# A COMPARATIVE STUDY OF THE SEVERITY OF THREE RACES OF LEAF RUST ON FIVE CULTIVARS OF WHEAT SEEDLINGS

By

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Thesis Approved: Thesis Advisor Dean of the Graduate College

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a tender age, between six and eighteen months, when loving care is most precious.

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#### CHAPTER I

#### INTRODUCTION

Wheat has long been known to be one of the major cereal crops of the world. Its consumption is not limited to the various ways in which man prepares it for food, since it also may serve as livestock pasture (15).

This crop had gained much popularity in the Plains areas of the United States for food and feed production. In many areas of Oklahoma and the rest of the Southern wheat-growing states, farmers plant earlier in the fall than is normally recommended for grain production specifically for a higher grazing potential.

Partly because of such concentrated and extensive cultivation it suffers damage and loss in yield from many diseases that attack it, including the disease called leaf rust of wheat caused by <u>Puccinia recondita</u> Rob ex. Desm. f. sp. <u>tritici</u> Erriks & E. Henn,. This is especially true among the winter wheat cultivars planted for grazing purposes in the southern plains because of the increased grazing season in the fall and therefore, an increased opportunity for the leaf rust pathogen.

Several studies already have been made on the effect of leaf rust on grain yield, both in quality and quantity of the grain, and more recently on its effect on production for cattle pasture.

Specific resistance has long been used in wheat cultivar development in attempts to control the disease (5). More recently, efforts have been made to find

and utilize generalized or non-specific resistance (3).

This study was designed to observe the response of certain selected wheat cultivars to attack by certain races of leaf rust in the early stages of growth, and to determine the effect of each race on root development by each cultivar.

#### CHAPTER II

#### LITERATURE REVIEW

The leaf rust fungus, <u>Puccinia recondita</u> f. sp. <u>tritici</u>, is found wherever wheat is grown over the world, but it is particularly severe in humid and sub-humid regions (5). It is the most common and most widely distributed of all rusts of wheat in the United States, and is a serious wheat pest in many other parts of the world also.

In some years, severe epidemics occur in part or in the entire central plains wheat regions of the United States. In 1938 for example, a leaf rust epidemic from Texas to Canada caused a loss estimated to be 30% or more (15).

Leaf rust is confined to the leaves and leaf sheaths. Heavy infections, therefore, greatly retard photosynthesis with the result that if the disease develops early, the entire leaves turn yellow and wither before the plants are 25 cm high (22). Marked interferance with leaf functions and a tendency to increase transpiration cause plants to mature early (15). Thus, when wheat plants are rusted in the early growth stage, the yield of a susceptible cultivar may be reduced as much as 94% (5). The effect of heavy rust infection is reflected not only in reduction of the grain yield and quality, but in straw yield and, in a deterioration of the root system (9, 11, 16, 23).

There is a pronounced correlation between the growth of the root and the

foliar portions of the plant, and the maintenance of a nutrient balance between the roots and the leaves is of great importance (19). It should not be surprising then that the effect of rust on the leaves should be closely associated with an effect on root development and function.

Johnson and Miller (13) found that inoculation of wheat seedlings with leaf rust resulted in root discoloration, a decrease in fibrous roots, and marked decrease in total root weight.

Hendrix and Lloyd (9), working with stripe rust, found that infected seedlings of wheat suffered morphological and anatomical abnormalities, such as reductions in pericycle cell number and a decrease in number of phloem cells. The total volume of roots had been reported to be markedly reduced, the severity of the reduction being dependent on the time of infection (13, 16). Early infections were more destructive than later infections.

Recently, Williams (23) reported on the discoloration and deterioration of roots of wheat plants infected with <u>Puccinia recondita</u> f. sp. <u>tritici</u>. He found considerable reduction in both weight and volume of the roots from infected plants. He also found that foliar growth was severely retarded as was the regrowth of seedlings infected with leaf rust after clipping to simulate grazing.

Using near-isogenic lines, one having a resistant and the other a susceptible response to leaf rust, he was interested in the effects of the rust on forage production. Under normal conditions in the field, he observed that forage production was virtually the same. But in severe disease conditions, he found that the susceptible isogenic line had a relatively poor forage production, compared to that of the resistant line. The development of physiologic specialization within morphological species of rust fungi is very pronounced in terms of varietal response to attack. The rusts may be monivorous (confined to a single host species or even variety) while many are plurivorous, (being capable of attacking and infecting many different hosts which may be related or unrelated)(8). For example, the leaf rust fungus <u>Puccinia</u> recondita f. sp. <u>tritici</u> embraces over two hundred physiologic races, some or many of which have probably arisen by mutation (5,7,10,12). Each of these races of leaf rust produce their effect on different specific cultivars of wheat, within the confines of the environment. If variations in temperature or light intensity occur, for example, there may be an increase or decrease in the development of the infection type for a particular host cultivar-pathogen race combination (7,11,13). Thus when the temperature is raised from  $17^{\circ}$  to  $25^{\circ}$  C, the cultivars Malakoff, Norka, and Democrat exhibit a more susceptible response to many races, while Carina, Bervit and Husser showed a more resistant response (7).

Specific resistance to leaf rust may be closely correlated with the age of the plant. Certain cultivars of wheat, when inoculated with certain races of the rust fungus, are highly susceptible in the seedling stage and moderately to highly resistant to the same race of the parasite at heading stage (11). Furthermore, the uppermost leaves of these same varieties were more resistant than the lower, older leaves. In certain cultivars, such as in Mammoth Red, the tips of the leaves were more susceptible to the rust than the younger tissue near the base and the opposite case had also been found (11).

Resistance is clearly a complex problem, and is affected by various factors, some of which are inherent in the host, and some inherent in the parasite. There-

fore, if the type and nature of resistance is to be studied for any given host--

parasite combination the environment must be maintained as constant as is possible.

#### CHAPTER III

#### MATERIALS AND METHODS

The five wheat cultivars used were:

(1) Danne C.I. 13876, having no known genes for resistance to leaf rust, was released in 1970 by the Oklahoma Agricultural Experiment Station and Crops Research Division, Agricultural Research Services, U.S. Dept. of Agriculture, from a group of wheat selections bequeathed to Oklahoma State University by the late Joseph E. Danne, a private plant breeder.

(2) Triumph 64 C.1. 13679 (TMP 64) $\frac{1}{a}$  also a selection produced and released by Joseph Danne, with no known genes for resistance to leaf rust.

(3) Wichita C.I. 11952, developed by the Kansas Agricultural Experiment Station from a cross between Early Blackbeard and Tenmarq which, again, has no known genes for resistance to leaf rust.

(4) A selection from the winter-spring cross of Bluebird/Sturdy//Gaza/Gabo/3/ Magnificient 27 made by Dr. J.A. Rupert at Davis, California, and having no known genes for resistance to leaf rust and cultures tested in 1970 at Oklahoma State.

<sup>1/</sup>C.1. numbers are assigned by the Germplasm Resources Laboratory, ARS, U.S. Dept. of Agriculture, Beltsville, Md.

(5) TAM W-101 C.1. 15324, a short-statured wheat released by the Texas Agricultural Experiment Station in cooperation with the Crops Research Division, U.S. Department of Agriculture, a selection from the cross Noria 16/3/ Nebraska 60/Medeterarean/Hope/4/Bison, which has no known genes for resistance to leaf rust in the seedling state, but has some resistance to this disease in the adult plant stage.

All of these wheat cultivars or selections are or have been grown extensively for wheat production in Oklahoma except the winter & spring wheat selection made by Rupert.

The races of <u>Puccinia recondita</u> f. sp. tritici used in these studies were UN1, UN2A and UN6B, which were isolated from field collections made in commercial wheat fields in Oklahoma. They were identified and classified on the basis of differential cultivars suggested by Basile (1) and two additional cultivars Westar C.I. 12110, and Wessel C.I. 13090. The designation "A" indicates pathogenicity on Wester, and the designation "B" indicates pathogenicity on both Westar and Wessel.

Uredospores used in the study were produced originally on the cultivar Danne. Five pots, each containing 20 seedlings, were inoculated when the seedlings were seven days old. Inoculation was accomplished by placing the pots horizontally on a greenhouse bench, moistening the seedling leaves by spraying them with a hand automizer containing tap water and a surfactant, Tween 20 (polyoxyethelene 20 sorbitanmonolaurate), at a rate of 3-4 drops per 1000 ml. of water. Uredospores from liquid nitrogen-storage vials were dusted on the moistened leaves and then the leaves were brushed together. The inoculated leaves were again sprayed with water

and surfactant and placed in a moist chamber overnight. These inoculated plants were placed in a growth chamber (Sherer-Gillet Model CEL 25-7) which was set at  $20^{+2}$  C with a 12-hr. photoperiod of 9687.8 lux light intensity at plant height.

Approximately 10 to 12 days after inoculation these plants had uredospores ready to serve as a source of inoculum for the plants of each wheat cultivar used in the test. This method was used to develop inoculum for all three races.

Seven days after inoculation of the plants which provided the inoculum source, ten Arasan (50% Thiram) treated seeds of each cultivar were planted in each of five 10 cm plastic pots filled with a soil mixture of four parts clay loam, one part sand, and part peat moss. The pots were pierced at the bottom, placed in other unpierced shorter plastic pots. Water was applied to the lower pot instead of the soil surface during the experiment. These pots were held in the growth chamber, each pot constituting one replication, for four days (one day after germination), and immediately thinned to five plants per pot, so as to assist in removal of the entire discarded seedling.

Inoculation of the cultivars to be tested was accomplished when the plants were seven days old. One pot of plants served as inoculum for five pots of each cultivar. Each was used separately. A modification of the brushing technique described by Browder (2) was used. Instead of vigorously brushing the plants to be inoculated with plants containing the inoculum, the plants to be inoculated were brushed gently only once, against the leaves of the plants to be inoculated. The inoculated plants were left in the moist chambers overnight and then transfered to the growth chamber. The pots were positioned in the growth chamber in a split pot design where races were the main plots. The chamber was again maintained at  $20^+_2$  C with a photoperiod of 12 hours of 9687.8 lux light intensity at plant level. The soil moisture was maintained at optimum, and nutrient solution of Hyponex (formulation 20-0-10) was supplied with the water every seven days.

In the same chamber, an uninoculated set of plants for each cultivar was grown for observation of differences in root growth and morphology.

Observation of the leaves of the plants started five days after inoculation, and a pustule count was started ten days after inoculation. Pustule numbers were noted for each primary leaf of each of the five plants per pot, and the number of pustules per plant per pot was recorded. Readings were taken every two days until the twentieth day after inoculation. At the end of 20 days, the plants were removed from the pots and the soil was carefully washed away in order to obtain root volume and dry weight. No attempt was made to separate the plants in one pot, rather, the entire root mass of 5 plants was measured and divided by the number of plants to obtain a mean weight per plant and root volume. Root volume was measured by water displacement using a 10 ml graduated cylinder. After the root volume was measured the roots were dried for 96 hours at 56 C and then weighed

### CHAPTER IV

#### RESULTS

In all the host-parasite combinations studied a high infection type developed and the response of most of the cultivars or selections was the same (Tables,1,11,111). At the end of 20 days Danne, Triumph 64, and the Rupert selection had about the same number of pustules regardless of which race was used. Wichita had noticeably fewer pustules, and TAM W-101 had very few pustules, again, regardless of the race used.

The progress of development of each of the three races on each cultivar or selection is presented diagramatically in Figures 1 through 5. With two exceptions the races seemed to progress similarly. The exceptions were with race UN2A on the Rupert selection (Fig.4) where a marked rise in the rate of increase of this race occurred between the 14th and 16th day after inoculation, and with race UN6B on Triumph 64 (Fig.2) where the progression of this race lagged considerably behind the other two races. Although the differences were not significant, race UN2A was the most aggressive, producing pustules faster than either UN1 and UN6B which was the least aggressive.

The volume and weight of the uninoculated control plants were similar, except that Wichita had considerably smaller root systems than the other cultivars used (Tables IV and V). Roots of inoculated plants were significantly

## TABLE I

## AVERAGE NUMBER OF PUSTULES PER PRIMARY LEAF OF 5 WHEAT CULTIVARS OR SELECTIONS MEASURED AT 6 DATES AFTER INOCULATION WITH RACE UN1 OF <u>Puccinia recondita</u> f. sp. <u>tritici</u>

Cultivar	Number of pustules/primary leaf after: - <sup>1/</sup>							
or	· · · · · · · · · · · · · · · · · · ·	· · · · · · · · · · · · · · · · · · ·	Days					
Selection	10	12	14	16	18	20		
Danne	18.0	69.0	127.5	139.6	157.2	191.9		
Triumph 64	9.2	29.0	56.0	90.7	151.2	185.7		
Wichita	7.9	15.7	37.1	74.1	108.7	139.5		
Rupert Sel.	15.4	72.6	97.0	127.7	143.2	175.2		
TAM W-101	0.0	0.0	4.9	7.0	8.4	12.9		

Each figure is the mean of the number of pustules on 5 replications of 5 leaves per replication.

## TABLE II

## AVERAGE NUMBER OF PUSTULES PER PRIMARY LEAF OF 5 WHEAT CULTIVARS OR SELECTIONS MEASURED AT 6 DATES AFTER INOCULATION WITH RACE UN2A OF <u>Puccinia recondita</u> f. sp. <u>tritici</u>

Cultivar		Numb	er of pustule	es/primary	leaf after	<u></u>
or	Contraction of the second of			Days		
Selection	10	12	14	16	18	20
Danne	13.8	74.4	123.1	151.7	173.3	195.4
Triumph 64	11.4	37.6	64.3	.95.1	159.9	196.1
Wichita	9.1	15.5	37.5	81.1	114.3	148.1
Rupert Sel.	13.4	50.7	103.3	158.5	181.9	195.0
TAM W-101	0.0	3.2	8.0	8.4	10.9	13.3

Each figure is the mean of the number of pustules on 5 replications of 5 leaves per replication.

1/

## TABLE III

## AVERAGE NUMBER OF PUSTULES PER PRIMARY LEAF OF 5 WHEAT CULTIVARS OR SELECTIONS MEASURED AT 6 DATES AFTER INOCULATION WITH RACE UN6B OF <u>Puccinia</u> recondita f. sp. <u>tritici</u>

Cultivar		Number	of pustules/	primary le	af after: –	<u>/</u>
or Selection			Days			
	10	12	14	16	18	20
Danne	12.6	60.1	112.6	128.3	150.3	178.6
Triumph 64	8.8	26.1	34.7	52.3	90.1	110.1
Wichita	6.1	13.7	26.9	49.4	85.2	114.5
Rupert Sel.	12.8	56.3	92.3	115.5	138.4	174.9
TAM W-101	0.0	3.2	5.0	8.4	10.9	13.3

 $\frac{1}{2}$  Each figure is the mean of the number of pustules on 5 replications of 5 leaves per replication.

reduced by an average of 30 to 50 percent both in volume and weight by rust infection. The effect of each race tended to be about the same except that the effect was somewhat less with UN6B than with the other two races.

## TABLE IV

## VOLUME OF ROOTS OF PLANTS OF 5 WHEAT CULTIVARS AND SELECTIONS INOCULATED WITH 3 RACES OF <u>Puccinia recondita</u> f. sp. <u>tritici</u> COMPARED WITH UNINOCULATED CONTROL PLANTS

			Volume, of r	oots in ml	•		
Cultivar		Inoculo	ated $\underline{1}$		Uninoculated		
or		Race			Race		
Selection	UN1	UN2A	UN6B	UNI	UN2A	UN6B	
Danne	2.5	2.4	2.6	3.5	3.5	3.6	
Triumph 64	2.8	2.8	2.9	3.5	4.2	4.1	
Wichita	1.7	1.5	1.6	2.5	2.3	2.5	
Rupert Sel.	1.4	1.3	1.8	3.0	2.9	3.1	
TAM W-101	2.4	2.7	2.5	3.9	3.1	4.2	

Each figure is the mean of 5 replications of 5 plants per replication. Each figure is the mean of 1 replication of 5 plants.

 $\frac{1}{2}$ 

## TABLE V

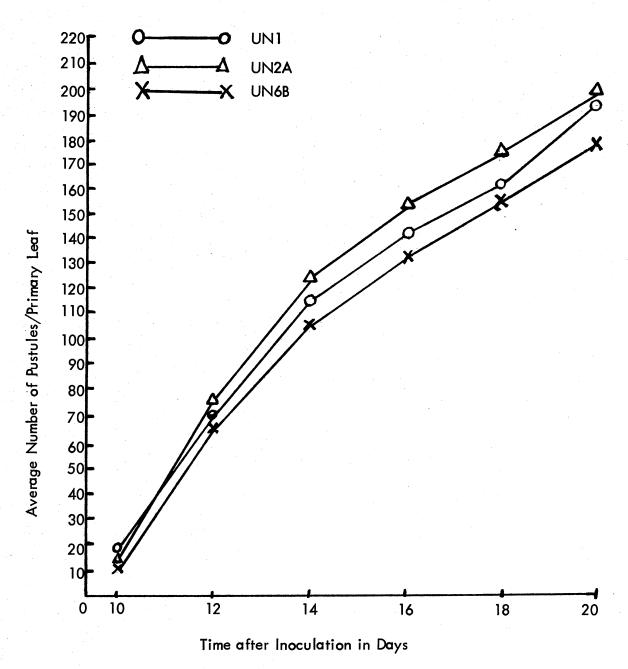
## VOLUME OF ROOTS OF PLANTS OF 5 WHEAT CULTIVARS AND SELECTIONS INOCULATED WITH 3 RACES OF <u>Puccinia recondita</u> f. sp. tritici COMPARED WITH UNINOCULATED CONTROL PLANTS

		We	ight of roots	in mg.		
Cultivar	Ir	oculated _	1	Uninoculated <u>2</u> / Race		
or		Race				
Selection	UNI	UN2A	UN6B	UN1	UN2A	UN6B
Danne	2.3	2.2	2.3	3.2	3.1	3.2
Triumph 64	2.4	2.7	2.8	3.2	3.5	3.6
Wichita	1.2	1.1	1.2	2.2	2.5	2.2
Rupert Sel.	1.2	1.1	1.5	2.9	2.9	3.1
TAM W-101	2.4	2.5	2.4	4.1	3.8	4.1

Each figure is the mean of replications of 5 plants per replication. Each figure is the mean of replications of 5 plants.

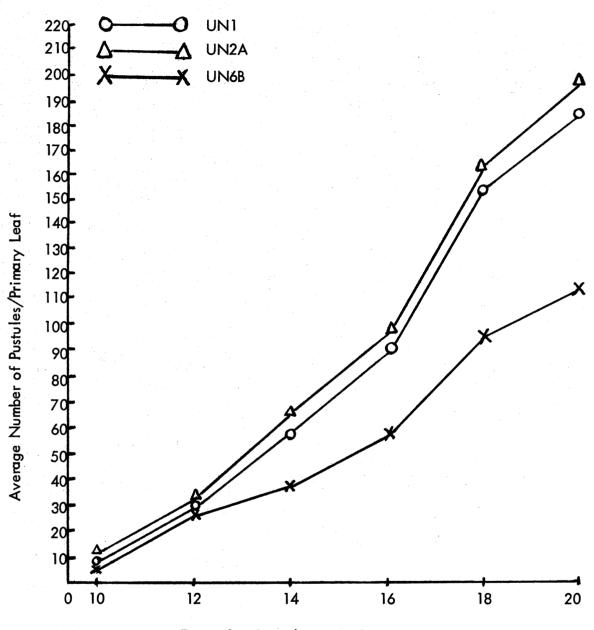
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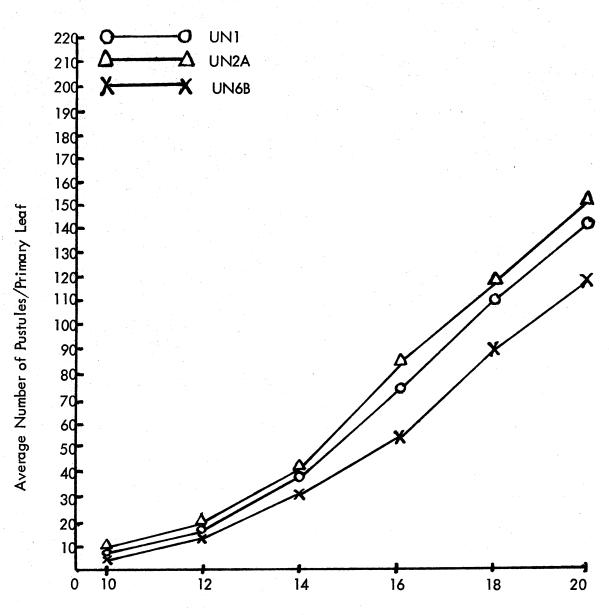
Rate of Increase in the Average Number of Pustules per Primary Leaf of the Cultivar Danne, for each of Three Races of <u>Puccinia</u> recondita f. sp. tritici







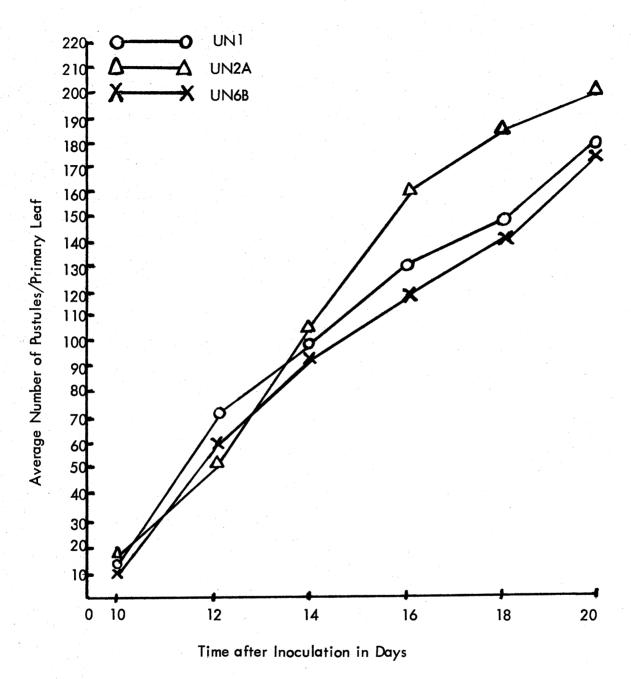
Rate of Increase in the Average Number of Pustules per Primary Leaf of the Cultivar Triumph 64, for each of Three Races of <u>Puccinia</u> recondita f. sp. tritici





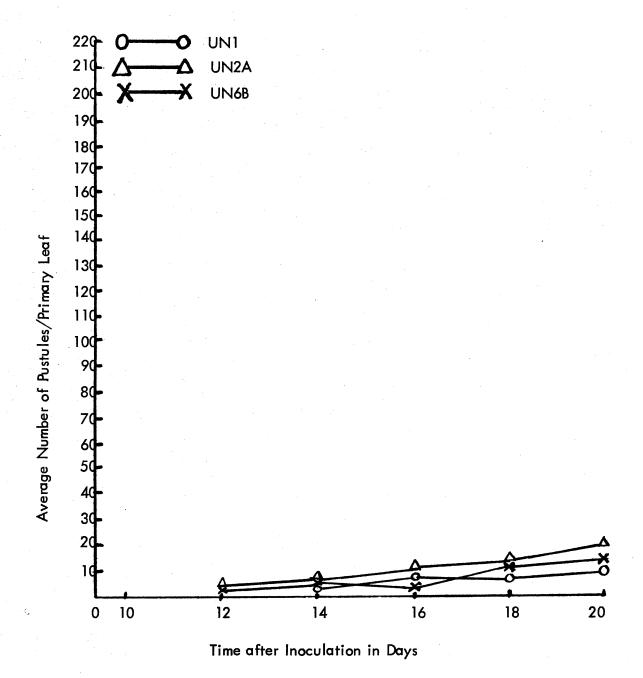


Rate of Increase in the Average Number of Pustules per Primary Leaf of the Cultivar Wichita, for each of Three Races of <u>Puccinia</u> recondita f. sp. tritici





Rate of Increase in the Average Number of Pustules per Primary Leaf of the Rupert Selection, for each of Three Races of <u>Puccinia</u> recondita f. sp. tritici





Rate of Increase in the Average Number of Pustules per Primary Leaf of the Cultivar TAM W-101, for each of Three Races of Puccinia recondita f. sp. tritici

### CHAPTER V

#### DISCUSSION

Caldwell (3) has referred to slow development of rust as a form of resistance and, therefore, wheat cultivars which have a smaller number of pustules at the early stages of development could be said to have this non-specific or generalized resistance such as the one reported here. Each of the cultivars used in this experiment were thought to be free of genes for specific resistance in the seedling stage although it was known that TAM W-101 has a gene for specific resistance which is only effective in the adult plant stage of growth. Nevertheless, the 5 cultivars used did not have the same amount of rust 20 days after infection. Danne, Triumph 64 and the Rupert selection had the most rust and were quite similar in response. This, in itself, was surprising since both Celik (4) and Smith (20) reported significantly fewer infections on Triumph 64 than on Danne when both were subjected to the same conditions. Wichita, which usually has a rust severity equal to or exceeding that of Danne, and almost always exceeding Triumph 64 in the field (24), had fewer pustules than either of those cultivars in the test. Perhaps the response of TAM W-101 was the most interesting, not only because it was the only cultivar in the test that had genes for specific resistance. Perhaps genes for specific resistance also are effective in exhibiting forms of non specific or generalized resistance.

The plants in this experiment ultimately became rather heavily infected. The effect on root growth and development was striking. The reductions in root growth of plants infected with rust over those not infected was in the range of 30 to 40 percent. This compares quite favorably with reductions reported by Williams (23) for plants having similar levels of infection.

In this study all three of the races seemed to perform similarly on all of the cultivars. Race UN6B was somewhat slower in development on the cultivars Triumph 64 and Wichita than the other races, and UN2A was somewhat more rapid in its development on the Rupert selection, but otherwise they were similar. Race UN2A was slightly more aggressive on each of the cultivars, and UN6B was the least aggressive. It is doubtful if those differences are significant in the study, but the relative aggressiveness of UN2A and UN6B compares favorably to data reported by Epperly (6) who found that uredospores of race UN2A germinated faster and at higher temperatures than uredospores on UN6B.

At the time of inoculation the primary leaves of all cultivars were approximately the same size and so they were never measured. By the end of 20 days after infection, however, there were some differences noted between cultivars. In retrospect, it may have been valuable to have the leaf size and converted the data to pustules per centimeter square. However, such a change would not have altered the major conclusions of the study.

### CHAPTER VI

#### SUMMARY

1. Five wheat cultivars Danne, TAM W-101, Triumph 64, Wichita, and a selection of winter x spring wheat cross made by J.A. Rupert, and three races of leaf rust were used to study relative rust development under controlled conditions.

2. Generally there were more pustules produced by race UN2A and UN1 than UN6B when all cultivars are considered.

3. Danne, Triumph 64 and the Rupert selection had more pustules than TAM W-101,20 days after inoculation.

4. The roots of the infected seedlings were reduced approximately 30 to 50 percent in both volume and weight, compared to the roots of healthy seedlings.

5. The root volume and weight of the roots of infected seedlings of TAM W-101 were reduced somewhat less than the other cultivars.

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