J. Bot. 49: 1095-1099.

Eyal, Z. 1972. Effect of septoria leaf blotch on the yield of spring wheat in Israel. Plant Dis. Rep. 56: 983-986.

Eyal, Z. 1981. Integrated control of septoria diseases of wheat. Plant Dis. 65: 763-768.

Eyal, Z., and Brown, M. B. 1976. A quantitative method for estimating density of *S. tritici* pycnidia on wheat leaves. Phytopathology 66: 11-14.

Eyal, Z., and Levy, E. 1987. Variatons in pathogenicity patterns of *M. graminicola* within *Triticum* spp. in Israel. Euphytica 36: 237-250.

Eyal, Z., Wahl, I., and Prescott, J. M. 1983. Evaluation of germplasm response to septoria leaf blotch of wheat. Euphytica 32: 439-446.

Eyal, Z., Scharen, A. L., Huffman, M. D., and Prescott., J. M. 1985. Global insights into virulence frequencies of *M. graminicola*. Phytopathology 75: 1456-1462.

Eyal, Z., Scharen, A. L., Prescott, J. M., and Van Ginkel, M. 1987. The septoria diseases of wheat: Concepts and methods of disease management. Mexico, D.F.: CIMMYT. 52 pp., 17 fig., 20 color plates.

Forni, C., and Zitelli, G. 1979. Preliminary results of studies on resistance sources to *Septoria tritici* in wheat. Genet. Agr. 33: 383-392.

Fournet, J. 1969. Propriétés et rôle du cirrhe du *Septoria nodorum* Berk. Ann. Phytopathol. 1: 87-94.

Gough, F. J. 1978. Effect of wheat host cultivars on pycnidiospore production by *Septoria tritici*. Phytopathology 68: 1343-1345.

Gough, F. J., and Merkle, O. G. 1977. The effect of leaf blotch on root and shoot development of wheat. Plant Dis. Rep. 61: 597-599.

Gough, F. J., and Smith, E. L. 1985. A genetic analysis of *T. aestivum* "Vilmorin" resistance to speckled leaf blotch and Pyrenophora tan spot. Page 36. *In*: Septoria of Cereals. Proc. of the Workshop. A.L. Scharen, ed. Bozeman, MT. 2-4 Aug. 1983. MSU. 116 pp.

Gough, F. J., and Tuleen, N. 1979. Septoria leaf blotch resistance among *Agropyron elongatum* chromosomes in *T. aestivum* 'Chinese' spring. Cereal Res. Commum. 7(4): 275-280.

Hess, D. E., and Shaner, G. 1985. Effect of moist period duration on septoria tritici blotch of wheat. Pages 70-73 *In*: Septoria of Cereals. Proc. of the Workshop. A.L. Scharen, ed. Bozeman, MT. 2-4 Aug. 1983. MSU. 116 pp

Hess, D. E., and Shaner, G. 1987. Effect of moisture and temperature on development of septoria tritici blotch in wheat. Phytopathology 77: 215-219.

Hilu, H. M., and Bever, W. M. 1957. Inoculation, oversummering, and suscept-pathogen relationship of *S. tritici* on *Triticum* spp. Phytopathology 47: 474-480.

Holmes, S. J. I., and Colhoun, J. 1975. Straw-borne inoculum of *T. nodorum* and *S. tritici* in relation to incidence of disease on wheat plants. Pl. Pathol. 24: 63-66.

Jeger, M. J., Jones, D. G., and Griffiths, E. 1983. Components of partial resistance of wheat seedling to *S. tritici*. Euphytica 32: 575-584.

Jlibene, M. 1990. Inheritance of resistance to septoria tritici blotch (*Mycosphaerella graminicola*) in hexploid wheat. PhD. Thesis. Univ. Missouri-Columbia. 86 pp.

Jones, D. G. 1985. Partial resistance, cultivar mixture, and epidemic developpement in the *Septoria nodorum*-wheat association. Pages 1-8 *In*: Septoria of Cereals. Proc. of the Workshop. A. L. Scharen, ed. Bozeman, MT. 2-4 Aug. 1983. MSU. 116 pp.

Kampmeijer, R. 1981. Epidat-data analysis for disease nurseries. Report of a joint project CIMMYT, Mexico. The Research Institute of Plant Protection (IPO), and the Department of Phytopathol. of the Agr. Univ., Wageningen, the Netherlands. 33 pp.

King, J. E., Cook, R. J., and Melville, S. C. 1983. A review of septoria disease of wheat and barley. Ann. Appl. Biol. 103: 345-373.

Krupinsky, J. M., Craddock, J. C., and Scharen, A. L. 1977. Septoria resistance in wheat. Plant Dis. Rep. 61: 632-636.

Lee, T. S., and Gough, F. J. 1984. Inheritance of septoria leaf blotch (*S. tritici*) and Pyrenophora tan spot (*P. tritici repentis*) resistance in *T. aestivum* cv. Carifen 12. Plant Dis. 68: 848-851.

Luthra, J. C., Sattar, A., and Ghani, M. A. 1938. Perpetuation and control of Septoria disease of wheat in the Punjab. Rev. Appl. Mycol. 17: 383.

Mackie, W. W. 1929. Resistance to *S. tritici* in wheat. Phytopathology 19: 1139-1140.

Madariaga, R. B., Gilchrist, D. G., and Martensen, A. N. 1989. Presence and role of *Mycosphaerella graminicola* in California wheat. Phytopathology 79: 1141 (Abstr.).

Malcolm, H. 1978. A host-specific toxin extracted from *S. tritici*. Pages 30-31. *In*: Proc. Aust. Septoria Workshop. Sept. 26-28, 1978. C.E.May, G.M.Murray, and J. Kuiper, eds. Agr. Res. Inst., Dept. of Agr., Wagga, Wagga, N.S.W., Australia. 65 pp.

Mann, C. E., Rajaram, S., and Villareal, R. L. 1985. Progress in Breeding for *S. tritici* resistance in semidwarf spring wheat at CIMMYT. Pages 22-26. *In*: Septoria of Cereals. Proc. of the Workshop. A. L. Scharen, ed. Bozeman, MT. 2-4 Aug., 1983. MSU. 116 pp.

May, C. E. 1983. Triticales as a source of resistance to speckled leaf blotch of wheat. *In*: Australian Plant Breeding Conference, Adelaide, 14-18 February, 1983. pp 114-115.

Metha, Y. R. 1989. Occurrence of *Septoria tritici* and its perfect state in Brazil. Pages 34-35. *In* Septoria of Cereals. Proc. of the workshop. P. M. Fried, ed. Zurich, Switzerland. July 4-7, 1989. 189 pp.

Narvaez, I., and Caldwell, R. M. 1957. Inheritance of resistance to leaf blotch of wheat caused by *S. tritici*. Phytopathology 47: 529-530.

Parlevliet, J. E. 1979. Components of resistance that reduce the rate of epidemic development. Ann. Rev. Pytopathol. 17: 203-222.

Patterson, F. L., Roberts, J. J., Finney, R. E., Shaner, G. E., Gallun, R. L., and Ohm, H. W. 1974. Oasis soft red winter wheat, resistant to septoria leaf blotch. Indiana Agr. Exp. Stn. Bull. 40. 5 pp.

Renfro, B. L., and Young, H. C. J. 1956. Techniques for studying varietal response to septoria leaf blotch of wheat. Phytopathology 46: 23-24.

Rillo, A. O., and Caldwell, R. M. 1966. Inheritance of resistance to *S. tritici* in *T. aestivum* subsp. "Bulgaria 88". Phytopathology 56:897.

Rillo, A. O., Caldwell, R. M., and Glover, D. V. 1970. Cytogenetics of resistance to wheat leaf blotch (*S. tritici*) in backcross derivatives of an Agrotricum line. Crop Sci. 10: 223-227.

Rosen, H. R. 1947. Breeding wheat to combine resistance to leaf rust, speckled leaf blotch, and glume blotch. Phytopathology 37: 524-527.

Rosielle, A. A. 1972. Sources of resistance in wheat to speckled leaf blotch caused by *S. tritici*. Euphytica 21: 152-161.

Rosielle, A. A., and Brown, A. G. P. 1979. Inheritance, heritability and breeding behaviour of three sources of resistance to *S. tritici* in wheat. Euphytica 28: 385-392.

Saâdoui, E. M. 1975. Contribution à l'étude des septorioses du blé au Maroc. Memoire de 3 cycle presenté à l'Institut Agronomique et Veterinaire Hassan II, Rabat, Morocco.

Saâdaoui, E. M. 1987. Physiologic specialization of *Septoria tritici* in Morocco. Plant Disease 71: 153-155.

Saari, E. E., and Prescott, J. M. 1975. A scale for appraising the foliar intensity of wheat diseases. Plant Dis. Rep. 59: 377-380.

Saari, E. E., and Wilcoxson, R. D. 1974. Plant disease situation of high-yielding dwarf wheats in Asia and Africa. Ann. Rev. Phytopathol. 12: 49-68.

Sanderson, F. R., Scharen, A. L., and Scott, P. R. 1985. Sources and importance of primary infection and identities of associated propagules. Pages 57-64. *In*: Septotria of Cereals. Proc. of the Workshop. A. L. Scharen, ed. Bozeman, MT. 2-4 Aug. 1983. MSU. 116 pp.

Santiago, J. C. 1970. Resultado das observacoes effectuadas en Marroc ena Tunisia respeifantes as doencas e pragas does cereals, principalmente em relacao aos trigos Mexicanos cultivados nestes paises: missoes de estudo relizados em 1969 a expensas das Missoes Americanas de auxilio tecnico a Marracos e Tunisia. Elvas, Portugal, Estacao de Melharamente de Plantas. 22 pp.

Scharen, A. L., ed. 1985. Septoria of Cereals. Proc. of the Workshop. Bozeman, MT. 2-4 Aug. 1983. MSU. 116 pp.

Scharen, A. L., and Eyal, Z. 1980. Measurement of quantitative resistance to *S. nodorum* in Wheat. Plant Dis. 64: 492-496.

Scharen, A. L., and Eyal, Z. 1983. Analysis of symptoms on spring and winter wheat cultivars inoculated with different isolates of *S. nodorum*. Phytopathology 73: 143-147.

Scharen, A. L., and Sanderson, F. R. 1985. Identification, distribution and nomenclature of *Septoria* spp. Pages 37-41. *In*: Septoria of Cereals. Proc. of the Workshop. A. L. Scharen, ed. Bozeman, MT. 2-4 Aug. 1983. MSU. 116 pp.

Schluter, K., and Janati, A. 1976. Les septorioses du blé au Maroc. Phytopathologia Mediterranea 15: 7-13.

Sewell, W. O., and Caldwell, R. M. 1960. Use of benzimidazole and excised wheat seedling leaves in testing resistance to *S. tritici*. Phytopathology 50: 654 (Abstr.)

Shaner, G., and Finney, R. E. 1976. Weather and epidemics of septoria leaf blotch of wheat. Phytopathology 66: 781-785.

Shearer, B. L., and Smith, L. D. J. 1978. Studies of the epidemiology of *S. nodorum* and *S. tritici* in Western Australia. Pages 11-14. *In*: Proc. Aust. Septoria Workshop. Sept. 26-28, 1978. C.E.May, Murray, and J.Kuiper, eds. Agr. Res. Inst. Dept. Agr., Wagga Wagga, N.S.W., Australia. 65 pp.

Shipton, W. A., Boyd, W. R. J., Rosielle, A. A., and Shearer, B.L. 1971. The common septoria diseases of wheat. Bot. Rev. 27: 231-262.

Stewart, D. M., Hafiz, A., and Abdelhak, T. 1972. Disease epiphytotic threats to high-yielding and local wheats in the Near East. FAO. Plant Protection Bull. 20: 50-57.

Tyagi, P. D., Joshi, L. M., and Renfro, B. L. 1969. Reaction of wheat varieties to *S. tritici* and report of an epidemic in North-Western Punjab. Ind. Phytopathology 22: 175-178.

Van Ginkel, M. 1986. Inheritance of resistance in wheat to *Septoria tritici*. Ph D thesis. MT. St. Univ. Bozeman. 102 pp.

Van Ginkel, M., and Scharen, A. L. 1987. Generatıon mean analysis and heritabılities of resistance to *S. tritici* in durum wheat. Phytopathology 77: 1629-1633.

Van Ginkel, M., and Scharen, A. L. 1988. Host-pathogen relationshıps of wheat and *S. tritıcı*. Phytopathology 78: 762-766.

Weber, G. F. 1922. Septoria dıseases of wheat. Phytopathol. 12: 537-585.

Wilson, R. E. 1985. Inheritance of resistance to *Septoria tritici* in wheat. Pages 33-35. *In*: Septorıa of Cereals. Proc. of the Workshop. A. L. Scharen, ed. Bozeman, MT. 2-4 Aug. 1983. MSU. 116 pp.

Yechilevich-Auster, M., Levy, E., and Eyal, Z. 1983. Assessment of interactions between cultivated and wild wheats and *S. trıtici*. Phytopathology 73: 1077-1083.

Zadoks, J.C., and Schein, R.D. 1979. Epidemiology and Plant Dısease Management. Ch. 3. Oxford Unıv. Press. NY. 427 pp.

CHAPTER III

COMPONENTS OF PARTIAL RESISTANCE TO SEPTORIA TRITICI BLOTCH

Abstract

Four bread wheat genotypes (BT4, BT6, BT93, and BT323) were evaluated for partial resistance to a single Moroccan isolate (88c) of *S. tritici*. BT93 was the most resistant genotype, differing significantly from the other selections in every experiment in which components of partial resistance were studied, including receptivity, % leaf necrosis, and incubation period. Small pinpoint lesions were the only evidence of infection on BT93 plants inoculated at the second leaf stage. The other three lines were equally susceptible with lesions often coalescing to form irregular patterns of chlorosis and necrosis without pycnidial formation. Receptivity and total number of lesions per gram dry weight in leaves of BT93 were always significantly less than in leaves of the remaining three wheat genotypes. Infected leaves of BT93 contained higher amounts of total chlorophyll per gram leaf tissue and lower amounts of leaf necrosis. Differences in incubation period were detected by determining the time (days) within which 50% of the lesions appeared on each selection. BT93 had the greatest effect on

delaying the appearance of lesions. Resistance of BT93 was consistently superior to the other three genotypes and was associated with lower receptivity, a longer incubation period, and reduced necrotic leaf area. These components complement each other to retard disease development, and indicate that BT93 may be a source of resistance to septoria tritici blotch in breeding programs.

Introduction

Septoria tritici blotch (STB), caused by *Mycosphaerella graminicola* (Fuckel) Schroeter (*Septoria tritici* Rob.ex Desm.), occurs worldwide (Eyal et al., 1987; King et al., 1983; Scharen, 1985; Shipton et al., 1971) with yield losses reported to range from 10-50% (King et al., 1983; Scharen 1985; Shipton et al., 1971). Resistance to STB was noted early in the 20th century (Beach, 1919; Mackie, 1929), but the absence of usable levels of resistance within *Triticum* spp. impeded breeding efforts. However, concerns about the effects of pesticides on environmental quality has led to a renewed interest in breeding for disease resistance, and wheat lines with genetic resistance continue to be sought. Many lines of hexaploid and tetraploid wheats as well as several wild relatives of wheat have shown high levels resistance to STB in field and greenhouse tests under naturally and artificially induced epidemics (Djerbi et al., 1974; Eyal, 1981; Lee & Gough, 1984; Rosielle, 1972; Tyagi et al., 1969; Yechilevich-Auster et al., 1983).

While immunity to STB has not been found in bread wheats (*T. aestivum* L.), many *Trıtıcum* spp. possess effective levels of partial resıstance. Thıs partial resistance appears to be more widely distributed among bread wheat cultivars wıth winter growth habit than among those with sprıng growth habit (Eyal, 1981; Wilson, 1985).

The expression of partial resistance is complex, and consequently so are ıts measurements, because the reactıon of a host cultivar to a pathogen isolate may occur ın some or in all phases of the infection cycle. Thus dıfferences between cultivars tested wıth one ısolate, or differences between isolates tested on one cultivar, may involve spore germınatıon, penetration, colonızation, ıncubatıon perıod, latent period, sporulation, infectious period, and so forth. As resistance can be expressed ın any of these phases, overall resistance is thought to reflect the sum of active components ın an ısolate-cultivar combınatıon (Zadoks & Schein, 1979).

Over the past few years, there has been an increasing awareness of the importance of the components of partıal resıstance to disease as resıstance mechanısms and as ımpedıments to the development of epidemıcs (Cunfer et al, 1988; Jeger et al., 1983; Jones, 1985; Parlevlıet, 1979). Some authors have suggested that the most precise approach to an evaluation of disease resistance is the measurement of ındividual components of partıal resıstance (Cunfer et al., 1988; Jeger et al., 1983; Jones, 1985). These components

have been evaluated occasionally as parameters to screen wheat for resistance to STB (Brokenshire, 1976; Gough, 1978; Shearer & Smith, 1978).

STB, formerly a minor disease in North Africa, is now a major disease problem of wheat in certain countries such as Morocco and Tunisia. This change is due in part to large-scale plantings of introduced high-yielding wheats, and to the occurrence of weather conditions favorable for the development of septoria disease (Djerbi et al., 1974; Saari & Wilcoxson, 1974; Stewart et al., 1972).

The destructiveness of STB in Morocco has been noted periodically since the late 1960's. A severe epidemic occurred in 1968-1969 when the growing season was unusually wet and cool (Santiago, 1970). All high-yielding varieties were susceptible to the disease, particularly Siete Cerros. Saâdaoui (1975) found *S. tritici* to be the predominant species, and Schluter & Janati (1976) reported losses of up to 18% in fungicide experiments. Thus, during the last decade STB has become a significant wheat disease in Morocco (Burleigh et al., 1991; Saâdaoui, 1987). An intensive effort to identify sources of resistance and incorporate this resistance into Moroccan wheats has been undertaken by the Moroccan Wheat Breeding Program. Several sources of resistance and potential varieties were introduced to reduce the genetic vulnerability that results from reliance on a few resistance sources (Jlibene, 1990). This resistance was further increased by selection.

The purpose of this study was to investigate some of the components of partial resistance to *S. tritici* in selected Moroccan bread wheat genotypes with diverse sources of resistance to STB.

Materials and Methods

Experimental Material

Four selected bread wheat genotypes (BT4, BT6, BT93, and BT323), with diverse genetic backgrounds, were obtained from the Moroccan Wheat Breeding Program for use in this study (Table I).

A single Moroccan isolate of *S. tritici*, designated 88c, was obtained from A. L. Scharen (SEA, USDA, Plant Pathology, Montana State University, Bozeman, MT 59715) and used to provide inoculum in all experiments. This isolate was selected because preliminary inoculations conducted with 88c and two other isolates from Oklahoma gave similar disease severity ratings on wheat.

Experimental Procedures

Seeds (50 per genotype) were germinated in a soil mix (containing sand, peat moss, and top soil in equal proportions by volume) in 5 cm clay pots. Seedlings were transplanted after they reached the first leaf stage into 15.2 cm pots (four seedlings per pot; nine pots per wheat genotype). Plants were grown in a growth chamber (model 15, Environmental Growth Chambers, Chagrin Falls, Ohio, U.S.A.)

TABLE I

LIST OF PEDIGREES OF FOUR SELECTED MOROCCAN WHEAT GENOTYPES

Genotypes	Pedigrees
BT4	Bobwhite's'= Bow's' CM33203-K-9M-9Y-4M-4Y-1M-0Y-1PTZ.
BT6	IAS20* 5/H567.71 CMH79.243-1Y-5B-4Y-1B-0Y-0PTZ
BT93	Thornbird's'/3/Burgus(2)/sort 12.13// Kal/Bb SWM 85-29-S3-S1.
BT323	RPB 709.71/COC SWM//Bow's' 85-74-S2-S2.

at 27 C, 70% relatıve humidity, and an 18 hr photoperıod (800 μE m^{-2} sec^{-1}).

When ınoculum was needed, cultures of ısolate 88c were transferred to 250-ml Erlenmeyer flasks containing 100 ml of lıquid medıum (10 g yeast extract, 10 g sucrose ın 1 L dıstılled water), placed on a shaker, and incubated at 21 C under continuous light provided by cool white fluorescent tubes. After 5-6 days, 1-2 ml of broth were transferred to each of several petri dıshes that contaıned fresh yeast-malt extract agar (4 g yeast extract, 4 g malt extract, 4 g sucrose, 15 g agar ın 1 L dıstılled water) and ıncubated for one week at 21 C in an incubator (model 1-35 LL, Percıval Instruments, Boone, Iowa, U.S.A.) under continuous fluorescent light. Under these conditions, *S. tritici* formed typıcal yeast-like colonies composed of budding conıdıa. A spore suspension was prepared by adding 3-5 ml sterile dıstılled water to each plate and gently scrapıng the agar surface with a rubber spatula. The resultıng suspensıon was strained through two-layers of cheese cloth and glasswool to remove the small mats of mycelium that ınterfered wıth inoculation of the plants. The concentratıon was adjusted to 10^7 spores per ml with an hemacytometer, and an all-purpose surfactant (Amway Corp., Ida, MI 49201) was added (0.3 ml/L) to the suspensıon as a wetting agent (Farıh et al., 1991).

Seedlings were inoculated shortly, after the second leaf was fully unfurled. Inoculum was atomized onto seedlings by means of a hand held utility paınt sprayer.

Each block of plants was sprayed as uniformly as possible from all directions until incipient run-off. Controls were sprayed only with water + surfactant. Seedlings were placed in a mist chamber equipped to provide a water-saturated atmosphere that is conducive to infection. After 96 hr, plants were moved to a growth chamber (model 15, Environmental Growth Chambers, Chagrin Falls, Ohio, U.S.A.) maintained at 21 C with an 18 hr photoperiod (800 μm m^{-2} sec^{-1}), with over 95% relative humidity. The experimental design was a randomized complete block with four genotypes as treatments and three blocks with eight subsamples per combination.

Incubation period (IP) data were collected in accordance with the operational definition given by Shaner (1980). To ascertain IP, the number of lesions per leaf surface (first & second leaves) was recorded within 18-22 days of inoculation. Lesions were marked by a small dot on alternate days with an indelible pen at each assessment period. Only the new visible lesions were marked and counted during subsequent assessment periods. Using the Statistical Analysis System (SAS Institute, Inc., Cary, North Carolina), lesion numbers at each assessment period were divided by the final number of lesions for each treatment combination, and the proportions of lesions visible, treated as probabilities, were transformed to Z values: Z = probit (proportion). Equations relating time (days) after inoculation to Z were calculated by linear regression.

The equation solved was of the form:
$T = b_0 + b_1 Z$. T_{50} (the number of days required for 50% of the lesions to appear) was readily obtained from the computer print-out of the regression analysis: $T_{50} = b_0$ (y-intercept for Z=0). Receptivity was determined as the final number of lesions visible per square centimeter of leaf tissue. The percentage necrotic leaf area was estimated indirectly by determining the relative water content (RWC) and total chlorophyll content in the leaf tissue. Relative Water Content was determined using the formula:

$$RWC = \frac{\text{fresh weight} - \text{dry weight}}{\text{sat. fresh weight} - \text{dry weight}} \times 100.$$

The arcsin-square root transformation of RWC was used before statistical analysis (Steel & Torrie, 1980). The saturated fresh weight was determined by soaking leaves cut into 2-3 cm lengths, in 20 ml of distilled water in petri plates for 24 hr. The leaf segments were then blot-dried and weighed for their saturated fresh weight, placed in a drying oven at 50 C for 48 hr and weighed again.

Determination of total chlorophyll content in leaf tissue was accomplished by immersing wheat leaf tissue in a test tube containing 10 ml of dimethyl sulfoxide (DMSO) (B.D.H. Chemicals Ltd., Toronto) and incubating in a water bath at 65 C for two hours. A 5 ml sample of chlorophyll extract was transferred to a cuvette and the optical density

(OD) values at 645 and 663 nm were read in a spectrophotometer against a DMSO blank (Hiscox & Israelstan, 1979). The amount of chlorophyll present in the extract was calculated on the basis of milligrams of chlorophyll per gram of leaf tissue extracted according to the following equation (Arnon, 1949):

$$\text{Mg tot. chlor./g tis.} = [20.2(D_{645}) + 8.02(D_{663})] \times \frac{V}{1000xW}$$

where:

D = OD reading of the chlorophyll extract at the specific indicated wavelength.

V = final volume of the DMSO-chlorophyll extract.

W = fresh weight in grams of the wheat tissue extracted.

Results

Four bread wheat genotypes, at the second-leaf stage, were evaluated for partial resistance to *S. tritici* under controlled conditions. The mist chamber-growth chamber conditions were conducive to development of STB. Components of partial resistance studied were total number of lesions per gram dry weight, receptivity, percentage leaf area necrosis, and incubation period. No lesions appeared on noninoculated control plants sprayed with distilled water + surfactant. Differences between BT93 and the other three genotypes were significant in every experiment for all the

components of partial resistance studied. BT93 was the most resistant showing either no lesions or small pinpoint lesions, while BT4, BT6 and BT323 were, in general, equally susceptible with lesions often coalescing to form irregular patterns of chlorosis and necrosis without pycnidial formation. Differences between these susceptible genotypes usually were not significant. The total number of lesions per gram dry weight was significantly lower in leaves of BT93 and higher in the remaining genotypes (Table II).

Differences in receptivity between wheat selections were detectable within 18-20 days of inoculation with BT6 the most receptive and BT93 the least receptive (Table III). Estimated damage caused by *S. tritici* in leaves of the four wheat genotypes studied, as determined by RWC and total chlorophyll content, was high on BT4, BT6 and BT323 and low on BT93 (Tables IV and V). Differences in incubation period on the second leaf were consistent with each genotype through the four experiments (Table VI). Incubation period was significantly longer in BT93 than in the other three genotypes. The range of number of days for incubation period was 5-8 days among the resistant and susceptible genotypes.

Discussion

We used one Moroccan isolate of *S. tritici* to demonstrate the effects of host genotype on components of resistance to STB in selected Moroccan wheat genotypes. All

TABLE II

LESIONS[x] OF SEPTORIA TRITICI BLOTCH ON LEAVES OF FOUR SELECTED MOROCCAN WHEAT GENOTYPES

Genotypes	E[y].1	E.2	E.3
BT4	7269 c[z]	7536 b	5901 b
BT6	3757 b	9314 b	7825 b
BT93	871 a	1756 a	1285 a
BT323	3823 b	7681 b	5096 b

[x] Lesions per gram dry weight of the first and second leaves

[y] Experiment number; Seedlings were inoculated using a concentration of 10^7 spores of *S. tritici* per ml, left in a mist chamber for 96 hr at ≈ 100% RH and then placed in a growth chamber for 18-22 days at 21 C, 90% RH and an 18hr photoperiod (800 μE m^{-2} sec^{-1}).

[z] Means in columns followed by the same letter are not significantly different according to Tukey's W procedure (P=0.05).
Each number represents a mean of 24 counts per treatment.
Noninoculated control seedlings showed no lesions.

TABLE III

RECEPTIVITY[x] AS MEASURED BY NUMBER OF LESIONS PER CM^2 OF LEAF TISSUE OF FOUR SELECTED MOROCCAN WHEAT GENOTYPES

Genotypes	E[y].4	E.5	E.6
BT4	12.8 c[z]	5.2 b	2.7 b
BT6	8.4 b	8.8 c	5.8 c
BT93	2.5 a	1.8 a	0.4 a
BT323	8.1 b	3.2 ab	2.9 b

[x] Receptivity= final number of lesions visible per square centimeter of leaf tissue

[y] Experiment number; Seedlings were inoculated using a concentration of 10^7 spores of *S. tritici* per ml, left in a mist chamber for 96 hr at ≈ 100% RH and then placed in a growth chamber for 18-22 days at 21 C, 90% RH and an 18 hr photoperiod (800 μE m^{-2} sec^{-1}).

[z] Means in columns followed by the same letter are not significantly different according to Tukey's W procedure (P=0.05).
Each number represents a mean of 24 counts per treatment.
Noninoculated control seedlings showed no lesions.

TABLE IV

LEAF DAMAGE ESTIMATED BY RELATIVE WATER CONTENT (%) IN LEAVES OF FOUR SELECTED MOROCCAN WHEAT GENOTYPES

Genotypes	E[x].2	E.3	E.7	Control
BT4	60.4 c[y]	59.2 c	63.6 c	91.4 a
BT6	71.7 b	73.0 b	75.4 b	91.1 a
BT93	88.0 a	86.2 a	89.3 a	92.4 a
BT323	71.1 b	71.0 b	74.6 b	93.6 a

[x] Experiment number; control corresponds to noninoculated seedlings.

[y] Means in columns followed by the same letter are not significantly different according to Tukey's W procedure (P=0.05).
Each number represents a mean of 24 measurements.

TABLE V

LEAF DAMAGE ESTIMATED BY TOTAL CHLOROPHYLL CONTENT[x] IN LEAVES OF FOUR SELECTED MOROCCAN WHEAT GENOTYPES

Genotypes	E[y].4	E.5	E.6	Control
BT4	1.13 b[z]	1.38 b	1.42 b	1.97 a
BT6	1.17 ab	1.47 b	1.43 b	1.94 a
BT93	1.58 a	1.98 a	1.88 a	2.01 a
BT323	1.18 ab	1.52 b	1.49 b	1.96 a

[x] Total chlorophyll content in mg/g leaf tissue.

[y] Experiment number; control corresponds to noninoculated seedlings.

[z] Means in columns followed by the same letter are not significantly different according to Tukey's W procedure (P=0.05).
Each number represents a mean of 24 measurements.

TABLE VI

INCUBATION PERIOD[x] OF SEPTORIA TRITICI BLOTCH IN LEAVES (first & second) OF FOUR SELECTED MOROCCAN WHEAT GENOTYPES

Genotypes	E[y].1	E.2	E.3	E.4
BT4	3.6 c[z]	3.2 b	4.2 b	5.1 b
BT6	4.3 bc	3.6 b	5.1 b	5.7 b
BT93	14.4 a	12.2 a	10.8 a	10.9 a
BT323	5.4 b	3.8 b	4.3 b	5.9 b

[x] Incubation period determined as the time (days) within which 50% of lesions appeared on each wheat genotype.

[y] Experiment number; Seedlings were inoculated using a concentration of 10^7 spores of *S. tritici* per ml, left in a mist chamber for 96 hr at ≈ 100% RH and then placed in a growth chamber for 18-22 days at 21 C, 90% RH and an18 hr photoperiod (800 μE m^{-2} sec^{-1}).

[z] Means in columns followed by the same letter are not significantly different according to Tukey's W procedure (P=0.05).
Each number represents a mean of 24 counts per treatment.
Noninoculated control seedlings showed no lesions.

isolates (three) tested previously on these wheat selections have resulted in the same ranking of wheat cultivars based on lesion number per gram dry weight (Farih and Hunger, 1990).

Under the controlled growth chamber conditions used in the present study, BT93 was consistently more resistant than the other three selections, with significant differences in every experiment for all components of partial resistance studied.

Infected leaves of the resistant cultivar BT93 contained higher amounts of total chlorophyll per gram leaf tissue and lower amounts of leaf necrosis which indicated limited growth of *S. tritici*. In most cases, disease symptoms with plant pathogens are assessed with the assumption that they reflect quantitatively the growth of the pathogen in the host (Parlevliet, 1979).

Although BT6 was the most receptive (Table III), the amount of leaf necrosis in leaves of BT4 (as estimated by RWC) was the highest (Table IV). Lesions in leaves of BT4 were larger and tended to coalesce.

In obligate parasite systems, the latent period has been defined as the time from inoculation to the time when 50% of the lesions (pustules or colonies) are visible (Parlevliet, 1979; Shaner, 1980). However, facultative parasites sporulate on necrotic tissue following moisture periods of sufficient duration. Thus, latent period, as defined above, would be dependent on sufficient periods of

free moisture following the appearance of necrotic leaf areas (Nutter & Pederson, 1985). In the *Septoria* - wheat association, pycnidial formation is favored by relatively long humid periods (Hilu & Bever, 1957; Holmes & Colhoun, 1971; Shaner, 1981; Shearer & Zadoks, 1972; Shipton et al., 1971). In such systems, the incubation period (visible lesions) may provide a measurement that closely parallels the latent period as defined for obligate parasites.

Differences in incubation period were detected by determining the time (days) within which 50% of lesions appeared on each wheat genotype. Delay in the appearance of lesions was greatest in BT93. Differences in incubation period are often assumed to reflect differences in growth rate of the pathogen in the host (Neervoort & Parlevliet, 1978; Parlevliet, 1979). Marked differences in disease severity occurred among *T. aestivum* varieties tested by Brokenshire (1976) and significant differences were also apparent for the incubation period.

Although only four selected Moroccan genotypes were studied, resistance of BT93 was consistently superior to the other three lines as evidenced by lower receptivity, reduced amount of leaf necrosis, and longer incubation period. These components could complement each other to retard disease development, and indicate that BT93 may be a source of resistance to septoria tritici blotch in breeding programs.

Literature Cited

Arnon, D. I. 1949. Copper enzymes in isolated chloroplasts. Polyphenoloxidase in *Beta vulgaris*. Plant Physiol. 24: 1-15.

Beach, W. S. 1919. Biologic specialization in the genus *Septoria*. Am. J. Botany 6: 1-33.

Brokenshire, T. 1976. The reaction of wheat genotypes to *S. tritici*. Ann. Appl. Biol. 82: 415-423.

Burleigh, J. R., Ezzahiri, B., and Roelfs, A. P. 1991. Assessment of cultivar performance and disease impact on cereals in Morocco. Plant Dis. 75: 65-73.

Cunfer, B. M., Stooksbury, D. E., and Johnson, J. W. 1988. Components of partial resistance to *Leptosphaeria nodorum* among seven soft red winter wheats. Euphytica 37: 129-140.

Djerbi, M., Ghodbane, A. G., Daaloul, A., and Varughese, G. 1974. Identification des sources de résistance à la septoriose du blé (*S. tritici* Rob. et Desm.). Ann. Phytopathol. 6: 495-496.

Eyal, Z. 1981. Integrated control of septoria diseases of wheat. Plant Dis. 65: 763-768.

Eyal, Z., Scharen, A. L., Prescott, J. M., and Van Ginkel, M. 1987. The septoria diseases of wheat: Concepts and methods of disease management. Mexico, D.F.: CIMMYT, 52 pp., 17 fig., 20 color plates.

Farih, A., and Hunger, R. M. 1990. Components of partial resistance to *Septoria tritici* in Moroccan wheat. Phytopathology 80: 967 (Abstr.)

Farih, A., Gough, F. J., Hunger, R. M., and Montana, J. R. 1991. Effect of surfactants and wheat leaf leachate on production, germination, and infectivity of *Septoria tritici* conidia. Phytopathology 81: 811 (Abstr.)

Gough, F. J. 1978. Effect of wheat host cultivars on pycnidiospore production by *Septoria tritici*. Phytopathology 68: 1343-1345.

Hilu, H. M., and Bever, W. M. 1957. Inoculation, oversummering, and suscept-pathogen relationship of *S. tritici* on *Triticum* spp. Phytopathology 47: 474-480.

Hiscox, J. D., and Israelstam, G. F. 1979. A method for the extraction of chlorophyll from leaf tissue without maceration. Can. J. Bot. 57: 1332-1334.

Holmes, S. J. I., and Colhoun, J. 1971. Infection of wheat seedlings by *Septoria nodorum* in relation to environmental factors. Trans. Br. Mycol. Soc. 57(3): 493-500.

Jeger, M. J., Jones, D. G., and Griffiths, E. 1983. Components of partial resistance of wheat seedling to *S. nodorum*. Euphytica 32: 575-584.

Jlibene, M. 1990. Inheritance of resistance to septoria tritici blotch (*Mycosphaerella graminicola*) in hexaploid wheat. Ph.D. Thesis. Univ. Missouri-Columbia. 86 pp.

Jones, D. G. 1985. Partial resistance, cultivar mixture, and epidemic development in the *Septoria nodorum*-wheat association. Pages 1-8 *In*: Septoria of Cereals.Proc. of the Workshop. A. L. Scharen, ed. Bozeman, MT. 2-4 Aug. 1983. MSU. 116 pp.

King, J. E., Cook, R. J., and Melville, S. C. 1983. A review of septoria diseases of wheat and barley. Ann. Appl. Biol. 103: 345-373.

Lee, T. S., and Gough, F. J. 1984. Inheritance of septoria leaf blotch (*S. tritici*) and Pyrenephora tan spot (*P. tritici repentis*) resistance in *T. aestivum* cv. Carifen 12. Plant Dis. 68: 848-851.

Mackie, W. W. 1929. Resistance to *Septoria tritici* in wheat. Phytopathology 19: 1139-1140.

Neervoort, W. J., and Parlevliet, J. E. 1978. Partial resistance of barley to leaf rust, *Puccinia hordei*. V. Analysis of the components of partial resistance in eight barley cultivars. Euphytica 27: 33-39.

Nutter, F. W., Jr., and Pederson, V. D. 1985. Receptivity, incubation period, and lesion size as criteria for screening barley genotypes for resistance to *Pyrenophora teres*. Phytopathology 75: 603-606.

Parlevliet, J. E. 1979. Components of resistance that reduce the rate of epidemic development. Ann. Rev. Phytopathol. 17: 203-222.

Rosielle, A. A. 1972. Sources of resistance in wheat to speckled leaf blotch caused by *S. tritici*. Euphytica 21: 152-161.

Saâdaoui, E. M. 1975. Contribution à l'étude des septorioses du blé au Maroc. Mémoire de 3 cycle presenté à l'Institut Agronomique et Vétérinaire Hassan II, Rabat, Morocco.

Saâdaoui, E. M. 1987. Physiologic specialization of *Septoria tritici* in Morocco. Plant Dis. 71: 153-155.

Saari, E. E., and Wilcoxson, R. D. 1974. Plant Disease situation of high-yielding dwarf wheats in Asia and Africa. Ann. Rev. Phytopathol. 12: 49-68.

Santiago, J.C., 1970. Resultado das observacoes effectuadas en Marroco ena Tunisia respeifantes as doencas e pragas does cereals, principalmente em relacao aos trigos Mexicanos cultivados nestes paises: missoes de estudo relizados en 1969 a expensas das Missoes Americanas de auxiliotecnico a Marracos e Tunisia. Elvas, Portugal, Estacao de Melharamente de Plantas. 22 pp.

SAS Institute. 1982. SAS User's Guide: Statistics. SAS institute, Cary, NC. 584 pp.

Scharen, A. L., ed. 1985. Septoria of Cereals. Proc. of the Workshop. Bozeman, MT., 2-4 Aug. 1983 MSU. 116 pp.

Schluter, K., and Janati, A. 1976. Les septorioses du blé au Maroc. Phytopathologia Mediterranea 15: 7-13.

Shaner, G. 1980. Probits for analyzing latent period data in studies of slow-rusting resistance. Phytopathology 70: 1179-1182.

Shaner, G. 1981. Effect of environment on fungal leaf blights of small grains. Ann. Rev. Phytopathol. 19: 273-296.

Shearer, B. L., and Smith, L. D. J. 1978. Studies of the epidemiology of *S. nodorum* and *S. tritici* in Western Australia. Pages 11-14 *In*: Proc. Aust. Septoria Workshop. Sept. 26-28, 1978. C.E. May, G.M. Murray, and J. Kuiper, eds. Agr. Res. Inst., Dept. Agr., Wagga Wagga, N.S.W., Australia. 65 pp.

Shearer, B. L., and Zadoks, J. C. 1972. The latent period of *Septoria nodorum* in wheat. I. the effect of temperature and moisture treatments under controlled conditions. Neth. J. Pl. Path. 78: 231-241.

Shipton, W. A., Boyd, W. R. J., Rosielle, A.A., and Shearer, B.L. 1971. The common septoria diseases of wheat. Bot.Rev. 27: 231-262.

Steel, R. G. D., and Torrie, J.H. 1980. Principles and Procedures of Statistics. Second ed. Mc Graw-Hıll, New York. 633 pp.

Stewart, D. M., Hafız, A., and Abdelhak, T. 1972. Dısease epıphytotic threats to hıgh-yıeldıng and local wheats in the Near-East. FAO Plant Protection Bull. 20: 50-57.

Tyagı, P. D., Joshi, L. M., and Renfro, B. L. 1969. Reaction of wheat varieties to *S. tritici* and report of an epidemic in North-Western Punjab. Ind. Phytopathol. 22: 175-178.

Wilson, R. E. 1985. Inheritance of resistance to *S. tritici* in wheat.Pages 33-35. *In*: Septoria of Cereals. Proc. of Workshop. A. L. Scharen, ed. Bozeman, MT. 2-4 Aug. 1983. MSU. 116 pp.

Yechilevich-Auster, M., Levy, E., and Eyal, Z. 1983. Assessment of interactions between cultivated and wild wheats and *S. trıtici*. Phytopathology 73: 1077-1083.

Zadoks, J. C., and Schein, R. D. 1979. Epidemiology and Plant Disease Management. Ch 3. Oxford Univ. Press. New York. 427 pp.

CHAPTER IV

MODE OF INHERITANCE OF SEEDLING PLANT RESISTANCE TO SEPTORIA TRITICI BLOTCH IN SPRING WHEATS

Abstract

Inheritance of seedling plant resistance to septoria tritici blotch (STB) was studied in a cross of two spring wheat selections, resistant BT93 and susceptible BT4. The parent genotypes, and the F_1, F_2, and F_3 generations were inoculated with conidia of Moroccan isolate 88c of *Septoria tritici*. Individual plants were classed as resistant or susceptible based on percentage of necrotic tissue that developed in inoculated leaves after incubation. F_3 families were classed as either homozygous resistant, segregating, or homozygous susceptible. The reaction of F_1 plants and segregating ratios in F_2 plants and F_3 families indicated that resistance was conditioned by two independent dominant genes.

Introduction

Mycosphaerella graminicola (Fuckel) Schroeter (Anamorph: *Septoria tritici* Rob. ex Desm.) causes septoria tritici blotch (STB) of wheat. The disease occurs in most major

wheat producing areas of the world and the pathogen is omnipresent even where conditions are only marginally conducive (Eyal et al., 1987; King et al., 1983; Shipton et al., 1971). STB can be pernicious and yield losses of slight to 60% have been attributed to natural infection (Brownell & Gilchrist, 1979; Eyal, 1972; Eyal et al., 1987; King t al., 1983; Shipton et al., 1971). Reports of increased occurrence and disease levels have elevated *S. tritici* to a pathogen of worldwide importance and have resulted in numerous research and crop improvement programs worldwide (Brown & Rosielle, 1980; Djerbi et al., 1974; Eyal, 1972; Forni & Zitelli, 1979; Gough & Smith, 1985; Mann et al., 1985; Metha, 1989; Scharen, 1985; Tyagi et al., 1969; Van Ginkel, 1986).

The increase in importance of the disease is largely due to the widespread replacement of tall, somewhat resistant cultivars with high-yielding, early maturing, short-strawed wheat cultivars (Djerbi et al., 1974; Saari and Wilcoxson, 1974; Stewart et al., 1972). Since the currently grown high-yielding wheat cultivars are disturbingly vulnerable to STB, the incorporation of resistance to *S. tritici* remains a high-priority breeding objective, worldwide.

Although incorporation of resistance to *S. tritici* into cultivated wheat is an objective in many breeding programs and several sources of resistance have been reported, little is known about the number and the nature of genes

controlling resistance and how they relate to each other. Monogenic resistance was reported to be recessive in an unnamed variety (Mackie, 1929), and to be dominant in cultivars Lerma 50 and P14 (Narvaez & Caldwell, 1957), Bulgaria 88 (Rillo & Cadwell, 1966), and Veranopolis and IAS-20 (Rosielle & Brown, 1979). Other reported modes of inheritance include duplicate dominant genes (Wilson, 1985) and single, incompletely dominant genes (Lee & Gough, 1984; Wilson, 1985). Digenic additive resistance was reported in cultivar Nabob (Narvaez & Caldwell, 1957), while two dominant genes appeared to control resistance of four winter wheats tested by Danon & Eyal (1986) to isolate ATCC 48507 of *M. graminicola*. Rosielle & Brown (1979) showed that resistance in the cultivar Seabreeze is determined by recessive genes at no less than three loci.

Severe outbreaks of septoria diseases have occurred in arid and semi-arid countries along the Mediterranean seacoast. In Morocco, a severe epidemic during the 1968-1969 season highlighted the potential importance of STB (Saari and Wilcoxson, 1974; Stewart et al., 1972). As a consequence, studies investigating the importance of the *Septoria* species and yield losses due to STB have been initiated (Saâdaoui, 1975; Schluter & Janati, 1976), and STB was found to adversely affect a significant portion of wheat production (Burleigh et al., 1991; Saâdaoui, 1987). As a result, wheat lines with genetic resistance are being sought, and a number of bread wheat cultivars with diverse

sources of resistance were introduced and their resistance was further increased by selection (Jlibene, 1990).

This paper reports the mode of inheritance of seedling resistance to STB in a cross of a resistant wheat genotype, designated BT93, with a susceptible genotype, designated BT4. Both wheat lines are new introductions to the Moroccan Wheat Breeding Program from the International Maize and Wheat Improvement Center (CIMMYT) headquartered in Mexico.

Materials and Methods

Two spring wheat lines, BT93 (Thornbird 'S'/3/ Burgus(2)/ sort 12.13//Kal/Bb 85-29-S3-S1) and BT4 (Bow'S' Cm 33203-k-9M-9Y-4M-4Y-1M-0Y-1PTZ), with contrasting reactions to *S. tritici* (Farih & Hunger, 1990) were used as parents in this study. Crosses between the cultivars were made in the greenhouse in 1989. Seeds produced by self-pollination of F_1 plants were composited to provide the F_2 generation, while seeds from individual F_2 plants provided F_3 families. Seedlings representing F_1, F_2, and F_3 generations were grown in a growth chamber (model 15, Environmental Growth Chambers, Chagrin Falls, Ohio, U.S.A.) maintained at 27 C, with an 18 hr-light period and 70% relative humidity. The two parents were always grown as checks next to the seedlings of each filial generation .

Inoculum preparation and application was essentially the same for all tests. Cultures of *S. tritici* isolate 88c

were grown first in 250 ml flasks containing 100 ml of liquid medium (10 g yeast extract, 10 g sucrose in 1 L of distilled water) for one week on a shaker at 21 C under continuous light provided by cool white fluorescent tubes. Plastic petri plates containing freshly prepared yeast malt extract agar (4 g yeast extract, 4 g malt extract, 4 g sucrose, 15 g agar in 1 L distilled water) were inoculated with 1 ml of liquid shake culture and incubated under continuous light at 21 C.

S. tritici produced yeast-like colonies composed of budding conidia on the agar surface. Spore suspensions were prepared by adding 2-3 ml of sterile distilled water to each plate and scraping the surface of the medium with a rubber spatula to dislodge the spores. Conidial suspensions derived from several plates were pooled and strained through two-layers of cheesecloth and glasswool. An all-purpose surfactant (Amway Corp., Ida, M1 49201) was added (0.3 ml/L) to the suspension as a wetting agent (Farih et al., 1991), and spore concentration was determined with a hemacytometer. Unless stated otherwise, the inoculum concentration used was adjusted to 10^7 spores/ml.

After the second leaf was fully unfurled, seedlings were inoculated. At this stage of growth, no sign of senescence was apparent on the seedlings including the first leaf. Inoculum was applied with a hand held utility paint sprayer, as uniformly as possible from all directions until incipient run off. Seedlings were then placed in a mist

chamber at 21 C equipped to maintain free moisture on the foliage. After 96 hr seedlings were moved to a growth chamber maintained at 21 C, with an 18 hr light period and at 95% relative humidity.

Visual assessment of symptoms were made when lesions became distinct, which was between seven to 15 days after inoculation. Assessment was conducted by estimating the percent leaf area covered by lesions on an individual seedling basis, on both leaves. In general, seedlings with percent necrotic leaf tissue equal to the resistant parent BT93 (characterized by no visible lesions, small pinpoint lesions, or little necrosis or chlorosis) were classed resistant while those similar to the susceptible parent BT4 (characterized by visible lesions or necrosis or both) were classed as susceptible. The F_3 families were classed as homozygous resistant, segregating, or homozygous susceptible. Seedlings per family ranged from 4-30, with mean of 14.2. Mature F_2 plants became infected with *Pseudomonas* sp. (inoculum source unknown) which severely reduced the quantity and the quality of seed produced. Chi-square tests were used to analyze data from the segregating populations.

Results and Discussion

Seven days after inoculation, lesions developed in the inoculated leaves of the susceptible parent BT4. Within 15 days after inoculation, the lesions coalesced to form

irregular patterns of chlorosis and necrosis. Either no visible or very small pinpoint lesions developed in leaves of BT93. Reaction of F_1 plants were similar to the reaction of BT93, the resistant parent. The absence of lesion development in the F_1 seedlings indicated that resistance of BT93 to culture 88c of *S. tritici* was conditioned by one or more dominant gene(s).

A total of 167 F_2 plants of BT93 x BT4 were classified for reactions to *S. tritici* (Table VII). Of the total, 154 seedlings were classified as resistant as BT93 and F_1 seedlings, and 13 developed large, spreading lesions similar to those in BT4. These numbers were a close fit to a 15:1 ratio expected when two independent dominant genes are segregating.

Mature F_2 plants became infected with *Pseudomonas* sp. (inoculum source unknown) which severely reduced the quantity and quality of seed produced. Consequently, only 64 F_3 families from tested F_2 plants were available for testing. The number of plants within segregating F_3 families frequently was too small to confidently separate those families segregating in a 3:1 ratio from those segregating in a 15:1 ratio. Thus, reactions of the F_3 families were classed into three groups as follows: 29 homozygous resistant, 31 segregating, and 4 homozygous susceptible (Table VII). This segregation was a close fit (P=0.75-0.90) to a 7:8:1 ratio (Table VIII) expected for two independent dominant genes that condition similar phenotypes. This

TABLE VII

SEGREGATION FOR RESISTANCE AND SUSCEPTIBILITY TO *SEPTORIA TRITICI* AMONG F_2 PLANTS AND F_3 FAMILIES FROM A CROSS OF RESISTANT GENOTYPE BT93 WITH SUSCEPTIBLE GENOTYPE BT4

Parents and filial generations	Numbers of F_2 plants and F_3 families			
	Resist	Int. (F_2) Seg. (F_3)	Susc.	P[x]
BT4	0	0	20	
BT93	20	0	0	
F_1	60	0	0	
F_2	154	0	13	0.25-0.50
F_3[y,z]	29	31	4	0.75-0.90

[x] Probability of fit to a 15:1 ratio of resistant and susceptible F_2 plants and to a 7:8:1 ratio of homozygous resistant, segregating, and homozygous susceptible F_3 families.

[y] Among 167 F2 plants tested only 76 produced seed because of *Pseudomonas* contamination that occurred in growth chambers. When planted only 64 F_3 families grew and were inoculated.

[z] Plants were rated twice within 7 and 15 days of inoculation by visual observation of lesions and comparing them to the two parents: resistant and susceptible.

TABLE VIII

OBSERVED REACTIONS OF F_2 PLANTS AND F_3 FAMILIES OF BT93 x BT4 INOCULATED WITH ISOLATE 88c OF *SEPTORIA TRITICI*, AND PROPOSED F_2 GENOTYPES AND SEGREGATION RATIO[x]

Reaction of F_2 plants	F_3	Proposed F_2 genotype	Proportion Of 16
R[y]	Homo. Res.	AABB	1
R	Homo. Res.	AABb	2
R	Homo. Res.	AAbb	1
R	Homo. Res.	AaBB	2
R	Seg. 15:1[z]	AaBb	4
R	Seg. 3:1[z]	Aabb	2
R	Homo. Res.	aaBB	1
R	Seg. 3:1[z]	aaBb	2
S	Homo. Sus.	aabb	1

[x] Based on data (text and Table VII) indicating that resistance was conditioned by two independent dominant genes.

[y] R = resistant, S = susceptible.

[z] Because some F_3 family sizes were too small to differentiate 3:1 and 15:1 ratios (P = 0.05), segregating families were combined in a single class representing 50% (8/16 ths) of all families.

supports the results obtained from tests of the F_2 plants.

Data derived from testing the F_2 and F_3 generations strongly suggest that resistance of BT93 is governed by two dominant genes. These genes confer a high level of resistance manifested by little or no necrosis. Such genetic control has also been reported by Danon and Eyal (1986) who found that resistance of four winter wheats to isolate ATCC 48507 of *M. graminicola* seemed to be controlled by two dominant genes.

Previous studies of host resistance have been conducted in the field where qualitative expression of resistance was difficult to detect. This may account for failure to identify other similarly controlled resistance.

Results from these experiments indicate that BT93 may provide effective resistance against STB. However, these experiments were conducted using only one isolate of *S. tritici*, and testing with additional *S. tritici* isolates is needed before BT93 should be considered as a source of resistance to STB.

Literature Cited

Brown, A. G. P., and Rosielle, A. A. 1980. Prospects for control of *Septoria*. J. Agric. 21: 8-11.

Brownell, K. H., and Gilchrist, D. G. 1979. Assessment of yield loss in wheat cultivars infected by *Septoria tritici* in California. Phytopathology 69: 1022-1023.

Burleigh, J. R., Ezzahiri, B., and Roelfs, A. P. 1991. Assessment of cultivar performance and disease impact on cereals in Morocco. Plant Dis. 75: 65-73.

Danon, T., and Eyal, Z. 1986. The inheritance of resistance in spring and winter bread wheats to two isolates of *Mycosphaerella graminicola*. Phytopathology 76: 1098 (Abstr.)

Djerbi, M., Ghodbane, A. G., Daaloul, A., and Varughese, G. 1974. Identification des sources de résistance à la septoriose de blé (*S. tritici* Rob. et Desm.). Ann. Phytopathol. 6: 495-496.

Eyal, Z. 1972. Effect of septoria leaf blotch on the yield of spring wheat in Israel. Plant Dis. Rep. 56: 983-986.

Eyal, Z., Scharen, A. L., Prescott, J. M., and Van Ginkel, M. 1987. The septoria diseases of wheat: Concepts and methods of disease management. Mexico, D.F.: CIMMYT. 52 pp., 17 figures. 20 color plates.

Farih, A., and Hunger, R. M. 1990. Components of partial resistance to *Septoria tritici* in Moroccan wheat. Phytopathology 80: 967 (Abstr.)

Farih, A., Gough, F. J., Hunger, R. M., and Montana, J. R. 1991. Effect of surfactants and wheat leaf leachate on production, germination, and infectivity of *Septoria tritici* conidia. Phytopathology 81: 811 (Abstr.)

Forni, C., and Zitelli, G. 1979. Preliminary results of studies on resistance sources to *Septoria tritici* in wheat. Genet. Agr. 33: 383-392.

Gough, F. J., and Smith, E. L. 1985. A genetic analysis of *T. aestivum* "Vilmorin" resistance to speckled leaf blotch and Pyrenophora tan spot. Page 36. *In* Septoria of Cereals. Proc. of the Workshop. A.L. Scharen, ed. Bozeman, MT. 2-4 Aug. 1983. MSU. 116 pp.

Jlibene, M. 1990. Inheritance of resistance to septoria tritici blotch (*Mycosphaerelle graminicola*) in hexaploid wheat. PhD. Thesis. Univ. Missouri- Columbia. 86 pp.

King, J. E., Cook, R. J., and Melville, S. C. 1983. A review of septoria diseases of wheat and barley. Ann. Appl. Biol. 103: 345-373.

Lee, T. S., and Gough, F. J. 1984. Inheritance of septoria leaf blotch (*S. tritici*) and Pyrenophora tan spot (*P.tritici repentis*) resistance in *T. aestivum* cv. Carifen 12. Plant Dis. 68: 848-851.

Mackie, W. W.1929. Resistance to *S. tritici* in wheat. Phytopathology 19: 1139-1140.

Mann, C. E., Rajaram,S., and Villareal, R. L. 1985. Progress in Breeding for *S. tritici* resistance in semidwarf spring wheat at CIMMYT. pages 22-26. *In* Septoria of Cereals. Proc. of the Workshop. A.L.Scharen, ed. Bozeman, MT 2-4 Aug., 1983. MSU. 116 pp.

Metha, Y. R. 1989 Occurrence of *Septoria tritici* and its perfect state in Brazil. Pages 34-35. *In* Septoria of Cereals. Proc. of the Workshop. P.M. Fried, ed. Zurich, Switzerland. July 4-7, 1989. 189 pp.

Narvaez,I., and Caldwell, R. M. 1957. Inheritance of resistance to leaf blotch of wheat caused by *S. tritici*. Phytopathology 47: 529-530.

Rillo, A. O., and Caldwell, R. M. 1966. Inheritance of resistance to *S. tritici* in *T. aestivum* subsp. "Bulgaria". Phytopathology 56: 897.

Rosielle, A. A., and Brown, A. G. P. 1979. Inheritance, heritability and breeding behaviour of three sources of resistance to *S. tritici* in wheat. Euphytica 28: 385-392.

Saâdaoui, E. M. 1975. Contribution à l'étude des septorioses du blé au Maroc. Mémoire de 3 cycle présenté à l'Institut Agronomique et Vétérinaire Hassan II, Rabat, Morocco.

Saâdaoui, E. M. 1987. Physiologic specialization of *Septoria tritici* in Morocco. Plant Dis. 71: 153-155.

Saari, E. E., and Wilcoxson, R. D. 1974. Plant disease situation of high-yielding dwarf wheats in Asia and Africa. Ann. Rev. Phytopathol. 12: 49-68.

Scharen, A. L., ed. 1985. Septoria of Cereals. Proc. of the Workshop. Bozeman, MT. 2-4 Aug. 1983. MSU. 116 pp.

Schluter, K., and Janati, A. 1976. Les septorioses du blé au Maroc. Phytopathologia Mediterranea 15: 7-13.

Shipton, W. A., Boyd, W. R. J., Rosielle, A. A., and Shearer, B.L. 1971. The common septoria diseases of wheat. Bot. Rev. 27: 231-262.

Stewart, D. M., Hafiz, A., and Abdelhak, T. 1972. Disease epiphytotic threats to high-yielding and local wheats in the Near Esat. FAO. Plant Protection Bull. 20: 50-57.

Tyagi, P. D., Joshi, L. M. and Renfro, B. L. 1969. Reaction of wheat varieties to *S. tritici* and report of an epidemic in North Westen Punjab. Ind. Phytopathology 22: 175-178.

Van Ginkel, M. 1986. Inheritance of resistance in wheat to *Septoria tritici*. PhD. Thesis. Montana State University. Bozeman. MT. 102 pp.

Wilson, R. E. 1985. Inheritance of resistance to *Septoria tritici* in wheat. Pages 33-35. *In* Septoria of Cereals. Proc. of the Workshop. A. L. Scharen, ed. Bozeman, MT. 2-4 Aug. 1983. MSU. 116 pp.

CHAPTER V

OCCURRENCE, DISTRIBUTION, AND SEVERITY OF SEPTORIA DISEASES IN MOROCCO

Abstract

Commercial fields were surveyed in 1991 for the occurrence, distribution and severity of septoria diseases on wheat in Morocco. Septoria blotch (SB) occured in 62% of fields examined throughout the wheat growing areas of Morocco. Seventy two percent of fields in the southern region (SR) were infested compared to 52% in the northern region (NR). Prevalence of SB on bread wheat was almost twice of that on durum wheat. Both species of *Septoria* occurred on wheat, with *S. tritici* being found in 82% of fields and *S. nodorum* being found less frequently (24%). On bread wheat *S. tritici* was eight times more frequent (96%) than *S. nodorum*; however, the frequency of occurrence of both species on durum wheat was the same. *S. tritici* was common in both regions surveyed while *S. nodorum* occurred twice as frequently in the SR as compared to the NR. SB severity was higher on bread wheat than on durum wheat, irrespective of the region surveyed. The ascogenous state (*Mycosphaerella graminicola*) of *S. tritici* was not found in Morocco, and further investigations are needed to

establish its presence. These results represent the first step in studying septoria diseases in Morocco.

Introduction

Wheat is a nutritious, convenient, and economical source of food. It provides about 20% of the world's food calories, and is a staple for nearly 40% of the world's population (Wiese, 1987). Diseases are considered one of the main limiting factors for wheat production. Over a hundred pathogens are described as causal agents of wheat diseases (Wiese, 1987), and septoria diseases caused by *Septoria* spp. are considered among the most important diseases of wheat.

There are two major diseases caused by *Septoria* spp., which are generally referred to collectively as septoria blotch (SB) (Eyal et al., 1987; King et al., 1983; Shipton et al., 1971). These are septoria tritici blotch (STB) incited by the fungus *Mycosphaerella graminicola* (Fuckel) Schroeter (Anamorph: *S. tritici* Rob. ex Desm.) and septoria nodorum blotch (SNB) caused by *Leptosphaeria nodorum* E. Muller [Anamorph: *S. nodorum* (Berk.) Berk.]. Both pathogens can be pernicious and yield losses attributed to these diseases have been reported to range from slight to 60% (Eyal et al., 1987; king et al., 1983; Saâri and Wilcoxson, 1974; Shipton et al., 1971; Stewart et al., 1972).

Normally *S. tritici* affects only leaves and sheaths, but in severe epidemic conditions, glumes and awns may become involved. *S. nodorum* usually affects all above-ground

parts including the glumes. Both species cause irregular, dead spots in which pycnidıa are eventually produced. Pycnidıa are just vısible to the unaided eye and appear on leaf lesions 10-20 days after infection (Eyal et al., 1987; Shipton et al., 1971; Weber, 1922). SB ıs assocıated wıth humıd conditıons, wıth *S. trıtıcı* more prevalent ın cooler climates and being active during the cooler portions of the growing season (Wiese, 1987).

The proliferation of susceptible wheat cultivars also has exacerbated the importance of SB in dryland wheat-growing areas. Severe outbreaks of septorıa diseases have occured in arıd countrıes along the Medıterranean seacost (Saâri and Wilcoxson, 1974; Stewart et al., 1972). In North Africa, SB, formerly a minor disease, is now a maȷor dısease problem of wheat. This change is due in part to large-scale planting of introduced high-yielding wheats, and the occurrence of weather conditions favorable for the development of *Septoria* diseases (Djerbi et al., 1974; Saârı and Wilcoxson, 1974; Stewart et al., 1972). In Morocco, a severe epidemic during the 1968-1969 season highlighted the potential importance of SB of wheat (Saâri and Wilcoxson, 1974; Stewart et al., 1972). As a consequence, some studies ınvestıgatıng the importance of the *Septoria* spp. and yıeld losses due to SB have been initiated (Saâdaoui, 1975; Schluter and Janati, 1976). Although no informatıon has been published since then, SB has been observed annually on wheat and has been particularly important sınce the mıd-1980's

(Burleigh et al., 1991; Jlibene, 1990; Saâdaoui, 1987).

The objective of this study was to determine the relative incidence of each septoria disease present on wheat, its geographic distribution, and the severity of SB on the species of *Triticum* grown in the different wheat growing areas surveyed in Morocco.

Materials and Methods

Morocco was divided into two large regions (North and South) based on moisture and temperature differences, with the northern region (NR) being, in general, more moist and cooler than the southern region (SR)(Burleigh et al., 1991). These two regions include the most distinctive and important wheat growing areas in the country. The survey covered most of the wheat growing areas within each region. The routes followed and some reference points are shown in Figs 1 & 2. The first field was selected at random and thereafter, a stop was made every 10-20 Km. The survey was made when the growth stage was flowering to milky-ripe according to the Feekes'scale (Large, 1954). One hundred thirteen commercial fields in the SR and 115 fields in the NR were inspected. In each field 50-100 plants were examined for *Septoria* symptoms. When necrotic lesions containing pycnidia were found, either on leaves or on spikes, 5-10 plants were collected for examination in a laboratory to determine the species of *Septoria* on the tissue. Disease prevalence was defined as the number of fields where SB was present divided

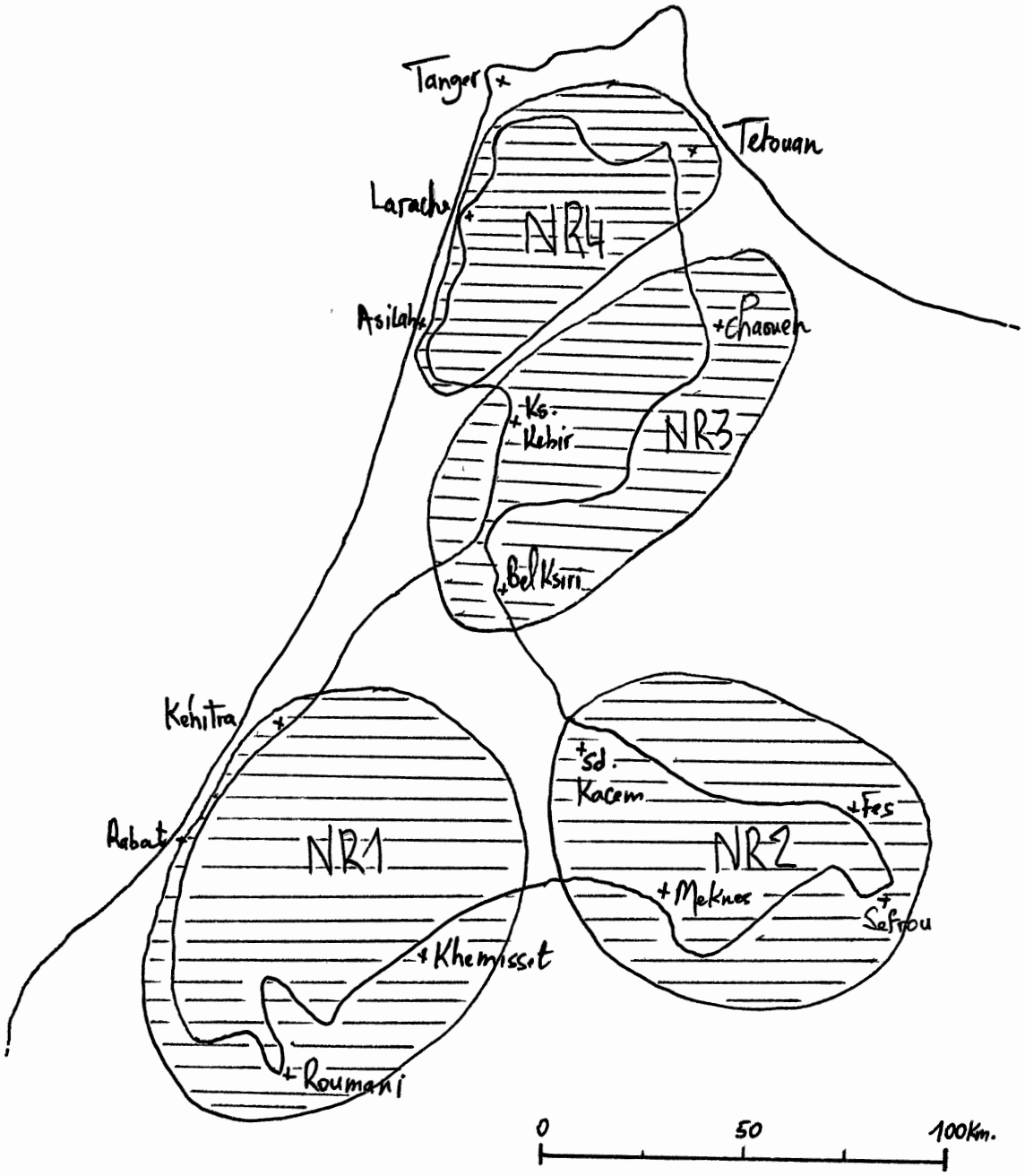

Figure 1. Important Wheat Growing Areas (NR1, NR2, NR3, and NR4), Routes Followed, and Reference Points in the Northern Region of Morocco

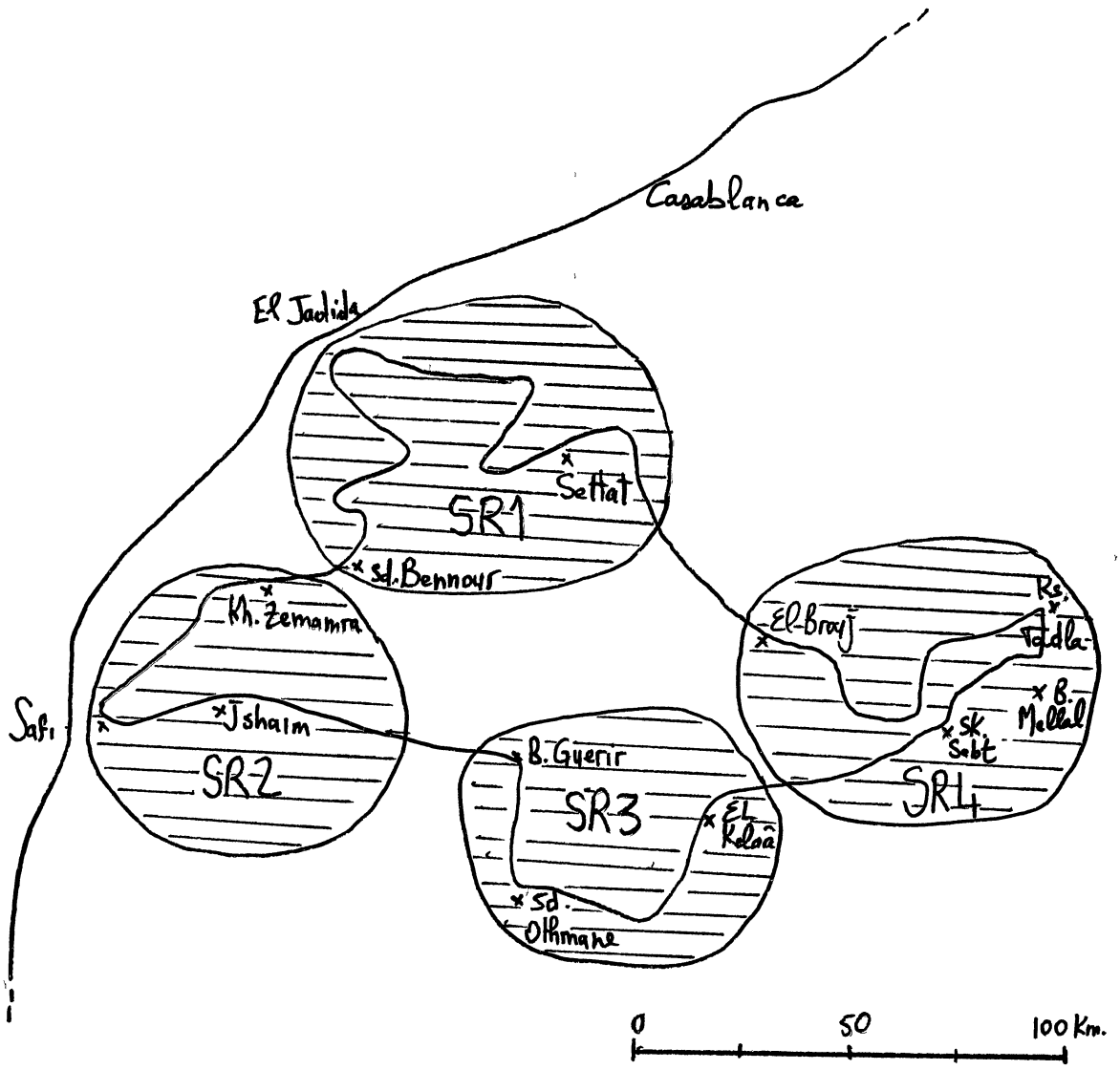

Figure 2. Important Wheat Growing Areas (SR1, SR2, SR3, and SR4), Routes Followed, and Reference Points in the Southern Region of Morocco.

by the total number of fields sampled (Nutter, Jr. et al., 1991).

Plants in each field were evaluated using a scale of 1-10 for an average severity index graduated in 10% increments from 0-90% necrosis of the leaf area on the upper half of the plant. An average rating was recorded for each surveyed field from the individually assessed plants and then for the area within each region (Gilchrist et al., 1982).

To identify the *Septoria* species present, tissue with necrotic lesions bearing pycnidia were moistened with sterile water to induce pycnidiospore liberation. Microscopic examination of spore morphology and dimensions were used to confirm the identity of the *Septoria* spp. present (Eyal et al., 1987). Isolations of random, single pycnidial culture were made to establish cultures representative of the area where the disease occurred for confirmation of pathogenicity and examination of conidia produced in culture.

Results

Laboratory evaluation of lesions on leaf and spike specimens collected from all diseased fields in 1991 survey showed SB to occur throughout the wheat growing areas in both regions of Morocco. Disease prevalence (DP), defined as the number of fields where the disease is present divided by the total number of fields sampled (Nutter, Jr. et al., 1991), averaged 62% in the two regions (NR & SR) surveyed

(Table IX). A difference between the two regions was observed with higher DP in the SR: 72%, compared to 52% in the NR (Table IX). SB was almost twice as prevalent (81%) on *T. aestivum* L. than on *T. durum* D. (43%) (Table X). Other foliar diseases also were observed, including stem, leaf, and yellow rusts; tan spot; and powdery mildew.

For the period studied SB symptoms on leaves were caused by *S. tritici* and *S. nodorum*. Symptoms on ears were observed only in one and four fields in the NR and the SR, respectively (Tables XI, XII, & XIII). *S. tritici* was the most prominent species, being identified in 82% of the samples. *S. nodorum* was found in 24% of the samples (Table XI).

On bread wheat *S. tritici* was eight times more frequent (96%) than *S. nodorum* (12%). However, on durum wheat there were almost no differences between the frequency of occurrence of the two species (Table XI).

During 1991, *S. tritici* was recorded at high frequency in both regions surveyed (NR: 85% and SR: 79%), but *S. nodorum* was recorded more frequently in the SR (32%) than in the NR (13%) (Tables XII & XIII). A comparison between STB and SNB showed the proportion of fields with STB was 6 times (85 vs 13%) and 2.5 times (79 vs 32%) more frequent than SNB in the NR and SR, respectively (Tables XII & XIII). The distribution of the two species within the four areas of the NR was consistent (Table XII); however, in the SR, a few fields with SB were found in the SR3 (Benguerir-Sidi

TABLE IX

PREVALENCE OF SEPTORIA BLOTCH IN NORTHERN AND SOUTHERN REGIONS OF MOROCCO

Fields	North	South	Total
Surveyed	115	113	228
With *Septoria*	60	82	142
Prevalence[x] (%)	52	72	62

[x] Prevalence defined as the number of fields where the disease is present divided by the total number of fields surveyed.

TABLE X

PREVALENCE OF SEPTORIA BLOTCH IN COMMERCIAL BREAD AND DURUM WHEAT FIELDS

Fields	Bread wheat.	Durum wheat	Total
Surveyed	116	112	228
With *Septoria*	94	48	142
Prevalence[x](%)	81	43	62

[x] Prevalence defined as the number of fields where the disease is present divided by the total number of fields surveyed.

TABLE XI

INCIDENCE OF THE *SEPTORIA* SPP. IN COMMERCIAL BREAD AND DURUM WHEAT FIELDS

Septoria spp.	Bread wheat		Durum wheat		Total	
	Number	(%)	Number	(%)	Number	(%)
S. tritici	90[x]	(96)	26[y]	(54)	116[z]	(82)
S. nodorum	11	(12)	23	(48)	34	(24)
S. tritici + *S. nodorum*	10	(11)	11	(23)	21	(15)

[x] Total number of bread wheat fields with SB is 94.

[y] Total number of durum wheat fields with SB is 48.

[z] Total number of wheat fields with SB is 142.

TABLE XII

INCIDENCE OF THE SEPTORIA SPP. ON WHEAT IN THE DIFFERENT AREAS OF THE NORTH REGION

Septoria	NR1[x]	NR2	NR3	NR4	Total[y]	(%)
S. tritici	12	15	12	12	51	(85)
S. nodorum	3	2	1	2	8	(13)
S. tritici + *S. nodorum*	2	0	1	3	6	(10)

[x] NR1 : Rabat-Khmisset.
NR2 : Meknes-Fes.
NR3 : Belksiri-Ksar-Chaouen.
NR4 : Larache-Tanger-Tetouan.

[y] The number of fields with *Septoria* in the NR was 60 out of 115 surveyed.

TABLE XIII

INCIDENCE OF THE SEPTORIA SPP. ON WHEAT IN THE DIFFERENT AREAS OF THE SOUTH REGION

Septoria	SR1[x]	SR2	SR3	SR4	Total[y]	(%)
S. tritici	24	18	1	22	65	(79)
S. nodorum	8	12	2	4	26	(32)
S. tritici + *S. nodorum*	4	6	-	5	15	(18)

[x] SR1: Settat-El Jadida-Sidi Bennour.
SR2: Khmiss Zmamra-Safi-Youssoufia.
SR3: Benguerir-Sidi Othmane-El Kelaâ.
SR4: Souk Sebt-Beni Mellal-Kasba Tadla-El brouj.

[y] The number of fields with *Septoria* in the SR was 82 out of 113 surveyed.

Othmane-El Kelaâ) area in comparison to other three SR areas. Mixed infection by *S. tritici* and *S. nodorum* was present but less important (Tables XII & XIII).

The severity of septoria blotch diseases, which was based on total area affected on the upper half of leaves, was higher on bread wheats than on durum wheats in all regions surveyed (Tables XIV & XV). In general, no differences existed in the severity of SB between the different areas within the NR for both classes of wheat grown (Table XIV); however, within the SR, SB was less severe in SR3 and SR4, and was mainly on bread wheat (Table XV).

Sanderson (1976) linked a *Mycosphaerella* species with *S. tritici* in New Zeland and proposed a new combination for the perfect state as *M. graminicola*. A survey was conducted in summer 1991, along the road Berrechid-Casa-Rabat to search for the sexual form of this fungus in Morocco. No pseudothecia were found on samples of wheat plants observed either in the field or under microscopic examination in the laboratory.

Discussion

Our investigation revealed that there are two species of *Septoria* on wheat in Morocco, namely *S. tritici* and *S. nodorum*. These two species with *S. avenae* f. sp. *triticea* are the most frequently reported causal agents of septoria diseases on wheat around the world but with different

TABLE XIV

SEVERITY[x] OF SEPTORIA BLOTCH IN COMMERCIAL BREAD AND DURUM WHEAT FIELDS WITHIN THE DIFFERENT AREAS OF THE NORTH REGION

Wheat field	NR1[y]	NR2	NR3	NR4
Bread wheat	30.0	15.0	25.0	20.0
Durum wheat	5.0	8.0	5.0	5.0

[x] Plants in each field were evaluated using a scale of 1-10 for an average severity index graduated in 10 % increments from 0-90 % necrosis of the leaf area on the upper half of the plant. An average severity rating was recorded for each surveyed field from the individually assessed plants and then for the area within the region.

[y] NR1: Rabat-Khemisset.
NR2: Meknes-Fes.
NR3: Belksiri-Ksar-Chaouen.
NR4: Larache-Tanger-Tetouan.

TABLE XV

SEVERITY[x] OF SEPTORIA BLOTCH IN COMMERCIAL BREAD AND DURUM WHEAT FIELDS WITHIN THE DIFFERENT AREAS OF THE SOUTH REGION

Wheat field	SR1[y]	SR2	SR3	SR4
Bread wheat	20.0	20.0	5.0	10.0
Durum wheat	5.0	8.0	5.0	5.0

[x] Plants in each field were evaluated using a scale of 1-10 for an average severity index graduated in 10 % increments from 0-90 % necrosis of the leaf area on the upper half of the plant. An average severity rating was recorded for each surveyed field from the individually assessed plants and then for the area within the region.

[y] SR1: Settat-El Jadida-Sidi Bennour.
SR2: Khmiss Zmamra-Safi-Youssoufia.
SR3: Benguerir-Sidi Othmane-El Kelaâ.
SR4: Souk Sebt-Beni Mellal-kasba Tadla-El Brouj.

relative importance in each country (Eyal et al., 1987; King et al., 1983; Richardson & Noble, 1970; Shearer & Calpouzos, 1973). Field and laboratory observations indicated that prevalence, distribution, and severity of SB are important on the two species of wheat in the cereal growing areas of Morocco. The much larger number of fields from which the SB causal agents were recovered in this study in comparison to earlier investigations (Burleigh et al., 1991; Saâdaoui, 1975; Schluter & Janati, 1976) gives a more complete indication of the distribution of the *Septoria* spp. based on geographical location and wheat species grown.

Septoria tritici was present almost as the singular species in all the samples with SB on bread wheat. Information collated by Burleigh et al. (1991), Saâri & Wilcoxson (1974), Saâdaoui (1975), and Schluter & Janati (1976) listed *S. tritici* as the predominant species on wheat in Morocco. Our results agree with this assessment and confirm that the main wheat growing areas of the country are all prone to serious STB epidemics. In a two-year study conducted on winter wheat in commercial fields in Michigan, distinct differences in the occurrence of *M. graminicola* and *L. nodorum* were observed (Hart et al., 1984).

This report confirms the presence of *S. nodorum* in Morocco. Although this species apparently is more limited in distribution and is mainly on bread wheat, its disease potential presents a concern for future spread and impact on wheat production in Morocco.

Although the presence and distribution of the two *Septoria* spp. have been mapped in a large extent of the cerealiculture, some important wheat growing areas (North-eastern part: Taza and Oujda provinces) have not been sampled. These regions should be surveyed in the future to fully assess the occurrence and distribution of SB in Morocco.

Symptoms caused by *S. tritici* and *S. nodorum* were frequently found on leaves and leaf sheaths, while symptoms on ears were observed only rarely. It is important to note that these fungi frequently occur in complexes and it is common to encounter the two *Septoria* spp. together on the same plant. Thus, 10% of the fields in the NR and 18% in the SR had mixed infections.

Morphology and size of conidia of *S. tritici* and *S. nodorum* found in Morocco were similar to those described in the literature (Eyal et al., 1987; Scharen & Sanderson, 1985).

SB prevalence was higher in the SR than in the NR (Table IX). This variability is due to the high frequency of occurrence of SNB in the SR (32%) compared to the NR (13%), since STB prevalence was similar between the two regions. It is not possible to deduce from this survey the reason for the variability in occurrence of SNB between the two regions; however, differences in date of seeding, rainfall, growth stage at the optimum time of infection, and temperature are contributing factors in explaining this

variabılıty (Hart et al., 1984; Hess & Shaner, 1987; Holmes & Colhoun, 1974; Shaw & Royle, 1989; Shearer & Wılcoxson, 1980; Wainshilbaum & Lipps, 1991; Wiese, 1987).

The sexual state of *S. trıtıcı* was not found ın Morocco, and the imperfect state ıs the most ımportant means for spread of the pathogen between successıve crops. In some countries where weather conditions are favourable for the development of the perfect state, ascospores play a major role as the primary source of ınoculum (Sanderson et al., 1985). Further investigations and more extensıve fıeld surveys are needed to establish the presence and the significance of the perfect state of *S. tritici* ın Morocco.

The results of the present investigation are the first step in studying of septoria diseases in Morocco. A culture collection including a number of isolates from different regıons and from the two specıes of *Tritıcum*, wıth dıverse vırulence patterns, has been establıshed. These ısolates will be used to screen for sources of resistance to *Septoria* and to facilitate a program of breeding for resıstance. Jlibene (1990), has been selecting cultıvars and breeding lines for STB to find sources of resistance effective for the condıtıons of our country.

Literature Cited

Burleigh, J. R., Ezzahiri, B., and Roelfs, A. P. 1991. Assessment of cultivar performance and disease ımpact on cereals in Morocco. Plant Dis. 75: 65-73.

Djerbi, M., Ghodbane, A. G., Daâloul, A., and Varughese, G. 1974. Identifıcation des sources de résıstance à la septoriose de blé (*S. tritici* Rob. et Desm.). Ann. Phytopathol. 6: 495-496.

Eyal, Z., Scharen, A. L., Prescott, J. M., and Van Ginkel, M. 1987. The septoria diseases of wheat: Concepts and methods of disease management. Mexico, D.F.: CIMMYT. 52 pp. 17 figures. 20 color plates.

Gilchrist, D. G., Brownell, K. H., and Martensen, A. N. 1982. Incidence and distribution of septoria diseases of wheat in California. Plant Dis. 66: 513-515.

Hart, L. P., Fulbright, D. W., Clayton, J. L., and Ravenscroft, A. V. 1984. Occurrence of septoria nodorum blotch and septoria tritici blotch in Michigan winter wheat. Plant Dis. 68: 418-420.

Hess, D. E., and Shaner, G. 1987. Effect of moısture and temperature on development of septoria trıtici blotch in wheat. Phytopathology 77: 215-219.

Holmes, S. J., and Colhoun, J. 1974. Infection of wheat by *Septoria nodorum* and *S. tritici* ın relatıon to plant age, air temperature and relative humidıty. Trans. Br. Mycol. Soc. 63: 329-338.

Jlibene, M. 1990. Inheritance of resistance to septoria tritici blotch (*Mycosphaerella graminicola*) in hexaploid wheat. PhD. Thesis. Univ. Missouri-Columbia. 86 pp.

King, J. E., Cook, R. J., and Melville, S. C. 1983. A revıew of *Septoria* diseases of wheat and barley. Ann. Appl. Biol. 103: 345-373.

Large, E. C. 1954. Growth stages ın cereals. Illustratıons of the Feeke's scale. Plant Pathol. 3: 129.

Nutter, Jr., F. W., Teng, P. S., and Shokes, F. M. 1991. Disease assessment terms and concepts. Plant Dis. 75: 1187-1188.

Richardson, J. J., and Noble, M. 1970. *Septoria* species on cereals-a note to aid their identification. Pl. Pathol. 19: 159-163.

Saâdaoui, E. M. 1975. Contribution à l'étude des septorioses du blé au Maroc. Mémoire de 3e cycle presenté à l'Institut Agronomique Hassan II, Rabat, Morocco.

Saâdaoui, E. M. 1987. Physiologic specialization of *Septoria tritici* in Morocco. Plant Dis. 71: 153-155.

Saari, E. E., and Wilcoxson, R. D. 1974. Plant disease situation of high-yielding dwarf wheats in Asia and Africa. Ann. Rev. Phytopathol. 12: 49-68.

Sanderson, F. R. 1976. *Mycosphaerella graminicola* (Fuckel) Sanderson Comb. nov., the ascogenous state of *Septoria tritici* Rob. apud Desm. N. Z. J. Bot. 14: 359-360.

Sanderson, F. R., Scharen, A. L., and Scott, P. R. 1985. Sources and importance of primary infection and identities of associated propagules. Pp. 57-64. *In*: *Septoria* of cereals. Proc. Workshop. A.L.Scharen, ed. August 2-4, 1983, Bozeman, MT. USDA-ARS Publ. No. 12. 116 pp.

Scharen, A. L., and Sanderson, F. R. 1985. Identification, distribution and nomenclature of the *Septoria* species that attack cereals. pp.37-41. *In*: *Septoria* of cereals. Proc. Workshop. A.LScaren, ed. August 2-4, 1983, Bozeman,MT. USDA-ARS Publ. No. 12. 116 pp.

Schluter, K., and Janati, A. 1976. Les septorioses du blé au Maroc. Phytopathologia Mediterranea 15: 7-13.

Shaw, M. W., and Royle, D. J. 1989. An epidemiologically based forcasting scheme for *Septoria tritici*. Pp. 107-109. *In*: *Septoria* of cereals Proc. Workshop. P. M. Fried, ed. July 4-7, 1989, Zurich, Switzerland. 189 pp.

Shearer, B. L., and Calpouzos, L. 1973. Relative prevalence of *Septoria avenae* f. sp. *triticea, Septoria nodorum* and *Septoria tritici* on spring wheat in Minnesota. Plant Dis. Rep. 57: 99-103.

Shearer, B. L., and Wilcoxson, R. D. 1980. Sporulation of *Septoria* species on wheat and barley in Minnesota. Tech. Bull. 323. Univ. Minn. Agric. Exp. Stn. 30 pp.

Shipton, W. A., Boyd, W. R. J., Rosielle, A. A., and Shearer, B. L. 1971. The common septoria diseases of wheat. Bot. Rev. 27: 231-262.

Stewart, D. M., Hafiz, A., and Abdelhak, T. 1972. Disease epiphytotic threats to high-yielding and local wheats in the Near East. FAO. Plant Protection Bull. 20: 50-57.

Wainshilbaum, S. J., and Lipps, P. E. 1991. Effect of temperature and growth stage of wheat on the development of leaf and glume blotch caused by *Septoria tritici* and *Septoria nodorum*. Plant Dis. 75: 993-998.

Weber, G. F. 1922. *Septoria* diseases of wheat. Phytopathology 12: 537-585.

Wiese, M. V. 1987. Compendium of wheat diseases. 2nd ed. Am. Phytopathol. Soc., St. Paul, MN. 112 pp.

APPENDIX

EFFECT OF SURFACTANTS AND WHEAT LEAF LEACHATE ON PRODUCTION GERMINATION AND INFECTION OF *SEPTORIA TRITICI* CONIDIA

To facilitate infection of wheat by *S. tritici*, researchers have added Tween 20, 0.5% gelatin, or nothing at all, to conidial suspensions of *S. tritici* prior to inoculating plants. Results from these inoculations have been inconsistent. The present work was undertaken to compare the effects of adjuvants on germination of conidia, length of germ tubes, and lesion development, and to determine if wheat leachate enhaces germination of conidia, growth by budding in culture, and production of lesions on inoculated plants.

Three isolates of *S. tritici* were used in these experiments; isolate 88C obtained from Morocco, and isolates 83A and AF12-3 obtained from Oklahoma. Three drops (0.075 ml) of a conidial suspension, containing about 200 conidia per ml, were placed on plain water agar and water agar amended with 0.3 ml of Tween 20 or Amway surfactant (Amway Corp., Ida, MI 49201). Conidia were considered to have germinated if, after either 20 or 24 hr at 21 C, they had terminal germ tubes at least as long as the parent conidium. Germ tubes were measured in μm; when more than one germ tube

was present, the longest was measured.

Wheat leaf leachate was prepared from 30 gm of fresh green wheat leaves (cultivars BT4, BT6, BT93, and BT323) by either soaking leaves in distilled water for 36 hr or by boiling in water for 1 hr. The leachate was then filtered, first through cheesecloth and glasswool, then through filter paper plus charcoal at least three times. Lesion counts were made 14 to 15 days after inoculation while lesions were still chlorotic flecks and coalescing was minimal.

As compared to the control, Tween 20 suppressed germination of conidia of *S. tritici* isolates 83A and 88C by 59-70% and 27-47%, respectively, (Table XVI). Germ tube growth was inhibited by 58-62% and 59% for each isolate (Table XVII), and Amway surfactant was less inhibitory for both germination and germ tube growth than Tween 20 (Tables XVI and XVII).

Leachate from resistant and susceptible wheat cultivars did not affect germination of conidia of the *S. tritici* isolates on water agar (Table XVIII). Tests comparing autoclaved and filtered leachate indicated that autoclaving for 20 min at 14 psi had no effect on germination of conidia (the percent germination did not deviate significantly from the control).

Inocula consisting of *S. tritici* conidia suspended in water supplemented with leachate from green wheat leaves, with or without Tween 20 or Amway, elicited more lesions per gram of inoculated leaf tissue than similar inocula lacking

TABLE XVI

EFFECT OF SURFACTANTS ON GERMINATION OF CONIDIA OF *S. TRITICI* isolates

	Experiment[x,y] I Isolate		Experiment II Isolate	
Treatment	83A	88c	83A	88c
Water agar	74.3 a[z]	73.0 a	70.0 a	85.0 a
Amway	65.0 b	75.0 a	55.0 b	62.0 b
Tween 20	22.0 c	53.0 b	29.0 c	45.0 c

[x] Randomized complete block design. EI: six blocks; EII: eight blocks, 100 conidia / treatment / block.

[y] Incubation: EI≈ 20 hr, EII≈ 24 hr, 21 C, constant light.

[z] Analysis: Tukey's W procedure p=0.05. Data transformed before analysis.

TABLE XVII

EFFECT OF SURFACTANTS ON GERM TUBE LENGTH (μm) OF CONIDIA OF *SEPTORIA TRITICI* ISOLATES

	Experiment[x,y] I Isolate		Experiment II Isolate	
Treatment	83A	88c	83A	88c
Water agar	34.1 a[z]	58.7 a	51.2 a	71.5 a
Amway	28.2 b	48.5 b	34.3 b	50.6 b
Tween	12.9 c	24.3 c	21.4 c	29.6 c

[x] Randomized complete block design. E.I: 6 blocks, E.II: 8 blocks, 20 observations/treatment/block.

[y] Incubation: E.I: 20 hr, E.II: 24 hr, 21 C, constant light.

[z] Analysis: Tukey's W procedure P=0.05.

TABLE XVIII

GERMINATION OF *S. TRITICI* CONIDIA ON WATER AGAR[w,x] CONTAINING LEAF LEACHATE[y] FROM RESISTANT AND SUSCEPTIBLE WHEAT LINES

Wheat source of leachate	Germination
Water agar	71.0 a[z]
BT4	72.0 a
BT6	65.0 a
BT93	68.0 a
BT323	69.0 a

[w] Randomized complete block design, 6 blocks, 100 conidia/treatment/block.

[x] Incubation: 24 hr., 21 C, constant light.

[y] Leachate: 30 g leaf tissue/L.

[z] Analysis: Tukey's W procedure, P=0.05.

leachate (Table XIX). Conidia production by three ısolates of *S. trıtıcı* was not statıstıcally dıfferent on the commonly used medium (yeast malt agar)and the newly modıfied yeast-sucrose medium with leaf leachate (Table XX).

TABLE XIX

EFFECT OF SURFACTANTS AND LEAF LEACHATE ON DEVELOPMENT OF SEPTORIA TRITICI BLOTCH LESIONS[v,w,x]

Treatment	Lesions/g. dry weight leaf tissue
Amway + leachate[y]	2046 a[z]
Leachate	1241 ab
Tween + leachate	1184 ab
Amway	1045 b
Water	832 b
Tween 20	620 b

[v] Randomized complete block design, 3 blocks, 7 plants/treatment/block.

[w] Inoculation: 10^7 conidia/ml., 4 days in mist chamber.

[x] Incubation: 15 days in growth chamber, 21 C, 16 hr light.

[y] Leachate: 30 gm tissue/L. Surfactant: .3 ml/L.

[z] Analysis: Tukey's W procedure p= 0.05

TABLE XX

CONIDIA PRODUCTION[v] BY THREE ISOLATES OF *SEPTORIA TRITICI* IN A MODIFIED YEAST-SUCROSE MEDIUM

Expt.[w]	Medium	Conidia/ml (10^8)		
		88c	83A	AF12-3
I	YMA[x]	1.9 a[z]	2.3 a	0.9 a
	YLL[y]	2.5 a	2.5 a	0.8 a
II	YMA	2.1 a	2.0 a	1.0 a
	YLL	2.4 a	2.3 a	1.0 a

[v] Incubation: 6-7 days growth, 21 C,constant light.

[w] Randomized comlete design, 3 blocks.

[x] YMA: 4 g sucrose, 4 g yeast, 4 g malt in 1000 ml;

[y] YLL: 2 g sucrose, 2 g yeast, 30 g fresh leaf tissue.

[z] Analysis: Tukey's W procedure P=0.05.

VITA

Ali Farih

Candidate for the Degree

Doctor of Philosophy

Thesis: COMPONENTS OF PARTIAL RESISTANCE, MODE OF INHERITANCE OF RESISTANCE TO SEPTORIA TRITICI BLOTCH, AND STATUS OF SEPTORIA DISEASES IN MOROCCO

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