CHAPTER 3

RESPONSES OF FOUR CULTIVARS AND TWO INBRED LINES TO *Exserohilum turcicum* AND MAIZE DISEASE SYMPTOMATOLOGY IN KENYA

(ABSTRACT)

Four maize cultivars namely; Coast Composite, Pwani1, Pwani4, Mzihana and two inbred lines were inoculated with eight isolates of *E. turcicum.* All the four cultivars from Kenya were susceptible. Mo17 inbred with polygenic resistance exhibited the smallest lesions while B73 inbred the known suscept had the largest lesions bearing profuse sporulation. Maize disease symptom expression was examined during post milk stage and plants were evaluated for lesion number, size and appearance of symptom complexes. The lesions dimensions recorded in Kenya are larger than those encountered in the literature. Lesions of over 400 mm by 20 mm were recorded for *E. turcicum,* 60 mm by 5 mm for *B. maydis* and 6 mm by 3 mm for *P. maydis* which were the most serious pathogens observed. The lesions of *E. turcicum* were observed in the middle third and lower part of the canopy while those of *P. maydis* were more dense in the upper third of the canopy. *B. maydis* infection occurred in a diffused manner but was more common in the lower leaves and less in the upper canopy. These three diseases could be found on a single plant or field. *P. maydis* a relatively unknown pathogen in Kenya, previously of minor importance, was observed in epidemic proportions in the western highlands, Lake Victoria basin and central regions. In the western Kenya maize belt *P. maydis* was dominant in the upper canopy including the top most leaves. Common rust caused by *Puccinia sorghi* a pathogen of maize in the western highlands was diffused throughout the plants with a concentration in the middle canopy where the infection occurred as single pustules to huge brownish blighted zones sometimes with tattered leaf surfaces covered with many pustules. Many of the lesions were sporulating in situ and were larger than any reported in the literature. Some individual plants exhibited intense and extensive chlorosis affecting large areas. This indicated that the varieties being grown in Kenya are very susceptible to the diseases encountered.

INTRODUCTION

Maize is the most important staple crop in Kenya with 1.6 hectares estimated in 1992. Yields are quite low with less than a ton per hectare; whereas a potential of 3 Tons/hectare is possible. About 60% of the maize consumed in the country comes from the Rift Valley and highlands to the west. The National Cereal Improvement Program is charged with maize improvement through development of maize hybrids and composites with superior yields, desirable agronomic characteristics including adaptability to the various climatic zones, and genetic tolerance to diseases and insects (Gebrekidan *et al*. 1992). Diseases and insects are serious constraints to yield improvement throughout the country. Southern leaf blight caused by *B. maydis,* lowland rust caused *by P. polysora,* northern leaf blight caused by *E. turcicum* and maize streak are the most important diseases in Kenya. The regional research center at Mtwapa is an ideal location for screening maize genotypes for genetic variability as the temperature and relative humidity are optimum for disease development (Gebrekidan *et al.* 1992, Njuguna *et al.* 1992). The other locations are Katumani for the arid lands, Embu for mid altitudes, Kitale for high altitudes and Ol-joro-Orok for very high altitude conditions (Gebrekidan *et al.* 1992).

Northern maize leaf blight is favored by mild temperature and high humidity (Ullstrup 1970). Heavy dews, cool temperatures, and frequent rains create good sustained environmental conditions for disease development (Jordan *et al.* 1983). Levy (1991) showed that isolates from different areas were different in parasitic fitness as was indicated by infection efficiency, sporulation and lesion size, while isolates of same location showed less variation. Inoculum in previous crop has been found to be critical in epidemic build up for subsequent cropping, especially in non tillage systems.

The reaction of a host plant to attack by a pathogen is normally expressed at the point of contact or infection court. Various manifestations, specific and characteristic of the interaction governed by host genetic expression, pathogen aggressiveness and the environment are factors that impact on both. These reactions or lesions are the symptoms that are examined to determine the nature of responses. The intensity or frequency of the responses or lesions define disease intensity and severity and consequently the damage caused.

Lesion size is one of the components of susceptibility of a host to a fungus (Populer 1978). Traut and Warren (1993) reported that susceptible lines of corn produced longer and wider lesions than those with partial or full resistance when inoculated with races of *Bipolaris zeicola*. Some races of the pathogen induced larger lesions than others and lesion types were genotype dependent, while shapes were race dependent. Temperature influences rate of lesion development, lesion size, and formation of spores (Warren 1975), and hence very critical in epidemic development. Hooker (1961) identified chlorotic lesion types of *E. turcicum* resistance maize cultivars characterized by chlorosis, late developing lesions with small necrotic centers surrounded by a light green margin, and greatly reduced sporulation. Susceptible cultivars expressed rapid developing necrotic susceptible lesions with abundant sporulation of *H. turcicum*. The resistance to *E. turcicum* was controlled by a single dominant gene, designated as *Ht* gene. Homozygous dominant plants rarely exhibit lesions. The average level of resistance or mean lesion area, the rate of increase in lesion size, and the shape of the lesion and size are influenced by host gene makeup as determined by contributions of each parent (Singulas *et al*. 1988). Other reports indicate that toxin producing pathogens or some races produce large and rapid developing lesions on susceptible hosts (Xiao *et al.* 1991, 1992, Traut and Warren 1993). Lesion size depends partially on the number of infection sites, genotypes, environmental conditions and aggressiveness of the pathogen. Lesion size or extent of tissue coverage was not adequate to measure resistance (Nicholoson and Warren 1975). Lesion size has also been used to estimate the resistance of several maize inbred lines to different isolates of race 3 of *B. carbonum* (Hamid *et al* 1982). Polygenic resistance is normally expressed by reduced number of lesions and decrease in lesion size and amount of sporulation (Ullstrup 1970). Adipala *et al.* (1993) inoculated Mo17 and B73 and nine cultivars from Uganda with races 0, 1, 23 and 23N of *E. turcicum* and reported mean lesion lengths of 67 mm and 41 mm for B73 and Mo17 respectively induced by races 0 and 1. Races 23 and 23N induced mean lesion lengths of 59 mm and 43 mm on B73 and Mo 17 respectively. Mo17 was more susceptible than most Ugandan cultivars tested. However, Mo17 had the smallest mean lesion lengths measuring about 120 mm compared to the Ugandan cultivars which had mean lesion lengths range of 126 mm to 267 mm. B73Ht1

had the smallest mean lesion of 84 mm lengths overall. B73 the known suscept had numerous but smaller mean lesion lengths than all Ugandan cultivars. This research was undertaken to assess genotypic responses to *E. turcicum* and evaluate field symptom expression in maize infected by *E. turcicum, P. maydis, B. maydis, P. sorghi and P. polysora*.

LITERATURE REVIEW

The most important maize diseases are rust (*Puccinia* spp.) and leaf blight in Kenya (Manwiller 1983). Pathogens that attack the leaves impact directly on the amount of dry matter stored in the grain and possibly leaf blights can induce up to 50% losses in yield (Ullstrup and Miller 1957). Disease severity is often assessed using various methods that utilize scales, keys, visual estimations and measurements (Adipala *et al*.1993, Horsfall and Barratt 1945, Saghaai Maroof *et al*. 1993, Slopeck 1989, Nicholson and Warren 1975, Robbins and Warren 1993, James 1968, Solomonovitz, Levy and Pataky 1992, Traut and Warren 1993).

LOWLAND RUST-*Puccinia polysora*

Lowland rust of maize caused by *Puccinia polysora* differs from common rust (*P. sorghi)* in pustule size, shape and color and is aggressive enough to kill the host, unlike *P. sorghi*. Highly resistant genotypes have smaller uredosori than moderately resistant or susceptible ones. Few uredosori rupture late in the resistant combinations (Subrahmanyam *et al.*1983). Once infection has been induced the disease progresses as the plant develops. *P. polysora* infects the leaves and sheath is severe on late planted maize (Roduel-Rodriguez *et al*, 1980). *Puccinia polysora* was first observed in Western Africa in 1949, and assumed to have come from the Western Hemisphere probably with maize germplasm. Uredospores are the primary inoculum and can initiate epidemics under favorable weather conditions that may account for up to about 60% loss in yield. During the period 1950-51 epidemic, high levels of southern rust were observed in West Africa (Rhind *et al.* 1952). Disease develops rapidly in warm weather and uredospores comprise both primary and secondary inoculum (Bailey *et al.* 1987). The pathogen has the potential of being destructive even if infection comes after anthesis (Raid *et al.* 1988).

Melching (1975) rated *P. polysora* as the most destructive of the rusts of maize. Severe losses can occur especially if infection occurs early (Zummo 1988). All maize grown in E. Africa and tested in the 1950s had no resistant reaction, but those from Mexico had major gene resistance (Storey and Ryland 1954). An isolate of *P. polysora* collected in Kenya from maize cultivar with a B73 background was found to be virulent to *Rp1* gene. Resistant lines normally show small chlorotic or necrotic flecks with no

sporulation. Ullstrup (1965) reported a dominant gene of resistance and designated it as Rpp9. In Kenya three physiologic races known as EA. 1, EA. 2 and EA. 3 have been reported (Ryland and Storey 1955, Storey and Ryland 1954, 1961). Six races were reported in the United States with the possibility of many others thought to occur (Robert 1962).

Maize lines with small pustules surrounded by chlorosis or necrosis were rated as resistant while maize with mature well formed pustules were considered susceptible (Hulbert *et al.* 1991). Rust occurs as small circular to elongate, powdery uredial pustules, initially brown then turn black as the telial stage develops (McGee 1990).

COMMON RUST -*Puccinia sorghi* Schw.

P. sorghi Schw. a macrocyclic heteroecious fungus, the causal agent of common rust, is an obligate pathogen of maize that occurs in all areas of the world where maize is grown. The fungus thrives in high humidity and moderate temperature conditions (Kim and Brewbaker 1976). Production of maize all year round increases the severity of epiphytotics (Brewbaker 1974). Uredospores are the primary source of inoculum and secondary spread (Kim and Brewbaker 1976). *P. sorghi* infects the leaves sheath, and the disease is severe on late-planted maize. Resistance to rust (*Rp)* is expressed as hypersensitive or fleck-like reactions and sources of this resistance has been identified (Hooker *et al.* 1955, Hooker and LeRoux 1957, Hooker 1969, Hagan and Hooker 1965, Hooker and Saxena 1971). Resistance for *P. sorghi* in the United States has been found in maize strains from Australia, Africa, Guatemala, Turkey, Uruguay, Yugoslavia and the United States. The resistance is expressed as chlorotic to necrotic flecks with small uredia (Hooker and Le Roux 1957, Hooker 1962). Two sources were from Kenya identified as Kitale flat white and Njoro flat white (Hooker 1962). A race able to infect maize with Rp genes has been described (Bergquist and Pryor 1984). Common rust has not been very damaging in the United States largely due to the incorporation of resistance genes(Hooker 1969, Melching 1975). Headrick and Pataky (1986) reported that rust developed rapidly at night temperatures of 24^{o}C and 16^{o}C and that on nights with 32^{o}C very few uredia formed but necrotic lesions formed without sporulating. Pathogenesis in resistant and susceptible combinations have been studied and reported (Hilu 1965).

Resistant varieties is the only feasible means of for control of common rust (Bergquist and Pryor 1984).

Phaeosphaeria maydis LEAF SPOT OF MAIZE

Phaeosphaeria maydis, a pathogen of maize, was first described in Brazil, and was later reported in India, Brazil, Colombia, Equador and Mexico (De Leon 1984). In India, the disease was observed in Sikkim and Kalimpong situated in West Bengal and Tarai area located in the state of Uttar Pradesh and was expressed as round, elongate or oblong bleached spots with brownish colored margins on the leaves of Amarillo de Cuba, Ganga Hybrid Makka 3 in addition to other genotypes. Perithecia and pycnidia containing conidia of *Phyllosticta* were observed on the lesions found on the leaf surfaces. Primary inoculum was found in crop debris of previously infected crop (Rane *et al*. 1966). In Kenya, the disease was reported as a minor pathogen of maize needing only medium level attention and hence considered not important (Njuguna *et al*. 1992).

Lesions caused by *P. maydis* are round to oblong and may coalesce and range in size from 0.3 to 2.0 cm (Botrel 1979*). P. maydis* lesions may also appear to be pale green or chlorotic, and become bleached with dried dark brown margins(Shurtlef 1980, Mcgee 1990).

NORTHERN MAIZE LEAF BLIGHT- *Exserohilum turcicum*

Exserohilum turcicum, the incitant of northern leaf blight, is favored by mild temperature and high humidity (Ullstrup 1970). Heavy dews, and frequent rains create good environmental conditions for disease development (Jordan *et al.*1983). Inoculum in previous crop residue has been found to be critical in epidemic build up for subsequent cropping especially in non-tillage systems (Pedersen and Oldham 1992). Levy and Cohen (1983) reported that disease is more aggressive in young susceptible plants with an optimum temperature for infection and lesion number at 20 ^{o}C. Lesion length increases with dew period lengthening and inoculum concentration, while infection occurred at 15-30^{o}C. Levy(1989) reported that pathogenic fitness and environmental conditions are very important in determining severity as the epidemics depend on the ability of *E. turcicum* to infect, grow and sporulate on maize plants. In the continental

United States the disease has been effectively controlled by the use of the dominant *Ht* gene (Smith and Kinsey 1980, Turner and Johnson 1980, Hooker 1961). A new cholorotic halo gene, different from *Ht* gene has been reported by Carson (1995a). This gene is of limited commercial value alone but may be useful in combination with *Ht* genes. Lesion size depends partially on the number of infection sites and the higher the density, the smaller each individual lesion becomes (Nicholoson and Warren 1975). Hooker (1961,1963) identified the chlorotic lesion types of *E. turcicum* resistant maize cultivars which were characterized by chlorosis, late developing lesions with small necrotic center surrounded by a light green margin, and reduced sporulation. Susceptible cultivars expressed rapid developing necrotic lesions with abundant sporulation of *E. turcicum*. The resistance was controlled by a single dominant gene designated *Ht* gene. Homozygous dominant plants rarely exhibit lesions. Similar results were presented by Ullstrup (1963) on line P.I. 217407 where small lesions were surrounded by chlorotic halos with very limited sporulation in resistant genotypes. Further work by Hooker (1963) also concluded that the resistant chlorotic lesion type was conditioned by a single dominant gene in the dent maize inbred line GE440. Results by Hilu and Hooker (1963 and 1965) showed that the initial symptoms after inoculation of inbred and hybrid seedlings of GE440 with *H. turcicum* were similar for susceptible and resistant lines. Within 2-7 days resistant and susceptible reactions appeared as minute white to light green flecks. On susceptible lines, these flecks developed into necrotic lesions that later wilted. No wilting was seen on chlorotic resistant lesions. Full disease development took 15 days. Sporulation was delayed 50-80 hrs and the population of spores per unit area reduced 60 times in the resistant lesions as compared to susceptible lesions. This is a situation normally seen in monogenic chlorotic lesion resistance but not in multigenic environments. Ceballos *et al*. (1991), reported that development of new races shorten the durability of the chlorotic resistant reactions which are controlled by monogenic genes. Polygenic resistance is normally expressed by reduced number of lesions and decrease in lesion size and amount of sporulation (Ullstrup 1970). Singulas *et al.* (1988) reported that average level of resistance, or mean lesion area, the rate of increase in lesion size and the shape of the lesion are influenced by host gene constitution, as determined by contributions of each parent. Lesions induced by *E. turcicum* are long or elliptical, 25 to

150 mm in length (Botrel 1979, Shurtlef 1980, McGee 1990). Lesion size increases in almost a straight line relationship with increasing dew periods and colonization temperatures (Nelson and Tung 1973). Pataky (1992) found that yield losses were significant when disease severity was high on the upper canopy leaves. Levy and Leonard (1990), Raymundo (1978) and Solomonovitz (1992) found that defoliation of the lower third of all the plant leaves showed no yield losses. Leath and Pedersen (1986) found that a cross between resistant B37*Ht3* and susceptible B37 had a severe chrolosis associated with resistant lesions resulting in high area under the disease progress curve values for resistant inbreds with low sporulation and secondary spread.

SOUTHERN MAIZE LEAF BLIGHT-*Bipolaris maydis*

Bipolaris maydis (Nisik.) Shoemaker, occurs as races 0, T and C . In an overwintering experiment, race O was found to have a higher saprophytic ability than race T; only about 4% of the recovered spores were race T (Blanco and Nelson 1972). Race C was reported in China infecting maize with C-cytoplasm (Wei *et al*. 1988). Spore production is influenced by temperature in both races with race T being most sensitive. More lesions formed at 30°C than at 15 or 22.5° C (Warren 1975). Lesion size increases in almost a straight line relationship with increasing dew periods and colonization temperatures (Nelson and Tung 1973). In the 1970s, *B. maydis* induced southern maize blight epidemic caused losses in maize with cytoplasmic male sterility gene (CMS-T) of more than US Dollar 1 Billion (Ullstrup 1972).

The lesions induced by *B. maydis* are oblong with parallel sides or spindle shaped to elliptical and range in size from 0.6 by 1.2-1.9 cm. *B. maydis* lesion size increases in almost a straight line relationship with increasing dew periods and colonization temperatures (Nelson and Tung 1973). *B. maydis* formed more lesions at 30^{o} C than at 15 or 22.5^{o} C.

MATERIALS AND METHODS

GENOTYPE EVALUTION FOR RESISTANCE TO *E. turcicum*

The experiments were done in the research greenhouses of the Kenya Plant Quarantine Station at Muguga where temperatures were constantly at 22^0 C. Cultures of *E. turcicum* were isolated from lesions of diseased maize leaves. One cm square pieces of diseased maize leaf were washed in distilled water and rinsed in sterile water. They were then placed in 10% bleach (sodium hypochlorite) for 1 to 2 minutes and placed between two sterile filter papers to dry out excess bleach. The treated diseased leaf pieces were incubated in moist chambers for 48 hrs. The sporulating pieces were placed in boiling tubes with 10 ml of sterile water and shaken to dislodge the spores. The suspension was serially diluted to 10^{-6} dilution and was checked at X10 magnification. The lowest dilution was then poured into plate of clear nutrient agar (oxoid). Single conidia were picked using a thin inoculating needle under a dissecting microscope. The spores were incubated at 22^{o}C in plates of potato dextrose agar (Oxoid). Culture propagation was done on potato dextrose agar where small blocks of 5mm square were placed at the center of the plates for 7 days. The cultures were ready for inoculation after 7 days and the plates were moistened with a few drops of sterile water and conidia harvested by dislodging them with a microscope slide and were placed in a beaker with sterile water. The spores were filtered through double cheese cloth and the concentration adjusted to 25,000 spores/ml by use of a hemocytometer. Two to three drops of Tween 20 was added to the inoculum as done by Warren (1975). Eight to ten plants for each test variety were planted in the greenhouse in clay pots with sterile loamy forest soil mixed with gravel and peat in the ratio 4:2:1 with a handful of 20:20:20 NPK fertilizer. The plants were ready for inoculation 14 days after emergence or at four leaf stage. They were spayed to runoff in the greenhouse and then covered by polythene sheets presprayed with sterile water for 48 hours. Seedling evaluations were done 15 days after inoculation based on the reactions as done by Hooker, 1961 and 1963 and Pratt *et al*. 1993. The reactions were classified as resistant or susceptible. The Kenyan varieties inoculated in the greenhouse were Pwani 4, Pwani1, Coast composite and Mzihana. Varieties Mo17 and B73 were also included. Length and width of five selected lesions per test plant was determined after 15 days as reported by Leath and Pedersen (1986).

EVALUATION OF MAIZE DISEASE SYMPTOM EXPRESSION IN KENYA

Maize farms were selected randomly with the assistance of Kenyan Ministry of Agriculture extension staff. Before the actual evaluations, we visited some of the maize growing areas of the Western Kenya to facilitate the development of the methods of examination and rating. A farm was selected away from the main roads and a point inside established as sample point. One plant was marked as the starting point. Seven plants were examined to South, North , West and East. Lesions were visually examined, total number of leaves counted, total number of leaves with lesions of each disease, total number of lesions per plant for *E. turcicum* and samples collected for lesion counting and measurement of dimensions. A plant was first examined and then individual leaves thoroughly checked. In most of the cases the diseases occurred in combination on the same leaves of a single plant. There were cases where lesions of different diseases overlapped but the selected individual lesions for a particular disease were always clearly defined or could be traced. The sizes of well defined lesions were determined by use of a ruler in millimeters and caution was taken to make sure that single isolated lesions with clear boundaries were measured. Initial infection foci were used to determine the extent of single lesions. Samples were taken for lesion size determination where five lesions induced by *E. turcicum* were measured per site for a total of 63 sites or 315 lesions, ten lesions induced by *P. maydis* for 35 sites or 350 lesions, and ten lesions induced by *B. maydis,* for 27 sites or 270 lesions. Lesion length and width of inoculated test plants were also examined and measured in millimeters. Analysis of variance and means comparisons using the Tukeys Studentized Range at $\alpha = 0.05$ was done using the General Linear Model. Data was analyzed after a square root transformation (SAS Institute, Cary, NC).

RESULTS

VARIETAL AND INBRED LINES RESPONSES TO ISOLATES OF *E. turcicum* IN KENYA

All inoculated plants developed pin head chlorotic spots after 48 hrs and later after 72 hours water soaked lesions developed in the susceptible reactions. Susceptible lines had big necrotic lesions that were water soaked with dark margins. The resistant reactions were characterized with chlorosis without extensive necrosis. The Coast area land race Mzihana exhibited severe chlorosis accompanied by necrosis with three isolates (Tables 1, 2).

Among the improved Kenyan lines only Pwani 1 was resistant to three isolates; Pwani4, Coast Composite and B73 a known suscept were all susceptible to all isolates while Mo17 the polygenic resistant was susceptible to two. The Coast local land race Mzihana was susceptible to two isolates from the Coast and resistant to three isolates from the Rift Valley. The lesions were longest in the susceptible inbred line B73 as the known suscept for comparison purposes with a mean length of 110 mm and a mean width of 3.4 mm while Mo17 had the smallest lesion length mean of 10 mm by 1.5 mm. (Table 3 and Figure 1). The improved coastal line Pwani 1 and Mzihana were seen as probably having resistance for they showed considerably small non-sporulating lesions. Tables 1and 2 represent the severity and reactions types respectvely and Figure 1 represents lesion sizes on susceptible plants.

MAIZE DISEASE SYMPTOM COMPLEXES IN KENYA

E. turcicum induced lesions were counted on twenty eight plants per location. Analysis of variance showed significant differences in lesion number between locations at $P<.0001$ (Table 6). Embu District in the mid altitudes had the highest number of lesions while the lowest number was in Njoro of Nakuru District. The largest lesions were induced by *E. turcicum* with a mean area of 8,279 sq mm (length x width) (Table 7). These were recorded at Njoro in Nakuru District on a variety PANA from South Africa (Figure 6). The lesions were long elliptical or keel shaped, with brownish necrotic zones more or less aligned to the main axis of the leaf. Individual lesions occurred singly or coalesced with others to give a blighted appearance. Extensive and sporulating lesions

on the husk and leaves on a severely infected Mzihana cultivar maize plant found in the Coast Province serves as an example of the severity of northern leaf blight as observed in the Coast Province (Figure 5). Frequently, many leaves had extensive lesions from the point of attachment to the tip which is an indicator of weak genetic resistance in an environment highly conducive for disease development and spread. Lesion expansion is directly related to the length of the dew period (Nelson and Tung 1973).

In the humid areas of the Coast and Lake Victoria region, *E. turcicum* induced lesions were observed sporulating profusely, with the lesion edge appearing dark with a conidial mass cover. Nakuru District had the largest lesion length mean of 284.4 mm and a mean width of 23 mm. This was found on PANA hybrid and also the smallest lesion length mean found on Hybrid 622 recorded as 51 mm by 8 mm which was an indication of resistance although this hybrid had severe rust infection (Table A18 in Appendix A; and Figures 6, and 9). The largest lesion measured was 452 mm long and 25 mm wide at Njoro on a newly introduced variety from South Africa (Figure 6). Leaves with lesions that occurred near the axil appeared pale. Many of the lesions were concentrated on the mid-third of the canopy with fewest at the top, while the lower leaves were wilting. The husks were also infected. The lesions were gray to whitish in color with a brownish border but sometimes a slight cholorosis was observed near the margins. Many lesions in the humid area had profuse sporulation (Figures 5, and 10). Leaves that were severely infected by *E. turcicum* rarely had other lesions induced by other pathogens, although, a few could be seen along the edges on green areas (Figures 8 and 10).

Analysis of variance of lesion areas (length x width) for *E. turcicum*, *P. maydis* and *B. maydis* showed significant differences between locations which is an indication of the local variety susceptibility and suitable environmental conditions for disease development respectively (Tables 4, 6, 8, 10 and Figures 2, 3 and 4).

P. maydis produced numerous lesions on the leaf lamina in all areas where maize was evaluated except the lowland coast. Many lesions coalesced and resulted in leaf wilting bearing blighted bleached, whitish, spotted zones. The whitish necrotic spots 1 to 10mm long by 1 to 4mm wide were surrounded by a dark border and frequently covered the entire leaf surface. Kirinyaga and Embu Districts had serious levels of *P. maydis* infection resulting into epiphytotics characterized by large spots exceeding 7 mm by

4mm covering the leaves. The spots were diffused over the leaf lamina with or without chlorosis and not confined between the veins (Figure 15). In the highlands, *P. maydis* was more severe in the upper third of the canopy and lesions could be found on the top most leaf. In central Kenya the distribution of lesions showed no pattern but was more or less diffuse all over the canopy (Figure 15). *E. turcicum* induced lesions were smaller on plants where lesions of *P. maydis* occurred densely (Figure 14). Some lesions of *P. maydis* developed on top of already established lesions of *B. maydis* (Figure 17).

The ANOVA indicated there were highly significant differences in lesion sizes in the various location at P<.0001 (Table 18). The comparison of means also showed differences in areas of lesions observed at α =.05 (Table 9), where the wetter humid areas had the most severe disease incidence. Nakuru District once again had the largest lesion areas with a mean of 21 sq mm induced by *P. maydis* followed by Kirinyaga with a mean of 17 sq mm and Thika with a mean of 15 sq mm in the mid altitudes. Embu district had the smallest lesion mean area of 3.22 sq mm (Table 7 and appendix A, Table A18).

B. maydis induced long or elliptical slender streak like necrotic spots, that were brownish in color occurring between and pallarell to the leaf veins about 1 to 60mm long and 1 to 5mm in wide. In the highlands where *B. maydis* infection was predominant, the leaves were variably blighted with numerous interveinal longitudinal lesions accompanied by limited chrolosis.

Both *P. maydis* and *B. maydis* could be found on the same plants especially on the lower leaves. *P. maydis* tended to increase towards the top while *B. maydis* towards the bottom of the canopy (Figures 11, 13, 16, and 17). This phenomenon may be explained by the methods of reproduction especially that *P. maydis* produces pycnidiospres which are likely to float higher up than the spores of *B. maydis* in the same micro-environment or be forcefully injected into the air stream from the fruiting bodies. The ANOVA for lesions induced by *B. maydis* is provided and shows significant difference at P<.0001 (Table 10). The comparisons of means indicated differences between locations at α =0.05 where Embu District had the largest disease lesion area with a mean of about 90 sq mm and Kakamega District at Musembe the smallest with a mean of 2.31mm (Table 11).

P. sorghi rust pustules were small rusty spots surrounded by a yellowish halo with or without torn leaf cuticle. In the highlands *P. sorghi* rust infection produced big irregular brownish rusty areas on the leaves with marked chrolosis and occasional blighting, erumpent pustules and wilting of huge leaf zones (Figure 9). *P. polysora* was confined to the Kenya coast but some isolated cases were seen in Kirinyaga District. In the coast, *P. polysora* was found sporulating profusely and the rusty yellowish pustules on the leaf surfaces occasionally exhibited chlorosis. Figures 2, 3, and 4 and appendix A 18, A19 and A 20 represents the lesion sizes as determined from field samples. Tables of total number of leaves and number of diseased leaves, ANOVA and means comparison are provided in the Appendix A1 to A16 for all diseases and all showed significant differences between locations.

DISCUSSION

Maize crop in all areas of Kenya was found to have a heavy pathogen load that undermined the leaf area and weakened the plants resulting in very poor crop stands. Favorable environmental conditions operating on poor disease resistance genome generated epidemics involving *E. turcicum, P. maydis, B. maydis P. sorghi* and *P. polysora.* Ideally, lowland rust is favored by 16 hours of free moisture on the plant for germination of uredispores (Hollier and King, 1985). In the 1950,s devastating yield losses of 50-60% occurred in various production areas of Africa due to lowland rust (Futrell, 1975).

Our results show that ideal temperature and moisture conditions coupled with susceptible genotypes could result in disease yield losses approaching those in the 50's. After evaluations of Ugandan maize germlasm for resistance to *E. turcicum*, Adipala *et al.* (1993) reported that all had necrotic susceptible reactions when inoculated with races 0, 1, 23 and 23N but the observed resistance had no resemblance to the known *Ht* gene expressions.

While the Kenyan Maize Improvement Program has done a tremendous job in producing maize varieties for the various ecological zones, it is evident the aspect of disease control by genetic resistance must be allotted more attention. This is because all the recommended varieties of 600, 500 series, Coast composite, Pwani series and all land races, etc were found to be very susceptible to two or more of the pathogens observed in the fields. It was interesting to observe Mzihana, a land race of the lowland coast in Kenya was resistant to isolates from the highlands while susceptible to coastal isolates. This tends to suggest that the races in the highlands are different from those in the lowlands and also points to selection based pathogen host interaction. In the continental United States the disease has been effectively controlled by the use of the dominant *Ht* gene (Smith and Kinsey 1980, Turner and Johnson 1980, Hooker 1961). Report by Smith and Kinsey (1980) suggested that a combination of *Ht* and *Ht2* or *Ht3* would provide resistance against race 1, 2 and 3. Report by Pataky (1994) showed that high levels of partial resistance with or without *Ht*-genes presented a spectacular approach in reducing damage from NLB on sweet corn which also eliminates the severe yield depressing chlorosis associated with *Ht* gene resistance in very susceptible backgrounds.

Studies by Carson (1995b) indicated that latent period is related to partial resistance which suggested that selection for increased latent period length would be more beneficial than selecting for reduced disease severity. Selection for increased latent period length can be done in environments without severe disease epidemics, and also breeding material could be assessed as seedlings for latent period length in the greenhouse during the off season. Levy (1991) showed that isolates from different areas were of different parasitic fitness as was indicated by infection efficiency, sporulation and lesion size, while isolates of same the location showed less variation. Inoculum in previous crop has been found to be critical in epidemic build up for subsequent cropping especially in non-tillage systems as reported by Pedersen and Oldham (1992) using race 2. While non tillage is not a common practice in Kenya, the heavy inoculum production means a lot of primary inoculum in subsequent plantings. According to Gevers (1975) the *HtN* major gene of resistance derived from the Mexican maize variety *Pepitila* is reasonably stable, but in some parts of the world the effects may fail to be expressed and genetic segregation may not behave like expected of dominant genes ratios, but does however remain in the tolerable limits of deviation of stability and segregation. He suggested the occurrence of biotypes in India able to overcome *HtN* gene resistance. The *HtN* gene is also background sensitive, and high temperatures reduced symptom expression on line B37*Ht3*. The Plants were evaluated at both 26^{o}C day/22 ^{o}C night and 22 ^{o}C day/18^{o} C night temperatures. There was observable weakened virulence at high temperatures. Combining *Ht1* and *Ht3* genes did not result in significantly less disease from those homozygous for each gene (Leath and Pedersen 1986). It appears that in Kenya the elite hybrids in the maize belt of the Western region were of low rust resistance backgrounds as evidenced by extensive chrolosis. Dunn and Namm (1980) reported gene dosage effects for the *Ht2* gene. Report by Smith and Kinsey (1980) suggested that a combination of *Ht* and *Ht2* or *Ht3* would confer resistance against race 1, 2 and 3.

Lesion size is one of the components of susceptibility of a host to a fungus disease (Populer 1978). Hamid *et al*. (1982) used lesion length to estimate the resistance of several maize inbred lines to different isolates of race 3 of *B. zeicola.* We examined field symptom expression as an attempt to gauge levels of resistance in the field as could be

indicated by lesion sizes and complexes of the various diseases. Our data present for the first time record of extremely large lesions of over 40cm long and 2.5cm in width for *E. turcicum*, 6 cm by 0.5cm for *B. maydis* and 1cm by 0.4 cm for *P. maydis*. Njuguna *et al*.(1992) reported lesion size for *E. turcicum* to be between 5 to 10cm long and 1.3cm wide in Kenya . However, we present evidence for the first time that lesions of over four times in size than those previously reported were observed in Kenya. Most of the lesions were observed in the upper canopy zones of the plants and hence may adversely affect yield. Pataky (1992) found that yield losses were significant when disease incidence was severe and present on the upper canopy. Their data agree with that of Levy and Leonard (1990). Serious yield depression in Kenya is therefore expected because most of the lesions were found in the upper third of the canopy. The critical population of leaves needed by the plants for dry matter production is in the upper canopy and is related to the final yield (Pataky 1992). Raymundo (1978) and Solomonovitz (1992) found that when the lower third of all the leaves were removed, no yield loss was observed. There is a clear indication that maize varieties grown in Kenya are very susceptible to all the pathogens inducing the observed diseases in this study. Many of the lesions were sporulating which is an indication of susceptibility (Hooker 1961), while extensive chrolosis was seen on some hybrids infected with *P. sorghi*. Maize observed in the Molo and Njoro areas of the Rift Valley exhibited severe chlorosis associated with rust infection, which probably may indicate a highly susceptible line was used for hybrid production. The Kenya environmental conditions were very conducive to disease development and therefore we suggest genotype evaluation for resistance be done under severe inoculum pressure and favorable environment. The locations with high rainfall and high humidity had severe disease, and more lesions which were sporulating. All genotype screening should be done for more than one disease per ecological zone as is the standard practice for maize breeders.

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