



Association of stripe rust (*Puccinia striiformis* west.) disease index with grain yield, growth rate, harvest index and other agronomic characteristics of wheat (*Triticum aestivum* L.)  
by Muhammad Aqil Khan

A thesis submitted in partial fulfillment of the requirements for the degree of DOCTOR OF PHILOSOPHY in Crop and Soil Science  
Montana State University  
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**Abstract:**

The relationship of grain yield, growth rate, harvest index and other agronomic characteristics of wheat (*Triticum aestivum* L.) in disease and disease free conditions was examined using three crosses each of spring and winter wheat with two generations in each, i.e., (Formula not captured by OCR), respectively. Effects of stripe rust (*Puccinia striiformis* West.) on plant traits and their relationships in control and inoculated hill plots were studied using ratios (inoculated/ control), phenotypic correlations, multiple regression, path coefficient analyses and heritability estimates.

The wheat lines from five of the six crosses used in these studies were developed previously emphasizing only the accumulation of additive genes for stripe rust resistance. This resulted in a decline in grain yield and other agronomic traits in subsequent generations. Concurrent selection for both agronomic characteristics and disease resistance is suggested to avoid erosion of variability for important agronomic characteristics.

Disease affected the expression of most of the traits studied, with effects being progressive with the increasing disease intensity. Disease effects, however, varied depending upon the time of infection. Infection reduced the growth of the plants and significantly lowered biological yield in infected lines. Maximum effects of infection in both experiments were noted on yield. Kernel weight was less affected by the infection compared to the other traits.

There was little effect of disease in changing the correlations of grain yield with other traits. Kernel weight, however, showed higher correlations with growth traits and grain yield in the inoculated than in the control plots. Harvest index was correlated with growth traits in the inoculated plots but not in the control plots.

The high correlation of harvest index (H.I.) with grain yield in these studies and the known association of hill plot (H.I.) with row plot grain yield is important. This relationship suggests that harvest index could be used with hill plots to help identify high yielding wheats with disease resistance.

Biological yield and harvest yield were the main determinants of yield in these experiments but had low heritabilities.

DEDICATION

Dedicated to God who insists man explore  
Him and His universe and to his Prophet -  
Muhammad (Peace Be Unto Him) who suffered  
so much from man teaching him his well being  
and  
to those pursuing natural truth.

ASSOCIATION OF STRIPE RUST (PUCCINIA STRIIFORMIS WEST.) DISEASE INDEX  
WITH GRAIN YIELD, GROWTH RATE, HARVEST INDEX AND OTHER  
AGRONOMIC CHARACTERISTICS OF WHEAT (TRITICUM AESTIVUM L.)

by

MUHAMMAD AQIL KHAN

A thesis submitted in partial fulfillment  
of the requirements for the degree

of

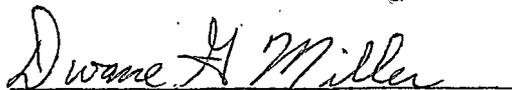
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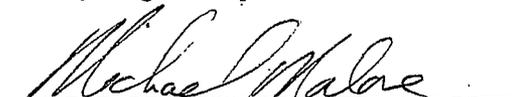
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Approved:

  
Chairperson, Graduate Committee

  
Head, Major Department

  
Graduate Dean

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## TABLE OF CONTENTS

		Page
	DEDICATION . . . . .	i
	VITA . . . . .	iii
	ACKNOWLEDGEMENTS . . . . .	iv
	TABLE OF CONTENTS. . . . .	v
	LIST OF TABLES . . . . .	vii
	LIST OF FIGURES. . . . .	x
	ABSTRACT . . . . .	xi
CHAPTER		
1	INTRODUCTION. . . . .	1
2	REVIEW OF LITERATURE. . . . .	3
	The Disease and the Pathogen. . . . .	3
	Effects of Stripe Rust on Wheat Plant . . . . .	5
	Concurrent Selection for Yield and Disease Resistance Under Disease Conditions . . . . .	10
3	MATERIALS AND METHODS . . . . .	12
	Experimental Materials. . . . .	12
	Field Design and Layout . . . . .	14
	Disease Control and Inoculation . . . . .	16
	Data Sampling and Analysis. . . . .	17
4	RESULTS AND DISCUSSION. . . . .	23
	Section 1: Spring Wheat Experiment . . . . .	23
	Variability and Disease Influence . . . . .	23
	Performance of Genotypes in Control and Inoculated Experiments and Their Comparisons. . . . .	30
	Association of Agronomic, Growth and Disease Traits. . . . .	47
	Path Coefficient Analysis . . . . .	56
	Heritability Estimates. . . . .	63
	Section 2: Winter Wheat Experiment . . . . .	65
	Variability and Disease Influence . . . . .	65
	Performance of Genotypes in Control and Inoculated Experiments and Their Comparisons. . . . .	73
	Association of Agronomic, Growth and Disease Traits. . . . .	87
	Path Coefficient Analysis . . . . .	96
	Heritability Estimates. . . . .	104

CHAPTER		Page
5	SUMMARY AND CONCLUSIONS . . . . .	107
	LITERATURE CITED. . . . .	112

## LIST OF TABLES

<u>Table</u>		<u>Page</u>
1	Number of lines in various generations of winter and spring wheat experiments . . . . .	13
2	Disease reaction of parents to stripe rust . . . . .	15
3	Mean squares of agronomic and growth characters of populations of various crosses in spring wheat experiment . . . . .	25
4	Coefficients of variation of agronomic and growth characteristics in spring wheat control and inoculated experiments . . . . .	31
5	Means of parents, $F_4$ and $F_6$ lines of the spring wheat cross-Centana/Polk and their comparisons. . . . .	33
6	Means of parents, $F_4$ and $F_6$ lines of the spring wheat cross-Sheridan/Centana and their comparisons . . .	35
7	Means of parents, $F_4$ and $F_6$ lines of the spring wheat cross-Shortana/Centana and their comparisons . . .	38
8	Mean disease indices of the parents, $F_4$ and $F_6$ of the three crosses in spring wheat experiment . . . . .	41
9	Correlation coefficients of agronomic and growth characters of $F_4$ and $F_6$ generation of spring wheat cross-Centana/Polk in control and inoculated plots . . .	48
10	Correlation coefficients of agronomic and growth characters of $F_4$ and $F_6$ generations of spring wheat cross-Sheridan/Centana in control and inoculated plots .	49
11	Correlation coefficients of agronomic and growth characters of $F_4$ and $F_6$ generations of spring wheat cross-Shortana/Centana in control and inoculated plots .	50

<u>Table</u>	<u>Page</u>
12 Combined correlation analysis of populations in control and inoculated experiments of spring wheat. . .	53
13 Estimates of direct and indirect path coefficients between grain yield and its components in spring wheat control experiment. . . . .	57
14 Estimates of direct and indirect path coefficients between grain yield and its components in spring wheat inoculated experiment . . . . .	60
15 Heritability estimates of various agronomic and growth characters of $F_4$ and $F_6$ generations of the three crosses in spring wheat experiment. . . . .	64
16 Mean squares of agronomic and growth characters of populations in various crosses in winter wheat experiment. . . . .	66
17 Coefficients of variation of agronomic and growth characteristics in winter wheat control and inoculated experiments. . . . .	71
18 Means of parents, $F_3$ and $F_5$ lines of winter wheat cross-Centurk/Itana and their comparisons . . . . .	74
19 Means of parents, $F_3$ and $F_5$ lines of winter wheat cross-McCall/Itana and their comparisons. . . . .	78
20 Means of parents and $F_3$ lines of winter wheat cross-MT6928/ID103 and their comparisons. . . . .	81
21 Mean disease indices of the parents, $F_3$ and $F_5$ of the three crosses in winter wheat experiment. . . . .	84
22 Correlation coefficients of agronomic and growth characters of $F_3$ and $F_5$ generations of winter wheat cross-Centurk/Itana in control and inoculated plots. . . . .	88

<u>Table</u>	<u>Page</u>
23 Correlation coefficients of agronomic and growth characters of $F_3$ and $F_5$ generations of winter wheat cross-McCall/Itana in control and inoculated plots. . . . .	89
24 Correlation coefficients of agronomic and growth characters of $F_3$ generation of winter wheat cross-MT6928/ID103 in control and inoculated plots. . . . .	90
25 Combined correlation analysis of populations in control and inoculated experiments of winter wheat. .	94
26 Estimates of direct and indirect path coefficients between grain yield and its components in winter wheat control experiment. . . . .	97
27 Estimates of direct and indirect path coefficients between grain yield and its components in winter wheat inoculated experiment . . . . .	101
28 Heritability estimates of various agronomic and growth characters of $F_3$ and $F_5$ generations of the three crosses in winter wheat experiment. . . . .	105

LIST OF FIGURES

FIGURE		Page
1	Pattern of relationships in path analysis and phenotypic paths. . . . .	21

## ABSTRACT

The relationship of grain yield, growth rate, harvest index and other agronomic characteristics of wheat (Triticum aestivum L.) in disease and disease free conditions was examined using three crosses each of spring and winter wheat with two generations in each, i.e., F<sub>4</sub>-F<sub>6</sub> and F<sub>3</sub>-F<sub>5</sub>, respectively. Effects of stripe rust (Puccinia striiformis West.) on plant traits and their relationships in control and inoculated hill plots were studied using ratios (inoculated/control), phenotypic correlations, multiple regression, path coefficient analyses and heritability estimates.

The wheat lines from five of the six crosses used in these studies were developed previously emphasizing only the accumulation of additive genes for stripe rust resistance. This resulted in a decline in grain yield and other agronomic traits in subsequent generations. Concurrent selection for both agronomic characteristics and disease resistance is suggested to avoid erosion of variability for important agronomic characteristics.

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## Chapter 1

### INTRODUCTION

Millions of tons of wheat are lost annually to diseases and pests throughout the world. An estimated 20 percent is lost to plant diseases, with approximately one-half in the storage and the other half in the field. A major part of this loss is attributed to rusts.

After discovery of Mendel's Laws earlier this century, the work initiated for understanding host-pathogen relationships led to breeding for disease resistance in crop plants. Genes for rust resistance in wheat were sought from near relatives, land races and other genera. However, the prolific nature of the pathogen defeated most of these efforts and the resistance in wheat varieties has lasted only 4 - 5 years in many instances.

Initially, breeding for disease resistance was aimed at producing immune varieties. Such reactions, as we now know, are determined by a few major genes which succumb to new races of rust organisms which originate soon after the new varieties are developed. The use of horizontal resistance has been advocated more recently. This resistance has a wider base due to many additive minor genes. Horizontal resistance represents a middle of the road approach where both host and pathogen coexist without seriously jeopardizing each other's existence, i.e., "live and let live."

Advocacy of such philosophy suggests an understanding of the interaction of the host with the pathogen, and, an understanding of how disease affects the expression of the characteristics of the wheat plant and their relationships. The objective of my research, therefore, included a study of the relationships of grain yield, growth rate, harvest index and other agronomic characteristics of wheat in disease and disease-free conditions.

## Chapter 2

### REVIEW OF LITERATURE

Most host-pathogen studies involve a particular natural or greenhouse disease situation. The results, therefore, relate to a kind of local situation of the host-pathogen relationship. Obviously, the pathogen affects the plants and therefore causes a deviation in the expression of their normal potential. But in the dynamic system both the pathogen and the host present an endless combination of virulence and resistance which are further compounded by the environment. In the pages that follow both the effect of disease on wheat, and the effect of resistance on disease development are reviewed.

#### The Disease and the Pathogen

Stripe rust, caused by the Basidiomycete Puccinia striiformis West. (syn. P. glumarum Ericks. and Henn.) was the disease chosen as a tool for the study reported herein. Stripe rust occurs in cool, moist climates. In the United States, stripe rust is mainly confined to the Pacific Northwest (15, 48) though its existence has been reported in other states (51). It is also a serious threat to the cereals in Northwest Europe-(29). Epidemic out-breaks of the disease have been reported in many countries (29, 45, 49, 64). The physiology, spread, epidemiology of the disease and genetics of the pathogen have been discussed by many authors (10, 26, 38, 29, 49, 50, 56).

The life cycle of the fungus is similar to the stem and leaf rusts of wheat except the sexual stage is lacking. The fungus, therefore, overwinters through asexual spores or mycelium (27, 54). High temperatures and drought discourage the growth of the pathogen (7, 27, 47). The pathogen survives these periods in cooler areas where spring planted wheat and native grasses provide the overwintering host (30, 52). In intermountain valleys of Montana, the infection is endemic and fungus overwinters as dormant mycelium in the leaves of winter wheat seedlings (54).

Climatic factors are important at each stage of the fungal growth (20, 23, 49, 53). In a water saturated environment the germination and leaf penetration of germ tubes occur in the temperature range of 2 - 15C (53) with an optimum of 7C. The germination decreases with exposure to sunshine (38) and decreases to almost nil after one day of exposure (49). The germination and growth of fungus is also dependent on the temperature at which the plants are grown (53).

While low temperatures are important for germination of the spores, once the disease is established higher temperatures may not necessarily be detrimental to the growth of the fungus as long as night temperatures are below 15C (53). The changes in the reaction type from one temperature regime to another were due to the activation of minor genes at higher temperatures (50, 55).

### Effects of Stripe Rust on Wheat Plant

References to yield losses due to stripe rust are numerous and well documented in the literature. In 1965, Zadoks (65) listed the years of stripe rust epidemics and the damages caused in 14 countries in Europe. Nagarajan and Joshi (44) similarly gave an account of rust epidemics in the Indo-Pakistan subcontinent. The situation in other countries has been reviewed by other workers (6, 9, 19, 31, 36).

The pathogen causes a progressive damage to the plant which usually is proportional to the degree and duration of infection (12). Generally the early infection causes more injury and therefore, the plants suffer more yield loss. Agrios (1) summarized the way cereal rusts, including stripe rust, affect plants. In his words:

"Rust may debilitate and kill young plants, but more often they reduce foliage, root growth, and yield by reducing the rate of photosynthesis, increasing rate of respiration, decreasing translocation of photosynthates from infected tissue and, instead, diverting materials into the infected tissue. The quantity of grains produced by rusted plants may be reduced greatly and the grains produced may be devoid of starch and may consist mainly of cellulosic materials that are of low or no nutritional value to humans."

Many workers have reported the effects of stripe rust on wheat. In greenhouse studies, Bever (5) reported that infection in early stages of growth greatly reduced plant growth, reduced root production, grain and straw yield, reduced plant height, kernel size, and number. Infection at the first leaf stage reduced root weight of the susceptible wheat variety by 87.6 percent, reduced dry matter production 55.7

percent and reduced the grain yield 65.1 percent. The quantity of water used per unit of dry matter was greatly increased. Delayed inoculation, on the contrary, had much less effect. The amount of injury was found to be related primarily to the time of inoculation with reference to the stages of development of the host plant. Infection was found to have similar though less effect on the resistant varieties even though very few rust pustules appeared on the plants. Root weight of resistant plants was reduced 24.5 percent; dry matter, 18.2 percent; and grain yield, 31.0 percent. Although every part of the plant was more or less affected, roots were most sensitive to the rust infection.

Batts (4) reported a 50 percent reduction of yield in a susceptible variety under natural infection. Under similar conditions, Nelson (46) found a reduction of 32.2 percent in a susceptible line and 14.5 percent in a line with intermediate resistance.

Allan, Vogel and Purdy (3) reported highly significant differences in yield based on infection types. In an earlier greenhouse study, the two senior authors (2) reported similar results between yield and infection types. The authors also noted the detrimental effects of the disease on test weight and tillering capacity of the plant.

McNeal and Sharp (42) studied the combined effects of severity and infection type on spring wheat under natural epidemic of stripe

rust in Montana. They found a highly negative correlation between yield and disease index (a product of severity and response symbol based on infection type). Lemhi, with high disease index (D.I. = 60) had maximum grain loss. Sunderman and Wise (61) reported significant differences caused by the rust in number of seed per head, kernel weight and the yield in susceptible Lemhi derivatives compared to resistant ones. The average yield of susceptible plants was reduced to 37.5 percent; kernel weight and number of seed per head were reduced to 19.9 percent and 17.6 percent, respectively. Susceptible plants also had lower percent protein. Similar results were obtained by Stewart and Hehn (60) in a comparison of the performance of four winter wheat varieties with disease and nondisease conditions. Susceptible varieties suffer heavy losses from infection. Protection of the crop with fungicide was found to increase yield many fold (33).

Doling and Doddson (11) explained the rust infection-yield loss relationship on a quantitative basis. They showed that loss was equal to three times the square root of the rust infection assessment when plants completed flowering. Mundy (43) showed that the amount of infection on the flag leaf at growth stage 11.1 (35) had a significant negative correlation with yield; the loss in yield was 0.022 ton/ha for each one percent increase in the level of yellow rust on the flag leaf. King (32) reported that severity of infection on the flag and second leaves at milk ripe stage was negatively correlated with grain

weight per ear and mean weight of single grain. The coefficients of regression of percentage loss on flag leaf rust severity were 0.41 and 0.39 for equations based on single grain weight and total yield per ear, respectively.

Hendrix and Fuchs (22) found that heavily infected plants had fewer leaves than the ones with lesser infection. Infection reduced the growth, number of tillers per plant and the number and size of leaves. In another study, the same authors (23) reported a reduction of 18.6 - 24.0 percent in tillers, 19.6 - 25.4 percent in straw and 18.3 - 30.8 percent in the number of kernels per head.

Disease effects are accentuated considerably under dry conditions (24). Stripe rust alone, water stress alone, and stripe rust plus water stress gave 31.6, 33.7 and 77.0 percent, respectively, less yield than uninfected plants grown with ample soil moisture. Rust infection also reduced the diameter and breakage strength of the stem (21). The reduction varied from a negligible level to more than 60 percent, depending upon the susceptibility of the variety. The reduction in the growth, especially roots, was due to the reduction in the diameter of stelar cylinders, number of the phloem cells and the mitotic index of the roots (25).

By selectively infecting the different parts of the plants at different times, Doodson, Manners and Myers (12) demonstrated that late infection affects the plants directly by reducing the number of grains

per ear, percentage fertility and weight of the grain. Early infection causes appreciable losses indirectly. Early infection is more extensive and reduces the number of tillers, number of spikelets per ear, number of flowerets per ear and grain weight. Regardless of the time of infection, rust decreased yield, the reduction being proportional to the degree of infection. The infection also caused a marked reduction in general growth and vigor, the growth of the leaves and their final length and breadth. The roots were affected more seriously than any other part of the plant with reduction in dry weight of as much as 78 percent in fully infected plants. Infection of the glumes reduced yield only half as much as the infection of the leaves. In a separate experiment (14) the same authors found that rust infection decreased  $\text{CO}_2$  uptake by 43.5 percent. Translocation of photosynthates was similarly affected. Reduced translocation was accompanied with accumulation of alcohol insoluble compounds in the infected leaves. Movement of photosynthates to the roots was reduced more noticeably and as a result the authors suggested that the primary role of rust infection was to impede translocation either by a disruptive process and/or by conversion of material into insoluble compounds (13). Livne (37) reported similar effects of stem rust infection on photosynthetic processes in wheat.

Manners (39), investigating the pattern of distribution of translocates in infected plants using  $^{14}\text{C}$ , showed that a greater proportion

of the translocates went to the shoot, and in particular, in the case of upper leaves, to the grains and smaller proportion to the roots and the tillers than was the case in healthy plants.

Stripe rust significantly reduced the rate of photosynthesis (18). Dark respiration showed an initial increase, reaching a peak at sporulation and then declined. The photochemical system, otherwise, was not affected by rust infection. Chloroplasts isolated from infected plants performed both the Hill reaction and photophosphorylation as well as the chloroplasts from the healthy plants. Continued infection, however, enhanced senescence differently from that occurring in healthy senescing leaves. Senescence was rapid and chloroplast breakdown pattern in infected leaves was different. Mares (40) and Mares and Cousen (41) reported similar results of rust infection on host tissue senescence. Cytologically, apart from the presence of collars around the neck of some haustoria, they observed little evidence of host cell injury. Cytoplasmic contents and golgi apparatus of the host increased, suggesting a stimulatory effect of the rust on the host cells.

#### Concurrent Selection for Yield and Disease Resistance Under Disease Conditions

Comeau and Barnet (8) investigated the effects of barley yellow dwarf virus (BYDV) infestation on harvest index (H.I.). The H.I.

proved to be a rather stable parameter under varied environmental ranges but was highly dependent on BYDV tolerance. They suggested the use of H.I. for estimating tolerance to BYDV and foliar diseases including rust diseases. For this purpose a "tolerance index" based on the ratio of H.I.(infected): H.I. (control) could be used (8) which would give a value independent of varietal differences and environmental variations. Simons (57), on the other hand, found a variable effect of crown rust of oats on harvest index. In some oat strains rust infection increased the harvest index while in others it decreased. He, however, states, "From the practical standpoint, these results suggest that there should be no problem in selecting for high yield combined with rust resistance in breeding programs."

Several workers investigated the predictive value of harvest index as an indirect selection criteria in control conditions. Frederickson (16) and Taylor et al. (62) found in a total of six experiments that harvest index (H.I.) in hill plots, was positively correlated with grain yield in row plots. Harvest index in hill plots, then, was a good predictor of grain yields in row plots.

This body of literature supports the previously stated research objective: to study the relationships of grain yield, growth rate, harvest index and other agronomic characteristics of wheat in disease and disease free conditions.

## Chapter 3

### MATERIALS AND METHODS

The study was conducted during the 1980-81 crop season at the Montana State University. Fort Ellis Field Research Laboratory, near Bozeman, Montana on a well drained silt loam soil. The climate was favorable for stripe rust during the early phase of crop growth with maximum temperature in the 15-24C range during May and 3/4 of June. Mild weather, coupled with plentiful precipitation, gave luxuriant crop growth and rapid spread of the pathogen, especially on fall planted winter wheat. The weather in late June changed to hot and dry which slowed the fungal growth and spread. Spring wheat, which was approximately in stage 6-7 on the Feeke's growth scale (35), had lower infection than the winter wheat plots.

#### Experimental Materials

Two experiments were planted, one with winter wheat and the other with spring wheat. A spring wheat experiment planted in the 1980 crop season was destroyed previous to harvest. The winter wheat experiment was planted on 27 September, 1980 and spring wheat on 22, 23 April, 1981. Each experiment consisted of populations derived from three separate crosses. The populations, in turn, were comprised of a variable number of entries of  $F_3$  and  $F_5$ , and,  $F_4$  and  $F_6$  generations of winter and spring wheat, respectively (Table 1). The populations

Table 1. Number of lines in various generations of spring and winter wheat experiments.

Crosses	Spring wheat	
	F <sub>4</sub>	F <sub>6</sub>
Centana/Polk	75	19
Sheridan/Centana	75	17
Shortana/Centana	75	20
	Winter wheat	
	F <sub>3</sub>	F <sub>5</sub>
Centurk/Itana	98	23
McCall/Itana	98	25
MT6928/ID103	94	-

were obtained by advancing  $F_2$  and  $F_4$  plants selected from Krupinsky's (34) minor gene lines derived from crosses involving parents listed in Table 2. A range of reaction to rust from susceptible to resistant should exist among these spring and winter wheat lines. One cross (MT6928/ID103), however, was added from another source because the seed of the winter wheat cross involving a resistant parent was not available in sufficient quantity from original Krupinsky's seed.

The spring wheat lines were advanced a generation each from  $F_3$  to  $F_4$  and  $F_5$  to  $F_6$  by planting at the University of Arizona Agricultural Research Farm, Mesa, Arizona in fall 1979. The spring wheat experiment, therefore, comprised of  $F_2$  derived  $F_4$  and  $F_4$  derived  $F_6$  generations while the winter wheat experiment had  $F_2$  derived  $F_3$  and  $F_4$  derived  $F_5$  lines.

#### Field Design and Layout

The experimental lines, together with parents, were sown in hill plots in a randomized block design in which crosses were randomized in blocks and the entries randomized within crosses. The hills were planted in a 30.5cm grid using 1.0g seed in each hill containing approximately 30 seeds (16,17). The number of  $F_3$  or  $F_4$  and  $F_5$  or  $F_6$  lines in each cross are given in Table 2. Populations, parents and filler cultivars, totaled 300 entries in the spring wheat experiment and 360 entries in the winter wheat experiment. The experiments

Table 2. Disease reaction of parents to stripe rust.\*

Cultivar	C.I.Number	Reaction type#
<u>Spring wheat</u>		
Centana	12974	3
Shortana	15233	3
Sheridan	13586	2
Polk	13773	1
<u>Winter wheat</u>		
Itana	12933	3
McCall	13842	3
Centurk	15075	2
ID103	-	1 **
MT6928	-	2 **

## Source:

\* Krupinsky (34).

\*\* Field observations.

# Scale = 0-4, 4 = susceptible.

were divided into control and inoculated treatment sections. Each was a duplicate of the other in regard to entries, number of replications, lay-out and randomization. Each treatment consisted of six replications planted adjacent to each other.

#### Disease Control and Inoculation

Control plots were maintained disease free by spray application of the fungicide, Bayleton (Triadimefon) @ 1 kg/ha. Bayleton is a systematic fungicide available as a wettable powder and provided excellent protection against rust. The fungicide was applied weekly starting 24th April and 15th May, 1981 on the winter and spring wheat plants, respectively. Later, however, the interval was increased to two weeks because the dry, hot weather slowed development of stripe rust.

Disease plots were inoculated on 15th May, 1981 with a field collection of stripe rust uredospores mixed with talc powder in 1:50 on a volume basis. The mixture was uniformly dusted on the plants late in the evening with a hand pump dust applicator. At the time of inoculation the winter wheat plants were at growth stage-6 (35), with the spring wheat plants only three weeks old. Cold and wet early spring weather facilitated rapid fungal spread and by anthesis most of the susceptible winter wheat lines were fully covered with rust. This facilitated two disease readings on winter wheat; one at the flag

leaf stage on 23, 24, and 25 June, 1981 and another at anthesis on 6, 7, and 8 July, 1981. Readings were taken on severity and infection types using a 0-4 and X scale for infection and 0-100 for severity (3, 59).

Dry hot weather in late June and early July slowed rust development. The severity of disease was much less on spring wheat resulting in one reading at the milk stage on 27, 28, and 29 July, 1981. Due to excessive drought conditions 8cm of water were applied to all treatments through sprinkler irrigation on 15 and 16 July, 1981 to facilitate infection.

#### Data Sampling and Analysis

Data were collected on plant height, heading date, number of tillers per hill, biological yield (grain + straw), harvest index and grain yield. Harvest index (H.I.) was calculated as seed weight/bundle weight, and expressed as a percentage. Growth rate (G.R.) was (bundle weight - seed weight)/days to heading. In addition, disease index was calculated on infected plots for each disease reading. Disease indices were obtained by multiplying the response symbol times severity.

Response symbols were obtained by assigning numerical values to infection type as follows: 4 = 1.0, 3 = 0.8, 2 = 0.4, and 0 = 0.0 (3). The few mesothetic infection types (X) were assigned a value of 0.6. Height was measured in cm from ground to the tip of the tallest tiller

in the hill excluding awns. Heading date in winter wheat was recorded as the number of days from January 1 until anthesis occurred in the main tiller of the hill. For spring wheat, heading was recorded from the planting date of 22 and 23 April, 1981. The number of tillers per hill was obtained by counting seed bearing tillers. Kernel weight was determined from the weight of 250 seeds selected at random from the plot yield. The count was taken using an electronic vibrator counter. The whole sample was counted to avoid bias due to large seed selectivity of the vibrating system.

For yield and biological yield determinations individual hills were cut at ground level, tagged and stacked on the raised pipes for drying. After ten days, when the straw was completely dry, the individual hill plot bundle was weighed to obtain biological yield, and threshed, using a single plant thresher to obtain grain yields. The winter wheat experiment was cut on 13, 14 August, 1981 and the spring wheat on 27 August, 1981.

The variable number of replications used for trait measurement were:

<u>Traits</u>	<u>Number of reps</u>
Number of tillers/hill	2
Plant height	3
Heading date	3
Growth rate	3
Kernel weight	3
Grain yield	6

Biological yield	6
Harvest index	6
Straw weight	6
Disease index	6

Disease effects were examined via ratios obtained by dividing the inoculated plot values by the control plot values (10). Individual crosses and ratios were analyzed as a completely randomized design. This was necessitated by the missing plots which resulted in an unequal number of entries in each replication. Although the results are slightly more conservative, the overall effect should be negligible due to the small degree of freedom for replications. Yield was analyzed as a test case according to both randomized block and completely randomized design. The results were similar for both cases.

The differences between the generations were tested using the t-test (58). Since the ratios of the inoculated/control should be one if disease had no effect, the ratios were tested against one. The data were averaged across the replications for all the traits and the means were used to calculate phenotypic correlation coefficients for each generation of each population in control and inoculated plots.

The direct and indirect effects of these traits on yield were partitioned by path analysis (63). The path coefficients were calculated using phenotypic correlation coefficients of various traits on grain yield and simultaneous solution of equations on page 22.

Figure 1 is the pattern of relationships in path analysis and phenotypic paths. The variables are (1) heading date, (2) plant height, (3) biological yield, (4) growth rate, (5) harvest index, (6) straw weight, (7) tiller number, (8) kernel weight, and (9) grain yield.

Heritability estimates were computed on a plot basis by using the general formula:

$$\text{Heritability} = V_g / V_g + V_e$$

where  $V_g$  and  $V_e$  are the genetic and error variance components respectively, for a trait obtained by equating observed mean squares to their expectation and solving for the variance components.





































































































































































































