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A Study of the Environmental Conditions Influencing the Development of Stem Rust in the Absence of an Alternate Host. IV. Overwintering of Urediniospores of *Puccinia graminis tritici*. V. The Period of Initial Infection of Urediniospores of *Puccinia graminis tritici* on Wheat. VI. Influence of Light on Infection and Subsequent Development of Urediniospores of *Puccinia graminis tritici* on Wheat

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COLLEGE OF AGRICULTURE UNIVERSITY OF NEBRASKA
AGRICULTURAL EXPERIMENT STATION
RESEARCH BULLETIN 35

**A Study of the Environmental Conditions Influencing
the Development of Stem Rust in the Absence
of an Alternate Host**

- IV. OVERWINTERING OF UREDINIOSPORES OF PUCCINIA GRAMINIS TRITICI
V. THE PERIOD OF INITIAL INFECTION OF UREDINIOSPORES OF PUCCINIA
GRAMINIS TRITICI ON WHEAT
VI. INFLUENCE OF LIGHT ON INFECTION AND SUBSEQUENT DEVELOP-
MENT OF UREDINIOSPORES OF PUCCINIA GRAMINIS TRITICI ON WHEAT

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A Study of the Environmental Conditions Influencing the Development of Stem Rust in the Absence of an Alternate Host

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IV. OVERWINTERING OF UREDINIOSPORES OF PUCCINIA GRAMINIS TRITICI

INTRODUCTION

In order to ascertain whether the urediniospores of *Puccinia graminis tritici* (Pers.) Erikss. and Henn. overwinter under the environmental conditions prevailing at Lincoln, Nebraska, a series of experiments was carried out extending over a period of 5 years. The results of this investigation are briefly presented.

EXPERIMENTAL DATA

1920-21. The experiment was begun July 15 by planting Prelude wheat. Every 2 weeks thereafter a 1/40th acre planting of Turkey Red (Neb. No. 60) was made until in January.

During the latter part of August a natural epidemic of stem rust occurred on the Prelude wheat, which had made a good growth during August. Later urediniospores were plentiful on the leaves of the first 4 plantings of Turkey Red. New uredinia developed from time to time until the middle of October.

Plants showing an abundance of uredinia in the first 5 plats were located by means of stakes, October 15. Every 2 weeks thereafter, leaves with uredinia were gathered and the percentage of spore germination determined. The number of spores which germinated decreased gradually at every reading until late in January, when a small percentage of the spores were still viable. No germination was obtained from any of the spore material gathered in February, at which time it was also difficult to find leaves with uredinia.

A close examination was made of the plants in these plats during the spring, but no uredinia were found until June 14. However, uredinia of stem rust were present at the same time on wheat plants in all the plats over the farm.

1921-22. The above experiment was duplicated and similar results were obtained.

1922-23. By means of an extended period of incubation, an attempt was made to determine whether overwintering of the urediniospores might not occur and for this reason the work was confined to the low temperature greenhouse. The results obtained have already been reported.¹ They show that when seedling plants were inoculated and incubated at the optimum temperature for infection and then placed at a constant temperature of 5° C., uredinia were not visible on any of the plants within 8 weeks after they were placed at this temperature.

1923-24. Two experiments of a similar nature were undertaken in the low temperature greenhouse. As in the previous experiments the period of incubation was extended for about 8 weeks under the conditions of the experiments, at which time uredinia were visible. It is extremely doubtful if overwintering, under the fluctuating field conditions of the Nebraska winter, can occur by means of an extended period of incubation.

Further, seedling plants were grown in paraffin paper containers (Sealrights) in the open field until they began to stool. During the forepart of November they were brought into the greenhouse, inoculated and incubated for 48 hours under optimum temperature and moisture conditions. The containers with the plants were then returned to the field and placed under a variety of conditions. At the end of each week a number of containers from each set were brought into the greenhouse and maintained for a period of 2 weeks under optimum conditions for the development of the disease, when a record was made of the number of plants infected and the degree of infection.

During the course of the winter the number of plants infected decreased with each succeeding week, until the last infection consisting of one pustule on a leaf appeared in January.

1924-25. The above experiment was repeated on a more extensive scale, with similar results, namely the last pustule appeared in the greenhouse on a leaf of a plant brought in from the field January 5, 1925. These experiments check with

¹Peltier, George L. A Study of the Environmental Conditions Influencing the Development of Stem Rust in the Absence of an Alternate Host. II. Infection Studies with *Puccinia graminis tritici* Form III and Form IX. Neb. Agr. Exp. Sta. Res. Bul. 25; 52 p. 12 plates. 1923.

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the one made on the viability of the urediniospores as determined by the percentage of germination of urediniospores produced naturally on plants in the field.

SUMMARY

After the month of January, no urediniospore germination was obtained from naturally infected wheat plants in the field, and no urediniospores developed on plants inoculated and incubated in the greenhouse, placed under various conditions in the field and subsequently brought back into the greenhouse. It is extremely doubtful whether overwintering of the urediniospores of stem rust can occur under field conditions by means of an extended period of incubation. Thus, it is safe to conclude from 5 years' results that the urediniospores of *Puccinia graminis tritici* do not overwinter under conditions that exist at Lincoln, Nebraska.

V. THE PERIOD OF INITIAL INFECTION OF UREDINIOSPORES OF PUCCINIA GRAMINIS TRITICI ON WHEAT

INTRODUCTION

By the period of initial infection is meant the time required by a pathogene after it reaches a susceptible plant part to enter the tissues. To determine what the period of initial infection of urediniospores of *Puccinia graminis tritici* might be, experiments were undertaken during the season of 1923-24 using urediniospores of Physiologic Form XXI.

EXPERIMENTAL DATA

Methods. A large number of 7-day-old seedlings of Little Club (C. I. 4066) were hand inoculated and all placed in an incubation chamber at the same time. The incubation chamber was held at 23° C. and all plants had an even film of water on the leaves. At stated intervals, 60 plants were withdrawn from the chamber and placed on a bench in a greenhouse held at a temperature of about 24° C., under a series of 1000 watt Mazda C lights. The artificial light served not only to give uniform conditions for all the plants but also to dry the film of water present on the plants, when taken out of the chamber, within a few minutes after being placed under them. These lights were run continuously for the period of the experiment. A record was made of the number of plants infected and the degree of infection two weeks after incubation. The results obtained from two experiments are listed in Table 1.

Results. The first infection subsequently developed on plants incubated for 3 hours. In other words, a urediniospore was able to germinate, produce a germ tube, and enter a stoma of a leaf within 3 hours. True, the number of plants infected was small but still some pustules were produced. With 6 hours' incubation there was a decided increase in the number of plants infected, and with each additional hour of incubation the percentage of infection was almost doubled, until at the end of 24 hours' incubation, almost 90 per cent of the plants were infected. When the length of incubation was extended beyond 24 hours from 92 to 100 per cent infection of the plants was obtained.

When incubation was less than 24 hours, only a few pustules were found on the plants, even tho the percentage of infec-

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TABLE 1.—*The per cent and degrees of infection obtained on wheat seedlings with urediniospores, when incubated for various periods of time under optimum conditions for infection*

Hours of incubation	Per cent of plants infected	Degrees of infection*		Hours of incubation	Per cent of plants infected	Degrees of infection*	
		Range	Mean			Range	Mean
1	0.0	0	0	24	89.0	1-3	2-
2	0.0	0	0	26	92.0	1-3	2
3	1.7	1	1-	28	93.0	1-3	2
4	1.7	1	1-	30	98.0	1-3	2+
5	1.7	1	1-	32	93.0	1-3	3-
6	17.0	1	1-	34	98.0	1-3	3-
8	35.0	1	1-	36	100.0	1-4	3
10	27.0	1	1-	38	100.0	1-4	3
12	28.0	1	1-	40	100.0	2-4	3+
14	25.0	1	1-	42	100.0	3-4	3+
16	33.0	1-2	1-	44	100.0	3-4	3+
18	43.0	1-2	1	46	98.0	1-4	3+
20	59.0	1-2	1+	48	100.0	3-4	3+
22	78.0	1-3	2-	50	100.0	3-4	3+

* Degrees of infection —

1. Trace, 5 uredinia or less per leaf
2. Slight, 5-10 uredinia per leaf
3. Moderate, 10-25 uredinia per leaf

tion was high. Even with 24 hours' incubation, less than 10 uredinia per leaf developed. However, the longer the incubation was continued, the greater were the degrees of infection, — in other words, the greater the number of pustules produced.

Under field conditions, then, the longer the optimum conditions for infection exist the more uredinia will be produced, altho some infection can occur when these conditions prevail for only a few hours.

VI. INFLUENCE OF LIGHT ON INFECTION AND SUBSEQUENT DEVELOPMENT OF UREDINIOSPORES OF PUCCINIA GRAMINIS TRITICI ON WHEAT

INTRODUCTION

In order to determine the influence of light on initial infection and development of stem rust of wheat, a series of experiments were undertaken using the series of artificial light chambers previously described.¹ Mazda lights of 50, 100, 200, 300, 500, 750, and 1000 watts were used. These lights were on continuously thruout the experiments. The plants placed in sunlight and in the dark were grown in the same greenhouse. The experiments reported below were all run in duplicate. Seven-day-old seedlings of Little Club (C. I. No. 4066) were used and were inoculated with the urediniospores of Physiologic Form XXI.

INFLUENCE OF LIGHT ON INITIAL INFECTION

The seedling plants were hand inoculated and 20 pots of 6 plants each were placed in each light compartment in suitable incubation chambers.

The plants were incubated for 48 hours under these lights and then placed on the bench in a greenhouse held at approximately 24° C. The mean temperature within the light compartments was 20.5° with a range of 1.5° C.

Two weeks later the percentage of infected plants and the degrees of infection were recorded. The data are listed in Table 1. The results show that light is not necessary for initial infection to take place, since as large a percentage of infection was obtained in total darkness as in sunlight or under the artificial lights. Likewise little or no difference was noted in the degrees of infection. Apparently light has no influence whatever on initial infection of wheat seedling by the urediniospores of stem rust.

¹Peltier, George L., and Goss, R. W. Control Equipment for the Study of the Relation of Environment to Disease. Neb. Agr. Exp. Sta. Res. Bul. 28; 16 p. 3 plates. 1924.

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TABLE 1.—*Influence of various intensities of light on initial infection by the urediniospores on wheat*

Light	Per cent of infection on plants	Degrees of infection †		Type of infection
		Range	Mean	
<i>Watts</i>				
50	97.0	1-2	2—	Normal pustules
100	96.0	1-2	2—	Normal pustules
200	92.0	1-2	2—	Normal pustules
300	93.0	1-3	2—	Normal pustules
500	95.0	1-3	2—	Normal pustules
750	96.0	1-2	2—	Normal pustules
1000	54.0*	1-2	1+	Normal pustules
Sun	100.0	1-3	2+	Normal pustules
Dark	99.0	1-3	3—	Normal pustules

* Difficult to keep a film of water on leaves during incubation.

† Degrees of Infection—

1. Trace, 5 uredinia or less per leaf
2. Slight, 5-10 uredinia per leaf
3. Moderate, 10-25 uredinia per leaf

INFLUENCE OF LIGHT ON INFECTION AND SUBSEQUENT DEVELOPMENT OF STEM RUST

Seedling plants were hand inoculated and 15 pots of 6 plants each were placed in incubation chambers in each light compartment. After 48 hours' incubation the plants were removed from the incubation chambers. The mean temperature within the light compartments for the period of the experiment was 19.8° with a mean range of 2° C. Notes were made of the degrees and types of infection, one and two weeks after inoculation. The results together with the percentage of infected plants are given in Table 2.

A decided difference in the development of rust in the different light compartments was noted. The most rapid development occurred in the compartments having the greatest intensities of light; in fact, the development of rust was apparently more rapid under the higher intensities of light than in sunlight (101.7 hours of actual sunshine for period of experiment) altho the final results at the end of two weeks were the same. At the lower intensities of light, pustules developed but they were small in size. However, this could be correlated directly with growing conditions of the wheat plants. The results show quite well that the development of rust after infection has taken place depends on the factors favoring the development of the host plant, which in this instance was light.

TABLE 2.—*Influence of various intensities of light on infection and development of stem rust of wheat*

Light	Per cent of plants infected	After 1 week			After 2 weeks		
		Degrees of infection †		Types of infection	Degrees of infection †		Types of infection
		Range	Mean		Range	Mean	
<i>Watts</i>							
50	99.0	1-2	1	Faint flecks	1-3	1+	Pustules very small
100	92.0	1-2	1	Faint flecks	1-3	1+	Pustules very small
200	100.0	1-3	1+	Faint flecks and occasional pustule	1-3	2-	Pustules very small
300	95.0	1-3	2-	Flecks and occasional pustule	1-3	2	Pustules small
500	99.0	1-3	2-	70% flecks	1-3	2+	Pustules normal
750	91.0	1-2	1+	30% pustules	1-3	2-	Pustures normal
1000	59.0*	1	1	50% flecks	1-2	1+	Pustules normal
Sun	100.0	1-3	2	30% pustules	1-3	3-	Pustules normal
				70% pustules			
				Faint flecks			

* Difficult to keep a film of water on leaves during incubation

† See Table 1

INFLUENCE OF LIGHT ON THE DEVELOPMENT OF THE DISEASE

As in the other experiment, seedlings were hand inoculated. These plants were incubated for 48 hours under ordinary greenhouse conditions in an incubation chamber held at 22.5° C. After incubation, 19 pots of 6 plants each were placed in each light compartment. Notes on the degrees and types of infection were made at the end of one week and two weeks. These are listed in Table 3.

A uniform percentage of infection was obtained in the incubation chamber, but the development of the disease depended on the amount of light that was available. At the higher light intensities, flecking and some pustules were recorded at the end of the first week; while at the lower intensities and in sunlight (93.1 hours of actual sunshine for period of experiment), flecking was very indistinct. At the end of 2 weeks, normal pustules were observed on the plants held in the sunlight and at the higher intensities of light. The development of rust was slow and some flecks were observed on the plants held at the low intensities of light, which again was correlated with the condition of the plants. The plants grown at the higher light intensities made a rapid growth and were of a dark green color as contrasted with the slower growing somewhat etiolated and

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TABLE 3.—*Influence of various intensities of light on the development of stem rust of wheat*

Light	Per cent of plants infected	After 1 week			After 2 weeks		
		Degrees of infection †		Types of infection	Degrees of infection †		Types of infection
		Range	Mean		Range	Mean	
<i>Watts</i>							
50	100.0	1-3	1	Faint flecks	1-2	1	Flecks and small pustules <i>a</i>
100	98.0	1-2	1	Faint flecks	1-2	1+	Flecks and small pustules <i>b</i>
200	98.0	1-2	1+	Flecks	1-3	2-	Small pustules <i>c</i>
300	98.0	1-3	2-	Flecks	1-3	2	Pustules <i>d</i>
500	97.0	1-3	2	Flecks	1-3	2	Normal pustules <i>e</i>
750	100.0	1-3	2	Flecks and pustules	1-3	2+	Normal pustules
1000	100.0	1-3	2+	Flecks and pustules	2-4	3-	Normal pustules
Sun	100.0	1-3	1+	Faint Flecks	1-3	3-	Normal pustules

† See Table 1

a. Look like those on resistant varieties

b. Slightly larger than *a*

c. Stand out from leaf

d. Somewhat normal

e. Normal

chlorotic plants produced at the lower intensities of light.

From these experiments we may conclude that light is not a factor in the initial infection of wheat by urediniospores of stem rust, but that it is essential for the development of the disease, in that light is essential for the growth of the host plant and as such contributes to the development of the pathogene, perhaps thru a food relation.

