PHYSIOLOGICAL STUDIES ON DISEASE RESISTANCE AND SUSCEPTIBILITY OF VULGARE WHEAT TO STEM RUST

By

YEHIA YOUSEF EL-HYATMY

B.Sc. Cairo University, 1961

M.Sc. Ain Shams University, 1969

THESIS

Submitted in Partial Fulfilment of the Requirements
for the Degree of

DOCTOR OF PHILOSOPHY

in

Plant Physiology

Plant Pathology Department
Faculty of Agriculture
Ain Shams University

1973

APPROVAL SHEET

This Thesis for the Ph.D. Degree

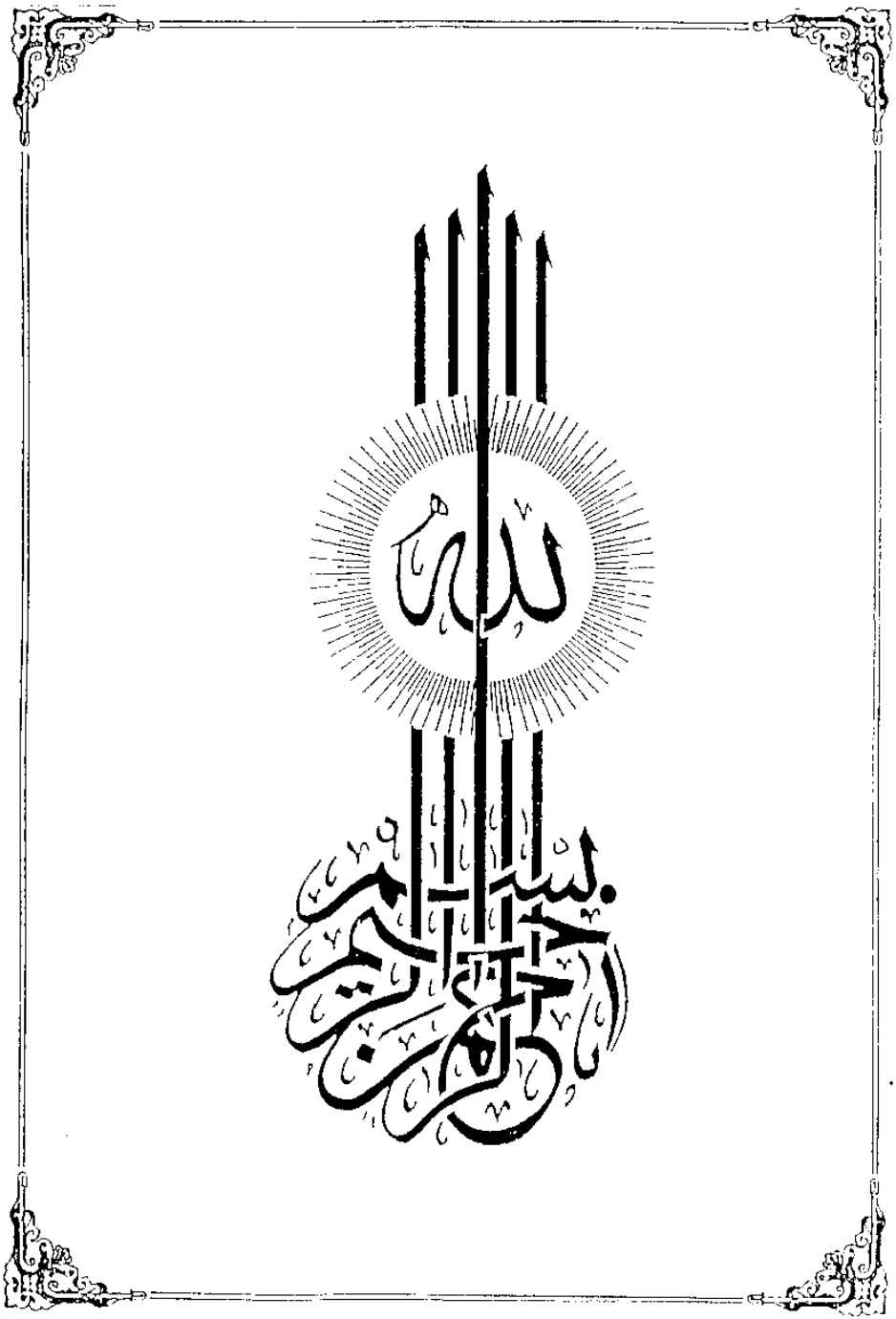
has been Approved by :

Tewfik Abdel Hak

M. Kad

Date : / / 1973

...ooOoo...



A C K N O W L E D G M E N T

This work has been carried out in the Department of Plant Pathology, Faculty of Agriculture, Ain Shams University under the supervision of Dr. A. Kadry (now on leave), Dr. M.A. Abdel-Halim, Associate Professor of Plant Physiology and Dr. El-Sayed H. Hassanein, Associate Professor in the Genetics Department.

The author wishes to express his deepest gratitude and sincere appreciations to Dr. M.A. Abdel-Halim and Dr. El-Sayed H. Hassanein for their supervision, guidance and encouragement throughout the course of this study.

Thanks are also due to the Director and Staff members of the Crop Physiology Research Department, Min. of Agriculture, A.R.E. for their cooperation and continuous help during this investigation.

...ooOoo...

C O N T E N T S

...ooOoo...

I N T R O D U C T I O N

Stem rust of wheat (Puccinia graminis tritici) is considered the most important and destructive disease attacking wheat, wherever it is cultivated. It occurs widely in the Near East and still remains a major problem, although many resistant cultivars have been produced. It is more common in the areas where wheat is planted under the irrigation system and where rain precipitation is high.

In agriculture, disease resistance is usually thought of as a cultivar phenomenon, and the breeding of disease - resistant cultivars of crop plants forms an important aspect of crop improvement. cultivar resistance appears to have dominated for more than half century. The cumulative results of their efforts to understand the mechanisms involved have been very small.

Disease resistance mechanisms are in fact present in all plants, even those which are known to be highly susceptible to specific diseases. This point is very important as it suggests that cultivars, resistance to specific diseases conform only one aspect of the much broader phenomenon of immunity. Such considerations have not been taken into

account in the interpretation of most results which have been reported in this field.

Various environmental factors may influence disease resistance namely, the effect of light, temperature, osmotic pressure and the permeability of the plasma membrane were inversely correlated with resistance to wheat stem rust. In addition, the nutritional balance of mineral salts in different soil conditions has also been reported frequently to offer an essential factor playing an important role in such respect.

The problem of host-parasite interactions, however, cannot be ignored if a satisfactory explanation of disease resistance mechanisms in plants is to be achieved. Their elucidation remains one of the major challenges and one of the most exciting fields in plant pathology.

The material used in this investigation was taken from the view of geneticists to have the same genetic background in order to determine the trend of resistance in the sister lines descendants from two cultivars.

It has been reviewed that the effects of infection on nucleic acids and protein synthesis, and on the formation and release of phenolic compounds would appear to be among the most rewarding topics for investigation (Shaw, 1963).

In order to meet with the above call, the present study was put forward as a humble effort to throw some lights on the complicated relationships between stem rust resistance and the different chemical components of the host plants in connection with their genetical constitutions.

REVIEW OF LITERATURE

Rusts (Uredinales) and powdery mildews (Erysiphaceae) include the causal organisms of many of the most important plant diseases. Both groups are obligate parasites, since they attack only the living tissues of their hosts and cannot as yet be grown routinely from spores on a synthetic medium. In studying host-parasite relations the ectoparasitic mildews offer the advantage that they are, except for the haustoria, readily separated from infected leaves, a feat not easily accomplished with the endoparasitic rusts.

The rusts and mildews exhibit an extreme degree of specialization of parasitism epitomized by Puccinia graminis While Puccinia graminis tritici Erikss. and Henn. is able to attack several cereal hosts, over 200 physiological races of it are at present distinguished by their uredinial reaction on the so-called differential cultivars of wheat. The genetic basis of specialization in the rusts and resistance in the hosts has been recognized virtually since the rediscovery of Mendel's laws, but it was not until 1927 that Stakman discovered the sexual stage in the life cycle of rusts, thus initiating studies on the genetics of these fungi.

Working with the flax rust fungus, Melampsora lini and its uredinial reaction on Linum usitatissimum, Flor (1956), recognized that it was possible to study the genetics of both members of the host-parasite system simultaneously. He was able to show that for each gene that determined resistance in the host, there was a specific related gene that determines virulence in the rust. Person (1959) pointed out that the evaluation of a gene - for gene relationship in a host - parasite system is an automatic result of mutation and natural selection for resistance in the host and for virulence in the parasite. He therefore, postulated that gene-for-gene relationships are the general rule in host-parasite systems and developed methods for analyzing them.

It is evident that knowledge of the genetic basis of host-parasite interactions is well advanced. It follows that physiological and biochemical studies on the rusts have a two-fold objective : to discover if possible why those fungi fail to develop from spores in axenic culture, and to investigate the biochemical basis of specialization and resistance, though it may be said at once that the gap between the genetic information and satisfactory answers to those questions is still a large and challenging one.

The host-parasite relations of the rusts and mildews

were reviewed earlier by Allen (1953 and 1959 a), who had also discussed the biochemistry of resistance. Other authors have dealt with the respiration of diseased plants (Urikami and Akazawa, 1959 and Millerd and Scott, 1962); as well as several other aspects of the physiology of disease (Farkas and Kiraly, 1962).

Physiological Aspects of Resistance and Susceptibility :

Resistance may take the form of exclusion (usually morphological rather than physiological), growth restriction after entry, or destruction after entry (Hart, 1949).Walker (1959), reported that the exclusion seems of minor importance, since usually the pathogen enters both resistant and susceptible hosts in the same manner, the expression of resistance appearing after invasion.

Various envrionmental factors may influence resistance. The effects of light and temperature on the infection of wheat by stem rust reflect the action of these factors on the differentiation of infection structures by the fungus. Hart and Forbes (1935), indicated that wheat stem rust but not leaf rust is an exception. It requires open stomates for penetration, and certain wheat cultivars with "functional resistance" against the forms exclude their germ tubes by having closed

stomates. This is a case of physiological surface resistance. They added that the light by its effect on stomata, may promote infection by stem rust. Yirgou and Caldwell (1963), also found that, light may aid infection by direct photosynthetic reduction of carbon dioxide within the leaf, since removal of CO_2 allowed penetration of closed stomata in the dark by P. graminis tritici and added CO_2 suppressed it.

Thatcher (1942),observed that the higher osmotic pressure in the host cells would make nutrients and water unavailable to the fungus, the ability of the parasite to thrive in the tissue of the host is correlated with a higher osmotic pressure in the invading cells. Alterations of resistance by shading, mineral nutrition, age and floating detached leaves on water were all correlated with changes in osmotic pressure.

Sen and Joshi (1957), stated that the permeability of the plasma membrane was positively correlated with susceptibility to wheat stem rust, possibly by making nutrients more available to the parasite. Permeability was correlated with electrical conductivity, which in turn was inversely correlated with resistance.

In this review, the following topics will be discussed:

,1) Hypersensitivity :

According to the gene-for-gene theory of rust resistance in flax, Flor (1955 and 1956), reported that each gene for rust reaction in the host has a complementary gene for pathogenicity in the parasite. The resistance in host and avirulence in the parasite are usually dominant traits. An antigen-antibody mechanism is suggested wherely antagonistic proteins are produced by complementary dominant genes, the result being a hypersensitive (immune) reaction. Anonymous (1960) and Doubly et al. (1960), reported that the susceptibility and resistance were related to the presence and absence of globulin antigens in flax lines. A rust race was virulent to flax cultivars containing its specific antigens and avirulent to cultivars lacking the antigen. This complementary gene theory may explain why resistance is much more common than susceptibility, since avirulence means resistance in either a resistant or a susceptible host, whereas virulence usually means susceptibility only in a susceptible host. In some instances a parasite with a gene for virulence can overcome the action of a complementary host gene for resistance.

Brian (1955), Braun (1959) and Braun and Bringle (1959), have reviewed pathogen - produced toxins as factors in the physiology of disease. Evidence of a correlation

between toxin production *in vitro* and pathogenicity is accumulating, but there are many exceptions. Substances shown to be correlated with disease symptoms (found by the pathogen *in vitro* and productive of typical symptoms when introduced into the host) include oxalic acid, lycomarismin, methionine sulfoxime, fusaric acid, vasinfuscarin, ethylene, indole acetic acid and victorin, Woolley (1959), indicated that some disease symptoms appear to result from the induction of specific deficiencies in the host as a result of extracellular toxins of the invader.

Allen (1959b), stated that, to demonstrate a substrate is associated with resistance to a certain disease, it must be isolated from resistant hosts at the site where infection occurs, then reintroduced into susceptible hosts where it must confer resistance similar to natural resistance. It may act by inhibiting spore germination, host penetration , or vegetative growth of the parasite.

Burrows (1960), made leaf sandwiches by stripping off the epidermis of two oat leaves, coating the exposed mesophyll cells with 0.5 percent agar, and then cementing them together with paraffin on the edges. When susceptible leaves were used, stem rust mycelium spread from one to the other. Resistant leaves do not become infected from susceptible leaves but transferred both the inoculum and ^{32}P to a susceptible