

CONTENTS

Introduction	1
Review of Literature	3
Materials and Methods	22
A- Survey of sorghum downy mildew	
B- Disease syndrome & causal agent	
C- Evaluation of Inoculation methods	
1-Oosporic inoculum	
2-Conidial inoculum	
D- Field screening of selected corn genotypes .	
E- Comparative reactions to pathogen propagules .	
F- Protein (SDS – PAGE) analysis of corn inbreds .	
G- Host range of <i>P.sorghii</i> in Egypt .	
H- Effect of fertilization and rate of application on SDM .	
Experimental Results	37
A- Survey of sorghum downy mildew	
B- Disease syndrome & causal agent	
C- Evaluation of Inoculation methods	
3-Oosporic inoculum	
4-Conidial inoculum	
D- Field screening of selected corn genotypes .	
E- Comparative reactions to pathogen propagules .	
F- Protein (SDS – PAGE) analysis of corn inbreds .	
G- Host range of <i>P.sorghii</i> in Egypt .	

H- Effect of fertilization and rate of application on SDM .

1-Under field infection .

2-Under artificial infection

Discussion	71
Summary	81
Literature Cited	88
Appendix	
Arabic Summary	

INTRODUCTION

Summary

Sorghum downy mildew, in Egypt, is incited by *Peronosclerospora sorghi*. The disease imposes an increasing hazards on corn and sorghum plantations, especially in lower Egypt. The disease causes bareness of infected plants, consequently, loss in seed production. Infection rate is increasing year after year wherever the environmental conditions are favourable.

The current study was designed to survey the disease in two important governorates of lower Egypt, to study the disease syndrome and confirming the pathogen identity, to compare the different methods of host plants inoculation, to screen some local corn genotypes for SDM resistance, to compare the reaction of some corn hybrids with their parental lines, to study the changes in proteins of resistant and susceptible corn inbred lines at different growth stages after inoculation, to study the host range of *P.sorghi* in Egypt, and to study the effect of fertilization regimes on disease incidence under natural and artificial inoculation.

The result of this study could be summarized in the following:-

A-The data collected during the survey (1998-1999) on corn revealed that the disease was found at six out of seven tested districts at Kafr El-sheikh governorate. Severity of the disease ranged from 0.0% to 15% during the two seasons with an overall mean of 7.9%. The highest mean incidence (15%) was recorded from Kafr El-sheikh district. El-hamoul district was found free from SDM. Considering the cultivated hybrids, the highest means of disease incidences were 13.6% and 12.9% in 1998 and 1999 with average of 13.3% observed

on the white T.w.c 310, whereas the lowest means were observed on the white S.c 122 (1.9 % and 1.3% in 1998 and 1999 with average 1.6%).

In Behera governorate, SDM was found in all the six tested districts. Disease incidence ranged between 3.7% and 12% during the two seasons with an overall mean 7.6%. The highest mean incidence (12%) was recorded at Shobrakheit district, while the lowest (3.7%) was recorded at Abu-El- matamir. Considering the cultivated hybrids, the highest mean disease incidence (12.6%) was observed on the T.w.c 310, whereas the lowest mean (1.25%) was observed on the S.C 122.

B- Disease syndrome on corn and sorghum was affected by growth stages. Initial symptoms, on both hosts, were expressed as plesionicrotic chlorosis at leaf base. Chlorosis occurs usually at the second-leaf and above. The new leaves, of systemically infected sorghum plants, exhibited complete chlorosis then striping with yellow or white stripes alternating with green ones parallel to leaf midrib. A white downy growth, characteristic to this disease, could be observed in the early morning on the undersurfaces of the infected leaves. This growth was found to be the conidiophores bearing conidia. Later in season, the white stripes turned into brown-reddish colour due to oospores formation. At maturity the infected leaves shred due to necrosis of the strips and the pressure of the formed oospores. The infected plants usually give sterile heads or poor seed-setting.

Chlorosis on infected corn plants may advance to cover the whole blade or be restricted to the basal part only with distinct margin between chlorotic and green portions which is called " half-leaf"

disease. The infected leaves were erect and narrow. The characteristic white downy growth could be observed on the undersurface of the infected leaves. Tassels of the systemically infected corn plants may express proliferation and phyloidy. An additional tassel may grow in place of the ear. Ear shank may be abnormally long. The plant may be barren giving no ear at all which, certainly, is a loss of seed yield.

The microscopic examination revealed that the fungus produced asexual true conidia on stout conidiophores. These were up to 250 μm tall, tapered towards a bulbous basal cell which was separated with a septum. The conidiophores were dichotomously branched on the top and end with tapered strigata, each bearing a single conidium. The conidiospores were hyaline, oval shape and measured 15X 27 μm to 18X 30 μm .

The sexual oospores were formed by late season. They were rare or absent in corn but, in sorghum, a lot of dense lines of oospores were observed in the mesophyll tissues between the fibro vascular bundles. The oospores were almost spherical, reticulately ornamented and measured 30 to 35 μm in diameter. They were light yellow, surrounded by dark layers, most probably the walls of the oogonium.

C- Data from inoculation methods showed that conidia were more efficient than oospores in inducing infection to both tested hosts. The most reliable oosporic inoculation method tested in this study was the usage of naturally infested soil (24% and 19.5% on sorghum and corn, respectively). Incubation of host seeds with the fungal oospores on moistened filter paper at 25 °C was less efficient (18.2% and 13.5% on sorghum and corn, respectively). Other tested methods were significantly less efficient than the previous two methods.

All conidial inoculation methods tested were successful in inducing higher disease incidence in both sorghum and corn compared to oosporic inoculation methods. The most significantly higher disease incidences on sorghum: 86%, 80.7%, and 75.2% were recorded from spray second leaf seedlings; spray sprouted seeds, and dropping in whorl methods, respectively. Dipping sprouted seeds in the conidial suspension was the least (43.9%). The highest significant disease incidences, on corn, were obtained through spray sprouted seeds and second leaf seedlings (58.3% and 53.8%, respectively). The least disease incidence was obtained through dipping sprouted seeds in the conidial suspension for 10 min.(33.3%).

D- The data concerning evaluation of corn genotypes reactions to SDM disease revealed that some inbred lines could be regarded as highly tolerant or resistant, e.g., Sd 62 (7.1%) and Sd 63 (6.6%). Others were moderately resistant, e.g., Sd 7 (19.2%) , Gz 603(16.7%), and Gz 628 (15.9%), susceptible, e.g., Sd 58 (36.2%) and Gz 650 (41.8%), or highly susceptible ,e.g., Gz 617 (59.9%), Gm.1002 (55.1%), and Gm.1021(68.5%). Seed yields of these inbred lines were mostly affected by disease incidences.

Among the single crosses tested in this study, S.c 107 and S.c 122 were found to be resistant under the experimental conditions (9% and 8.5%, respectively), at the same time, both were significantly higher in seed yields (7.71 and 8.58 kg/plot, respectively). On the other hand, S.c 10 and S.c 155 (43.8% and 31.3%, respectively) were susceptible and S.c 161 was highly susceptible to SDM (61.7%) and produced less seed yields (6.45,5.47 and 4.63 kg/plot, respectively).

Also, among the three way crosses, tested in this study, T.w.c 322 was found to be more resistant to SDM (6.9 %) and gave the highest seed yield (7.27% Kg/plot) whereas, T.w.c 310 was highly susceptible (68.8%) and gave significant low seed yield (4.9 Kg/plot). Other three way crosses were either moderately resistance (T.w.c 321), moderately susceptible (T.w.c 323), susceptible (T.w.c 320 and 324).

E- Reaction of corn hybrids and their parental lines to oosporic and conidial inoculum of *P.sorghi* indicated that corn genotypes resistant to oosporic inoculation were, in most cases, resistant to conidial inoculation at two-leaf stage. For example, Gz 603 and Gz 628 were resistance to oosporic inoculation (0.0% and 4.7% respectively) and were, also, resistant and moderately resistant to conidial inoculation at second-leaf stage (6.7% and 14.9%, respectively). The S.c 122, involved both resistant parents, was found to be resistant too (5.9%). Meanwhile, the yellow inbred lines Gm 1002 and Gm 1021 were, both, moderately susceptible to oosporic inoculation (21.2% and 20.9%, respectively), and highly susceptible to conidial inoculation (81.4% and 77%, respectively). The S.c 155, involved both highly susceptible parents, was found to be highly susceptible (53.9%). The S.c 10 was an exception in this study, it was found to be susceptible to the conidial inoculation (34%), although it involved two moderately resistant inbred lines; Sd 7 (15.3%) and Sd 63 (16.6%).

F- Results of studying buffer soluble protein, extracted from fresh leaves of resistant (Sd 62) and susceptible (Sd 58) corn inbred lines at different growth stages after inoculation and analysis by SDS-PAGE,

showed marked differences in the number and intensities of the protein bands. However, regardless of inoculation treatment, two protein bands of MW's 52 and 29 KDa were more intensive in the susceptible plants of (Sd 58) at two and four-leaf stages. On the other hand, some protein bands were detected in certain inbred at certain growth stage as different transient gene expression in each of the tested inbreds.

Regarding inoculation treatment, a polymorphic protein band of MW 63 KDa was detected in the inoculated resistant inbred plants (Sd 62) at two-leaf stage, which might be assumed related to resistance at this early stage. Another polymorphic band of MW 43 KDa was detected in the inoculated plants of (Sd 58) at four and five leaf stages, which assumed to be responsible for susceptibility at those late stages of growth.

Polymorphic protein bands of MW's 20, 17 and 13 KDa were marked at different growth stages, regardless of inoculation. A steady increase of protein bands; 20 and 17 KDa, at two and three leaf stages, then steady decrease at the following growth stages was observed. Meanwhile, steady decrease in a band of 13 KDa at two and three-leaf stages, then steady increase in the four and five -leaf stages. These might be related to different gene expressions at different developmental stages in both inbred lines.

G - The host range study demonstrated that *P.sorghii* had a narrow host range and was restricted to plant species that belong to *Sorghum* and *Zea* spp. Disease incidence due to oosporic or conidial inoculations were: 23.1% and 78.6% on *Sorghum bicolor* (grain sorghum), 22.2% and 54.8% on *S. bicolor* (green fodder), 20.8% and

72.4% on *S.bicolor* (sweet sorghum), 42.9% and 60% on *S.sudanense* (Sudangrass), and 31.3% and 82.1% on *S.halepense* (Johnson grass).

The disease incidences in *Zea spp.* due to oosporic and conidial inocula, were: 13.3% and 63.2%, and 15% and 45.5% on *Zea mays* and *Euchlaena mexicana* respectively.

H- Fertilization with one single nutrient element (N, P, or K) resulted in higher SDM incidences compared to any combinations of these three major elements. Means of disease incidence under natural infection were 32.7%, 26.2%, and 25.3% for nitrogen, phosphorous, and potassium, respectively, while it was 23.1%, 19.1%, and 12.6% for (NPK)₁, (NPK)₂, and (NPK)₃, respectively. Means of barren plants were not significantly different. Seed yields were 2.061, 2.280, and 2.418 kg/plot for nitrogen, phosphorous, and potassium, respectively, while it was 2.523, 3.353, and 3.608 kg/plot for (NPK)₁, (NPK)₂, and (NPK)₃, respectively.

Under artificial infection, where numerous conidia were released by Sudangrass infector row, the same trend was observed. Disease incidences were higher at single nutrient element treatments (63.9%, 55.4% and 59.9% for N, P, and K, respectively) compared to significantly lower disease incidences: 48.4%, 38.8%, and 35.7% for (NPK)₁, (NPK)₂, and (NPK)₃, respectively. Seed yields were similarly different. Means of seed yield were 1.342, 1.599, and 1.402 kg/plot for N, P, and K, respectively, while these were 1.975, 2.976, and 3.250 kg/plot for (NPK)₁, (NPK)₂, and (NPK)₃, respectively.