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The University of Minnesota
Agricultural Experiment Station

Sunflower Rust

By D. L. Bailey
Division of Plant Pathology and Botany

UNIVERSITY FARM, ST. PAUL

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SUNFLOWER RUST

By D. L. Bailey[1]

INTRODUCTION

Altho the sunflower has been grown for a long time in this country for ornamental purposes, it is only recently that the economic value of the plant itself has been recognized. The seed is important as a source of a highly prized edible oil and of an oil cake rich in nitrogenous matter. Preliminary tests in Nebraska, Colorado, Montana, and Michigan have established also the desirability of sunflower silage and indicate a wide range of usefulness for it. Because of this the sunflower seems destined to achieve its chief significance in this country as an ensilage crop in those northern states and in adjoining parts of Canada where corn can not be grown to advantage.

Several serious diseases of the crop, however, have appeared. One of the most destructive of these is the sunflower rust. This disease occurs commonly throughout the region in which sunflowers promise to be most extensively cultivated, and under favorable conditions it causes serious damage through defoliation. For this reason rust may prove a limiting factor in the use of sunflowers unless some satisfactory means of control can be developed.

HISTORICAL SUMMARY

Sunflower rust was first described by Schweinitz in 1882, from material collected in Pennsylvania. Subsequent to his description of it, *Puccinia helianthi* Schw. was reported from Canada, Germany, Austria, Italy, Serbia, Roumania, Sweden, and Russia.

At present, sunflower rust is distributed practically throughout the United States and occurs commonly on several species of *Helianthus*. According to Saccardo (13), it occurs on the following hosts: *Helianthus angustifolius* L., *H. annuus* L., *H. californicus* Schw., *H. decapetalus* L., *H. divaricatus* L., *H. doronicoides* Lam., *H. grosseserratus* Mart., *H. heterophyllus* Schw., *H. hirsutus* Rafin., *H. laetiflorus* Pers., *H. maximiliani* Schrad., *H. mollis* Lam., *H. occidentalis* Ridd., *H. rigidus* Desf., *H. strumosus* L., and *H. tuberosus* L.

Altho *Puccinia helianthi* was described in 1882, comparatively little work has been done on it. Kellerman first demonstrated that the rust as it occurs in this country is euautoecious; and Kellerman, Carleton, and Arthur subsequently investigated the identity of the rust on various wild hosts. Their results are rather conflicting and inconclusive, but

[1] Formerly a member of the staff.

seem to indicate that there is some biologic specialization within the species *P. helianthi* Schw., and that cultivated *H. annuus* is a common host for all forms of rust. Nothing further has been reported on the life history of the fungus, on its relation to the host, or on control measures; yet these have assumed great significance since the rust has become important economically.

OBJECTS OF THE INVESTIGATION

The investigation was undertaken to determine (1) normal life-history of *Puccinia helianthi*, (2) variations from the normal life history, (3) conditions under which infection takes place, (4) influence of external factors on development of the rust, (5) relation between the rusts on wild and on cultivated sunflowers, and (6) possible means of control.

LIFE HISTORY STUDIES

CAUSAL ORGANISM

Sunflower rust was first described by Schweinitz from material collected on *Helianthus mollis* Salem, in Pennsylvania, and was named by him *Aecidium helianthi-mollis*. Therefore the causal organism of sunflower rust is technically correctly designated as *Puccinia helianthi-mollis* (Schw.) Arthur and Bisby (4). The cumbersomeness of the trinomial probably explains why the *mollis* has been dropped, and the pathogene is commonly referred to as *Puccinia helianthi* Schw.

The telia may be scattered or gregarious and frequently are confluent. They are usually oval, two to three millimeters in diameter, pulvinate, compact, and brownish-black in color. The teliospores are smooth, chestnut brown in color, oblong-elliptical or pear-shaped, and slightly constricted at the septum. The apex is thickened (6-9 microns) and is usually round, altho occasionally somewhat flattened. The upper cell is similar in color to the lower or a shade darker, and is slightly larger. The lower cell usually tapers slightly toward a rounded base. The spores vary considerably in size. The range, as indicated by measuring a hundred spores, was 30 to 46 by 18 to 26 microns and the average was 39.1 by 23.9 microns. The pedicels are hyaline, persistent, and generally much longer than the spore, at times reaching a length of 110 microns.

The pycnia occur in small clusters, which are usually isolated but frequently coalesce. They are honey-colored at first, later becoming orange. The pycnospores are small, oval, hyaline, and appear shiny and viscous in mass.

The aecia are arranged on orbicular spots which frequently coalesce into broadly-expanded oblong lesions. The isolated clusters are from

three to five millimeters in diameter but the expanded spots are often a centimeter and a half in length. The aecia are orange-red in color with white laciniate margins. Ordinarily they appear from ten to fourteen days after the pycnial pustules develop, and while most common on the under surface of the leaves, are also to be found on the upper surface, on the petioles, and on young stems. The aeciospores are orange-red to pale orange in color and somewhat variable in shape, typically ellipsoidal but sometimes almost polygonal. They are finely echinulate and have four median germ pores. They vary from 21 to 28 microns by 18 to 20 microns.

The uredinia are chestnut brown in color, round, scattered or confluent, and pulverulent. They present an entirely different appearance on the upper and lower surfaces. (Plate III.) On the upper surface they are small and usually located on pale green or yellowish spots, while on the lower surface the sori are much more massive, darker in color, and there is little associated chlorosis. The uredinia occur on leaves, petioles, young stems, and involucral bracts. The urediniospores are sub-globose, elliptic, or obovate, and yellowish brown in color. They are echinulate and have four median germ pores. The measurements of a hundred spores varied from 23 to 27 by 17 to 22 microns and averaged 23.7 by 20.8 microns.

TYPICAL LIFE HISTORY

Sunflower rust is a typical euautoecious rust; that is, telial, pycnial, aecial, and uredinial stages are all produced on the one host. All four stages have been produced and studied in the greenhouse.

Under greenhouse conditions telia follow uredinia at varying lengths of time, apparently depending largely on the condition of the host. Under optimum conditions for the host, telia begin to form about a month and a half after the uredinial stage first appears. At the end of two months both stages are present in about equal proportions, and by the end of the third month the telial stage has almost entirely replaced the uredinial stage. This course is much varied, however, by external conditions; in general, conditions unfavorable to the host hasten the production of telia.

Thus, by cutting down the light intensity telia began to form within two weeks after the uredinia were produced and had entirely replaced the latter at the end of a month. This was the normal course of events under greenhouse conditions during December and January and was induced later by allowing only weak diffuse light to reach the plants. Similarly high temperatures (75 to 85° F.) with low soil moisture induced the formation of telia within the same period. At temperatures averaging approximately 55° F. uredinia developed very slowly and were replaced by telia within two weeks.

This ready formation of telia is very important in fortifying the fungus against adverse conditions, for the onset of these quickly induces the production of a resistant spore form which tides the fungus over until conditions become suitable for growth. Then the teliospores germinate, producing sporidia which cause infection and give rise to the pycnial stage.

Pycnia appear from eight to ten days after inoculation with teliospores and after about the same period are followed by the aecia.

Altho Woronin (17) reported in 1872 that he had been successful in obtaining the aecia of sunflower rust on *Helianthus annuus* from teliosporic infection, his observation was not confirmed until 1900, and during this period the rust was regarded by most authorities as a hemi-form. Sydow (14), in his first description of the rust, questions the accuracy of Woronin's observations and ventures the opinion that the aecia reported on various species of Helianthus in North America belong either to a heteroecious form or to an isolated *Aecidium*. However, Kellerman in 1900 (10) and Jacky (14) and Arthur (1) in 1903 demonstrated conclusively that the rust is euautoecious. Sydow, in 1902 (14), in correcting his earlier error, concludes that on account of the rarity with which the aecial stage has been reported the fungus is able to omit this stage and propagate itself altogether through the uredinial and telial generations. McAlpine (11) states that the aecia never have been reported in Australia, altho the rust is plentiful.

In this country the aecial stage, while relatively uncommon, is by no means rare and it may play a significant rôle in the life-history of the rust. In fact, except for short-cycling, it has not been shown that the rust can propagate itself here without the aecial stage; and aecia have commonly been found in Saskatchewan as late as July 28 on cultivated sunflowers. This would indicate that the aecial stage might be an important factor in the early stages of the epidemiology of the rust.

Uredinia are formed within from five to seven days after inoculation with either aeciospores or unrediniospores. This is the repeating stage and, under natural conditions, infections continue to take place until unfavorable conditions of moisture or temperature prevent further infection and induce the formation of telia.

VARIATIONS FROM TYPICAL LIFE HISTORY

The normal development of pycnia and aecia does not always follow teliosporic infection. There is a distinct tendency for the rust to omit the aecia and develop uredinia after the production of pycnia. The uredinia produced by short-cycling subsequent to pycnial formation usually had a very distinct appearance. They developed below pycnia

and were confined to definite, light-colored, slightly hypertrophied spots. The length of the incubation period also served to distinguish them from ordinary uredinia which usually developed within from five to seven days after inoculation. Short-cycled uredinia, on the other hand, do not develop until two or three days after the pycnia and then are limited to the region directly beneath them.

Nothing is known of the nuclear phenomena accompanying this shortening of the life cycle. This phase proved extremely difficult to investigate, since one had to wait for the development of the uredinium to determine whether short-cycling had occurred in any given instance. By that time the sorus was underlaid by a hopelessly intertwined mass of hyphae which were for the most part empty. Consequently, the nuclear condition could not be determined, nor could any fusion of hyphae be traced.

Short-cycling seems to occur rather infrequently and the conditions governing it are not at all understood. It was observed only on rather heavily infected leaves which suggested that a nutrient relation might be involved. Attempts to induce short-cycling by adverse host conditions did not yield conclusive results. The phenomenon occurred so erratically that it was difficult to ascertain definitely the cause. It occurred on plants incubated at approximately 60° F. as well as on plants kept dry at 75° F. but it did not do so consistently. Moreover, sometimes there was fully as much short-cycling under conditions which were apparently entirely favorable to the host.

Efforts to cause the omission of the uredinial stage were not successful. Subjecting the host to low and high temperatures, diminished light, and lack of moisture resulted in the early formation of telia as already mentioned but did not completely inhibit the formation of the uredinial stage.

Carleton (8) apparently suspected that short-cycling occurred in *P. helianthi*. In discussing the comparatively infrequent occurrence of aecia, he says: "The aecidium occurs rarely in comparison with the occurrence of other stages, but is to be found on a number of hosts and occasionally in considerable abundance. This rarity of its occurrence, together with the occurrence of spermagonia so often with the uredo, may be accounted for by the fact that the uredo is often produced by direct teleutosporic infection." He gives no other evidence for his statement and no other reference in the literature consulted has referred to the phenomenon at all.

In our experience, altho short-cycling was not frequent, it occurred often enough and was so clear that no doubt remains that it actually does take place.

PHENOMENA OF INFECTION

METHODS OF INOCULATION

In artificial inoculations the method of application of spores does not seem to be an important factor in the development of the rust. Uredinial infection is equally successful whether the spores are spread on moistened leaves or applied in suspension. Heavy infection also resulted from spreading dry spores on dry leaves and incubating in a moist chamber for forty-eight hours. In this case, however, considerable moisture condensed on the leaves before the end of the incubation period.

The usual method of inoculating with teliospores was to smear the spores on moistened leaves and then to incubate for forty-eight hours in a moist chamber. After spraying spore suspensions on the plants with an atomizer and then incubating as above, satisfactory, though usually lighter, infection developed. The suspension of well-soaked telial material above the plants in moist chambers for three days also proved a good method of inoculation.

Heavy infection resulted when aeciospores were applied either in suspension or directly on moistened leaves and the plants incubated for forty-eight hours.

Dissemination of the various spore forms in nature is probably largely by wind, for sunflower plants in isolated locations are frequently rusted. The results of artificial inoculations indicate that the manner of inoculation is relatively unimportant and that the transfer of spores by any means is followed by infection when moisture conditions become suitable.

SPORE GERMINATION

Teliospores.—When floated on the surface of water, teliospores may begin to germinate within two hours. After twelve to twenty-four hours a much elongated four-celled promycelium has formed. Each cell of the promycelium develops a hyaline, globose sporidium. (Plate II, Figures 1, 2, and 3.) The sporidia germinate soon after they are produced.

Rather frequently the promycelium, instead of forming sporidia, branches and grows indefinitely. This type of germination has been observed also on the epidermis of artificially inoculated leaves.

Teliospores are not sharply limited in their temperature requirements. They germinate well at temperatures from 6 to 21° C. The optimum for germination of spores floating on distilled water was about 18° C.

Aeciospores.—Aeciospores germinate very rapidly. Freshly produced spores may begin to germinate within an hour; and within eight

hours practically 100 per cent usually germinate. Each spore sends out one germ tube from a median germ pore.

Urediniospores.—Urediniospores produced in the greenhouse germinated within an hour and a half. In one instance 15 per cent of the spores had germinated within two hours, and some of them had produced germ tubes 46 microns long. This is of great significance in insuring the development of rust, as short periods of favorable moisture conditions, even a light dew, would be sufficient for spore germination.

The optimum temperature for the germination of urediniospores, when floating on distilled water, is approximately 18° C. and the maximum probably slightly above 28° C. Germination tests were made in incubators running at higher temperatures, but no spores germinated except in one case. In this instance 5 per cent of the spores had germinated and the incubator registered 36° C. when the spores were removed. As this result could not be obtained again, it seemed possible that the temperature of the incubator must have dropped sufficiently to permit germination. The relative percentage of germination of urediniospores at various temperatures is shown in Table I.

TABLE I

PERCENTAGE OF GERMINATION OF UREDINIOSPORES AT VARIOUS TEMPERATURES

Trial	Temperatures				
	6-9° C.	23-26° C.	18° C.	30° C.	40° C.
	Percentage of germination				
1	4	30	40	tr	0
2	tr	16	17	tr	0
3	tr	44	41	1	0
4	0	40	68	tr	0
5	tr	38	48	0	0
6	1	36	49	0	0
7	5	18	51	0	0
8	5	4	10	1	0
9	tr	61	56	tr	0
10	3	25	36	5	0

tr (trace) = less than one per cent germination. Each percentage the average of two or four hanging-drop cultures.

A light smear of paraffin oil on the cover slip of Van Tieghem cells was found to have a marked influence in increasing the germination of uredinospores. This influence did not usually extend to the spores submerged in the center of the drop but was very pronounced around the edge. As a rule only a trace of germination occurred, even under optimum temperature conditions, when a spore suspension without an oil film was used in hanging drops. The effect of the oil smear does not seem to be explicable on a direct chemical basis, because of the marked inactivity of the saturated hydrocarbons. The physical action would seem to be largely a matter of reducing surface tension, which would increase the ease of wetting and thus regulate the oxygen supply. That an oxygen relationship may be involved is suggested by the fact that spores will not germinate even if an oil film is present if the lower part of the cell contains potassium pyrogallate solution; and if the spores are floated on water in open dishes they germinate almost equally well whether an oil film is present or not. (Table II.) Moreover, if spores are floated on the drops in Van Tieghem cells instead of being immersed in the medium, they germinate well in the absence of an oil film.

TABLE II

EFFECT OF AN OIL FILM ON UREDINIOSPORE GERMINATION

Percentage of germination in hanging drops at 18°

With an oil film	Without an oil film
53	0
59	0
32	tr
95	4
31	tr
48	tr
42	0
94	tr

tr (trace) = less than one per cent.

VITALITY OF SPORES

Teliospores.—Teliospores germinate over a long period of time. In October cultivated *Helianthus annuus* was inoculated with mixed uredinial and telial material from both wild and cultivated *H. annuus*. About forty-five plants were inoculated and along with the uredinial infection that developed, five pycnia were produced. This indicated that a small percentage of teliospores germinate without a rest period. Therefore further investigations were made as soon as freshly produced telia were obtained in the greenhouse. With the exception of one instance, where a heavy infection was obtained, such spores consistently produced a light infection, one to six pycnial pustules being

produced on each leaf. The percentage of teliospores that germinate without a rest period must therefore be very small. This was further indicated by germination tests with freshly produced teliospores, for in these tests it was only rarely that any spores germinated.

As time went on, increasingly heavy infection resulted from telial material produced and stored outdoors, so that by the middle of February from 15 to 20 per cent of the spores were germinating, and consistent heavy infections were being obtained. How long these spores will retain their vitality is not known, but since collections of the pycnial stage have been made in Saskatchewan as late as the latter part of July, germination obviously continues over a long period in nature. One lot of teliospores, produced in the field and overwintered outdoors, was placed in the ice-box in June. At that time 80 per cent of the spores germinated. Six months later most of the spores were no longer viable, altho occasionally a few telia were found all the spores of which germinated vigorously.

Telial material collected on November 16, 1920, was stored in darkness and light, outdoors and inside, as well as at the following temperatures in darkness: ice-box (6-9° C.), 18° C.; room temperature, 30° C., 40° C., and 45° C. Inoculations were made from all of these on March 5, and the spores kept at 40° and 45° were no longer viable. Those that had been stored at 18° C. produced about the same infection as spores stored outdoors, and this infection was much heavier than that from spores kept in the ice-box. This agrees with Woronin's (17) observation that the teliospores of this rust germinate equally well whether kept dry in a room or taken from leaves which had laid under the snow all winter. At this time somewhat better infection was obtained from material stored in light than from that stored in darkness.

No satisfactory explanation of the varying requirements in regard to a resting period by the teliospores was discovered. Efforts to shorten the dormant period of resting spores by the citric acid treatment, as used with success on teliospores of *Puccinia graminis tritici* by Thiel and Weiss (16), were altogether unsuccessful. Varying the strength of the citric acid from 1 to 10 per cent and the time of treatment from ten minutes to three hours, likewise gave no positive results. Acetic and hydrochloric acids were substituted for citric in similar treatments; and the spores were exposed to ether vapor for various intervals, but only negative results were obtained.

The observation pointed out above, that spores stored at room temperature produced heavier infections in March than those stored under more humid conditions in the ice-box and outdoors, seems to

indicate that drying is an important factor in determining the length of the resting period.

Aeciospores.—The combined effect of various temperatures and relative humidities on the viability of aeciospores was tested in the following experiment: Aeciospores were stored in relative humidities of 0, 20, 40, 60, 80, and 100 per cent at 6, 21, 23.5, and 33° C. as well as outdoors in darkness and light. Various relative humidities were maintained in 125-cc. wide-mouthed bottles by half filling the bottles with sulphuric acid of the concentration necessary to produce the desired relative humidity. A small paper basket attached to the inner surface of the cork suspended a sample of the aecia in the controlled atmosphere of the bottle. The aecial material used was developed on Mammoth Russian in the field by artificial inoculation early in June. The experiment was begun on June 27. Freshly-opened aecia of as nearly uniform a stage of development as possible were used. Germination tests were made after 3, 7, 10, 16, 23, and 56 days. These were performed in the following way: A small piece of leaf tissue on which there were at least two aecia, was lightly shaken over a Syracuse dish partly filled with distilled water. These were incubated at 19° C. and counts were made after eight hours. It was found that aecia could be depended on for one copious spore shower only, and consequently there was not sufficient material for more frequent tests. The results are presented in Table III and summarized in Plate I.

A study of these results indicates that aeciospores do not retain their viability for long periods of time. Thus, at the end of fifty-six days, even at low temperatures, only about five per cent of the spores were viable under optimum conditions of relative humidity. At higher temperatures no spores survived. Throughout the temperature range of from 8 to 24° C., a larger percentage of spores remained alive at the end of twenty-three days when stored in 80 per cent relative humidity than at any other humidity. At higher temperatures a lower humidity is more favorable. The results indicate that throughout an extended range of temperature, humidity is a much more important factor in relation to the viability of aeciospores than temperature. In fact, the percentage of germination at the end of twenty-three days, throughout the complete range of temperature, except at 33° C., is almost a constant function of the relative humidity. Moreover, this relationship holds in the two series stored outdoors, where the fluctuating temperatures might be expected to exert a more deleterious effect. These results are so consistent that it seems fair to conclude that humidity is the outstanding factor in determining the length of time aeciospores will retain their viability at temperatures to which they are likely to be exposed in nature.

TABLE III

VITALITY OF AECIOSPORES IN RELATION TO TEMPERATURE AND HUMIDITY

Storage temperature	Per cent relative humidity	Length of storage period in days: 3	7	10	16	23	56
		Percentage of germination					
33° C.	0	3	tr	0	0	tr	†
	20	2	tr	0	0	0	†
	40	3	6	6	tr	0	†
	60	12	10	15	3	0	†
	80	5	0	tr	0	tr	†
	100	tr	s	s	s	s	†
10-31° C. (Outside in light)	0	10	s	5	tr	tr	0
	20	9	5	2	2	0	0
	40	6	35	38	7	4	0
	60	2	90	62	14	12	0
	80	2	85	90	21	21	0
	100	8	s	s	s	s	s
10-31° C. (Outside in darkness)	0	0	4	1	tr	0	0
	20	tr	10	25	1	2	0
	40	1	15	50	7	9	0
	60	15	60	60	18	14	0
	80	5	90	95	23	16	0
	100	4	s	s	s	s	s
23.5° C.	0	tr	tr	0	0	0	†
	20	18	12	12	2	0	†
	40	15	32	56	9	8	†
	60	15	60	55	13	12	†
	80	20	70	70	12	12	†
	100	40	tr	s	s	s	†
21° C	0	0	0	tr	0	tr	†
	20	35	15	12	2	0	†
	40	30	12	35	10	7	†
	60	60	40	60	12	10	†
	80	95	45	94	18	20	†
	100	90	tr	0	s	s	†
8° C	0	tr	tr	2	tr	0	0
	20	20	16	7	3	0	0
	40	23	18	35	5	4	4
	60	35	70	60	14	15	5
	80	38	85	†	20	18	3
	100	40	28	tr	s	s	s

tr = trace (less than one per cent).
s = no spores available; aecia overgrown with saprophytes.
† = no test made.

TABLE IV

VITALITY OF UREDINIOSPORES IN RELATION TO TEMPERATURE AND HUMIDITY

Storage temperature	Per cent relative humidity	Length of storage period in days					
		7	17	24	38	47	185
		Percentage of germination*					
28° C.	95	10	0	tr	0	0	0
	80	6	9	tr	0	0	0
	60	17	18	tr	2	0	0
	40	18	14	3	1	1	0
	20	17.5	22	tr	0	0	0
	0	0	0	0	0	0	0
20-23° C.	95	5	1	tr	0	0	0
	80	14	3	3	4	tr	0
	60	21	26	19	30	5	0
	40	24	25	5	18	24	10
	20	20	12	14	15	12	tr
	0	17	0	0	0	0	†
19-21° C.	95	3	0	0	0	0	0
	80	17.6	6	2	9	1	†
	60	21	23	12	6	tr	0
	40	17.5	15	14	25	19	5
	20	19	12	11	20	18	†
	0	18	0	tr	0	0	0
19-21° C.	95	1	tr	0	tr	0	0
	80	14.6	8	tr	4	tr	tr
	60	13	6	5.5	8	3	0
	40	10	8	4	32	6	2
	20	9	6	7	12	4	1
	0	10	0	0	0	0	0
8° C.	95	8	0	tr	0	tr	0
	80	19	6	tr	1	2	0
	60	22	30	15	19	13	3
	40	15	25	19	28	21	12
	20	20	16	16	10	12	20
	0	54	12	12	8	0	0
Outdoors Mean 0° C. Range of daily means +29 to −28° C.	95	2	0	tr	0	tr	†
	80	†	2	tr	tr	0	0
	60	10	9	1	3	0	0
	40	28	12	8	13	18	†
	20	26	12	10	22	19	1
	0	11	0	0	0	0	tr

* tr = trace (less than one per cent).

† = no test made.

Urediniospores.—The relation of temperature and humidity to the viability of urediniospores was investigated by an experiment exactly similar to that described with aeciospores. The urediniospores were obtained from Mammoth Russian in the field. The results of germination tests made after 7, 17, 24, 38, 47, and 185 days of storage are presented in Table IV. The results are difficult to interpret. The wide divergence between the results obtained in the two series at from 19 to 21° C. indicates that some uncontrolled factors were operating to obscure the effect of the two under consideration. The most probable of these is variability in age of the spores at the time the experiment was begun. It is much more difficult to choose uredinia of a uniform stage of development than aecia; and the comparative uniformity of the results in the two experiments supports the idea that this difference was probably largely responsible for the extreme variability in the results of the experiment with urediniospores. It is evident, however, that urediniospores are much more resistant than aeciospores. Relative humidities of from 20 to 40 per cent are optimum for a wide range of temperature and in general, humidity seems to be less important in relation to viability than in the case of aeciospores.

The fact that urediniospores remain viable for at least six months when stored at from 8 to 23° C. and at from 20 to 40 per cent relative humidity, suggests that urediniospores may overwinter. During the first year of these investigations there was little suitable material for a study of overwintering and February 17 was the latest date on which viable spores were found. In 1921 more satisfactory material was under observation, and in the last test, which was early in March, 3 per cent of the urediniospores germinated vigorously. Field evidence supports the idea that urediniospores survive the winter, because uredinia are found early in the spring without accia being present, or the two may appear about the same time.

ENTRANCE INTO THE HOST PLANT

The time required by the fungus to enter the host was determined by the influence on infection of different periods of incubation in moist chambers. This was investigated in detail for the uredinial stage. Very heavy infection resulted when plants were incubated for twenty-four hours, and there was little difference in the degree of infection when they were incubated for a longer time provided they remained uninjured. The infection became lighter as the incubation period was shortened to less than twenty-four hours, altho some infection resulted after only six hours' incubation. This is very significant when correlated with the fact that sunflower rust flourishes in dry regions, for evidently new infections can occur when there are only light dews.

The simplest method of ascertaining how the germ tubes of the various spore forms enter the host was found to be by the direct microscopic examination of the epidermis removed from artificially inoculated cotyledons. Because of its thickness, the epidermis could readily be stripped off the leaf. It then was stained with aqueous eosin and examined under the microscope.

The germ tubes of the urediniospores enter through the stomata. The germ tube forms an appressorium above a stoma. A minute penetration tube passes through the stomatal opening and a substomatal vesicle is developed. From this, hyphae grow in all directions. This is well brought out by Figure 5 of Plate II. That the method of penetration in the older leaves is similar to that of the cotyledons is indicated by Figure 4, Plate II, which is drawn from a section of an older leaf. The substomatal vesicle and appressorium are both visible and are connected by a tube passing through the stomatal opening.

The germ tubes of the aeciospores also enter through the stomata in the same manner as do those of the urediniospores. This is shown in Figure 6 of Plate II, which is a view of the inner surface of a stoma three days after inoculation, and demonstrates stomatal penetration. Substomatal vesicles beginning to send out hyphae in all directions are shown in the figures, and at a slightly different focus the remnants of appressorium could be seen.

Altho it has not actually been observed, there is little doubt that the germ tubes of the sporidia penetrate the epidermis directly. Many infected leaves were examined but there was no evidence of stomatal penetration. In a few cases there was some evidence that the promycelium had grown out indefinitely and had penetrated the leaf directly without forming sporidia.

LENGTH OF INCUBATION PERIOD

The length of the incubation period of any stage of the rust is not sharply fixed. It is greatly influenced by external conditions shortly after infection has taken place. Under optimum conditions pycnia follow teliospore infection in from ten to twelve days; and aecia are developed within the next eight or ten days. Aeciospores produce uredinial infection normally within seven days, while plants fleck following urediniospore inoculation within five days. The influence of various factors on the incubation period of the uredinial stage was more extensively studied than that of any of the other stages.

The age of the host prolongs the incubation period but does not alter the final degree of infection. When other conditions are favorable, young leaves develop uredinia in the greenhouse five or six days

after inoculation. Ten inoculations of the mature leaves of blossoming plants indicated that they become infected readily. The incubation period varied from six to eight days.

Light has a marked effect on the rapidity of the development of rust. In two trials the incubation period in strong diffuse light was two days longer than in direct sunlight. In weak diffuse light, such as resulted from passing ordinary diffuse light of the latter part of February through three thicknesses of cheese-cloth, the incubation period was increased in three trials, ten, ten, and twelve days respectively. The development of the rust may be inhibited altogether if the light is cut down still more.

The pronounced influence of temperature on the development of rust was brought out by the following experiment. Six pots of plants were inoculated in a similar manner with the same spore suspension on November 28 and incubated for two days in a moist chamber. Four of the pots were transferred to a cold incubator where the temperature remained fairly constant between 40 and 50° F. The other two were retained in the greenhouse as checks at approximately 75° F. On the latter rust developed normally, flecks appearing after five days. At the end of a week no change had taken place in the plants in the cold incubator, but flecks appeared on one of the plants two days after it was transferred to the greenhouse. Uredinia appeared two days later. After two weeks a second plant was brought into the greenhouse and after three days flecks appeared and a normal infection developed. The last was brought in at the end of four weeks and reacted similarly. Thus, for at least twenty-eight days the mycelium remained alive but dormant in the infected leaves without giving any evidence of its presence.

A second experiment was made under more variable temperatures, ranging from 36 to 70° F. but averaging approximately 55° F. The plants did not fleck until from sixteen to eighteen days after inoculation. Shortly after this the plants in three of the four pots were killed by root rots, but those in the fourth pot lived a month longer. Only weak subepidermal uredinia developed on these plants. Within two weeks from the time the uredinia developed they were almost completely replaced by telia, but even the telia failed to rupture the epidermis.

The upper limit of the temperature at which infection can take place has not been determined; but as a few urediniospores germinated at a temperature of 30° C., high temperatures are probably not often a limiting factor, if moisture conditions are favorable.

The wide range of temperature at which infection can develop partially accounts for the occurrence of the rust under such varied climatic conditions as were indicated in the discussion of distribution.

The fact that the rust may long remain dormant if external conditions are unfavorable and that it may develop rapidly when conditions again become suitable, offers a plausible explanation of the sudden appearance of rust over large areas. For instance, a cold spell might prevent the development of infections previously initiated, but it would not inhibit spore germination or the initial stages of infection. Therefore the return of favorable conditions would be marked by the development, within a few days, of a great number of infections which would normally have been spread over the whole period.

HOST RANGE AND BIOLOGICAL SPECIALIZATION

According to our present knowledge, *Puccinia helianthi* Schw. must be regarded as confined to the genus *Helianthus* L. One report of its occurrence on *Rudbeckia hirta* L. was investigated by Sydow who reports (14) that the host probably was *H. angustifolius* L. It occurs widely throughout the genus *Helianthus* as specified above under "Hosts." *Puccinia heliopsidis* Schw., which occurs on *Heliopsis laevis* Pers. and *Heliopsis scabra* Dunal, is morphologically indistinguishable from *P. helianthi* but is retained by Sydow as a separate species, since it has not been shown to be biologically identical with *P. helianthi*.

There has been much speculation as to whether there are several biological forms of the rust. Woronin and Jacky (14) claimed that the rust from *H. annuus* L. would not transfer to *H. tuberosus* L. and on this basis, after a very limited number of infection experiments, Jacky divided the species into two. He retained the name *P. helianthi* for the form on *H. annuus* L. and called the form on *H. tuberosus* L., *P. helianthorum*. Sydow did not consider the evidence sufficient to warrant breaking up the species and does not recognize Jacky's *P. helianthorum* (14).

Carleton in 1901 (6) first made cultural experiments with the rust in this country. He showed that the species was autoecious here and was of the opinion that "there is no distinction of host forms."

Arthur (1), in 1903, reported that teliospores from *H. grosse-serratus* Mart. were successfully sown on *H. grosse-serratus* and *H. maximiliani* but failed to infect *H. strumosus*. In the following year teliospores from *H. mollis* failed to infect *H. strumosus, H. tuberosus, H. grosse-serratus, H. rigida,* and *H. maximiliani*. There was slight infection on *H. tomentosus* and heavy infection on *H. mollis* and *H. annuus*. In 1905 (3), Arthur inoculated fifteen species of *Helianthus* with teliospores from *H. mollis, H. grosse-serratus,* and *H. laetiflorus*. He summarizes his results as follows: "Each set of spores grew upon the species of host from which derived but not upon the other two species, except that spores from *H. laetiflorus* sown on *H. mollis* gave

a tardy showing of pycnia without further development. Also each set of spores grew luxuriantly on *H. annuus* and made a feeble growth on *H. tomentosus*, but on all other species failed to infect or make a feeble growth." He concludes that *P. helianthi* Schw. is "a single species having many races, for which *H. annuus* acts as a bridging host." From his results it would seem that "bridging host" implied only a common host.

Kellerman (10), in 1905, infected *H. annuus* with teliospores from *H. tuberosus* but failed to infect *H. tracheliifolius, H. mollis, H. maximiliani, H. decapetalus, H. grosse-serratus, H. orgyalis,* and *H. kellermani.* Teliospores from *H. grosse-serratus* infected *H. annuus, H. orgyalis, H. tracheliifolius, H. kellermani, H. giganteus, H. grosse-serratus* and *H. decapetalus.* This work shows that Jacky's division of the species as noted above is invalid. With reference to the existence of specialization within the species Kellerman says, "Recent inoculation work leads me to think there is but one valid species and there are no recognizable 'biologic' forms." The negative results reported above he considers uncertain, since the results of different years have not agreed in all particulars.

EXPERIMENTS ON BIOLOGICAL SPECIALIZATION

In 1921 urediniospores from *H. scaberrimus* Ell., wild *H. annuus* L., and *H. subrhomboideus* Rydb., three wild forms which occur very commonly in the West, readily infected cultivated *H. annuus.* Teliospores from wild *H. annuus* and *H. maximiliani* Schrad. and from three collections of cultivated *H. annuus* also infected cultivated sunflower heavily.

The reaction of eight horticultural and cultivated varieties of sunflower to six cultures of rust was determined. The varieties used were Double California, Chrysanthemum-Flowered, Miniature I, Miniature II, Orion, Giant Russian, Mammoth Russian, and *H. cucumerifolius.* Three cultures of rust were obtained from cultivated sunflower, one from wild *H. annuus,* one from *H. scaberrimus,* and one from *H. subrhomboideus.* Altho there were variations in the severity of infection, all varieties were susceptible to all cultures of rust and there was no indication of specialization.

In 1922 the following cultures of rust were obtained: three from *H. tuberosus* L., one from *H. maximiliani* Schrad., one from *H. hirsutus* Raf., one from Mammoth Russian, and one from a horticultural variety called Chrysanthemum sunflower. These cultures were transferred to seedlings of the following species: *H. giganteus* L., *H. scaberrimus* Ell., *H. hirsutus* Raf., *H. grosse-serratus* Mart, *H. divari-*

catus L., *H. maximiliani* Schrad. two collections of *H. tuberosus* L., and Mammoth Russian.

The results are shown in Table V and indicate that three distinct biologic forms were differentiated on the above-mentioned hosts.

TABLE V

HOST REACTION TO BIOLOGIC FORMS OF *Puccinia helianthi*

Source of rust	Hosts									Biologic Form No.
	H. giganteus	*H. tuberosus* (1)	*H. tuberosus* (2)	*H. scaberrimus*	*H. hirsutus*	*H. grosseserratus*	*H. divaricatus*	*H. maximiliani*	*H. annuus* (Mammoth Russian)	
H. tuberosus	I*	I	I	I	I	I	I	I	S+	I
H. tuberosus	S	I	I	I	I	S	I	S	R	II
H. maximiliani	S--	I	I	I	I	S	I	S	R	II
Chrysanthemum sunflower	I	I	I	I	I	I	I	I	S+	I
H. tuberosus	S	I	I	I	I	S	I	S	R	II
H. annuus (Mammoth Russian)	I	I	I	I	I	I	I	I	S+	I
H. hirsutus	S	S—	I	I	S	S—	I	S	R+	III

I = immune.
R = resistant.
S = susceptible.

It is interesting to notice that the Mammoth Russian used in this work was a differential host. Forms I and II can readily be distinguished on Mammoth Russian. Form I produces a normal heavy infection, uredinia being developed on both upper and lower leaf surfaces; and usually there is little or no chlorosis associated with the infection. Plate III, Figure 1, shows this type of infection. Form II, on the other hand, infects Mammoth Russian only weakly. Uredinia are developed only on the upper leaf surface, are small, and a sharp chlorosis, which sometimes merges into necrosis, is associated

with each pustule. This type of reaction is shown in Plate III, Figure 2. Mammoth Russian is even more resistant to Form III than to Form II, but as a vigorous development of III may easily be mistaken for a weak development of II, and vice versa, these two forms are not clearly differentiated on Mammoth Russian. An additional collection on *H. divaricatus* from Pennsylvania produced only flecks on Mammoth Russian. This indicates the existence of a fourth form, but as it did not develop any rust the culture was lost before its identity could be established.

The wild species, as will be noted in Table V, were either immune or susceptible; no intermediate expressions of resistance were observed. The fact that Form I, which is the most virulent one on Mammoth Russian, is incapable of infecting any of the wild species used is very striking, but is probably not of great significance. Since sunflowers are practically self-sterile, the specific limits in the genus are not well defined, and it is inevitable that a large number of genotypes will be included in each species. Therefore the reactions indicated for the particular collection of the species worked with can not be expected to hold for all samples of the same species. This conclusion is thoroly substantiated by the results obtained. It will be noticed that no one of the three cultures of rust obtained from *H. tuberosus* was able to infect either of the two collections of *H. tuberosus* used as hosts. Further it will be observed that Form III infects one collection of *H. tuberosus* but not the other. The specific identity of the host in this case is not open to question, since *H. tuberosus* is the best characterized and most easily distinguished species of the genus. Therefore the marked variability in resistance of different populations of the same species in this genus is established, and this fact may readily account for the apparently conflicting results of previous culture work with this rust. Because of this fact, too, it is obviously unfair to conclude that wild species of *Helianthus* have little relation to the form of rust which normally infects cultivated varieties, altho this conclusion would seem justified by a casual study of Table V.

As the wild species are not pure lines and the resistance of each species is extremely variable, the results obtained by testing the reaction of particular samples of several species to given forms of rust can not be applied generally. Therefore there seems to be little advantage in following out this line of investigation past the point of demonstrating the actual existence of biologic specialization. This has been indisputably established in this investigation. Mixtures of Forms I and II even on the same leaf, of Mammoth Russian, have been separated several times, and the forms so isolated have been cultured in

some instances for ten urediniospore generations on Mammoth Russian, and each time they have produced the reaction typical of the form concerned.

The existence of biologic specialization of *P. helianthi* is of practical significance in relation to the development of resistant varieties. Since it is not possible to isolate all the biologic forms that exist or to determine the reaction of apparently resistant varieties to them, the only practical method of investigating their resistance is to grow them in many widely-separated localities for a considerable period of time.

CONTROL

INFLUENCE OF FERTILIZERS ON RUST DEVELOPMENT

An experiment was carried out to determine whether the severity of a rust attack could be influenced to any marked degree by the use of various fertilizers. Sodium nitrate, potassium chloride, and treble superphosphate were used alone and in various combinations, and one plot was fertilized with barnyard manure. Plate 1, each block of which represents one square rod, indicates the various combinations of fertilizers used and the rate at which they were applied.

PLAN OF FERTILIZER PLOTS INDICATING FERTILIZERS USED AND RATE OF APPLICATION, IN POUNDS PER ACRE, IN 1921

N — 300 PO_4— 600	PO_4— 600	PO_4— 600 K — 600	PO_4— 600 K — 300	Check	N — 600 PO_4— 600
N — 300 PO_4— 300	PO_4— 300	K — 600 PO_4— 300	K — 300 PO_4— 300	Manure 20 t/acre	N — 600 PO_4— 300
N — 300	Check	K — 600	K — 300	N — 300 K — 300 PO_4— 300	N — 600

N = Sodium nitrate.
K = Potassium chloride.
PO_4 = treble superphosphate.

The nitrates and manure were added in two applications, one at seeding and the other about two months later. In all other cases a single application was made just before seeding. When the plants were about a foot high the plots were sprayed with a suspension of urediniospores of *Puccinia helianthi* from greenhouse cultures. This was repeated about two weeks later and these two inoculations were found sufficient to institute an infection which developed rapidly into a severe epidemic.

At the end of the growing season there were no significant differences in the severity of the rust attack in the various plots. In all

cases 100 per cent of the plants were heavily and uniformly rusted. Marked differences in yield were apparent and so yield data were obtained with the hope of finding a fertilizer or combination of fertilizers whose presence might enable the crop to escape serious damage even under the conditions of a serious epidemic.

The yield results in 1921, however, proved extremely erratic and yields could not be correlated consistently with any fertilizer. As the early part of the growing season had been abnormally dry, it seemed probable that the fertilizers had not become available in significant amounts until late in the season and hence had had little chance to influence the plants. Moreover, border effects were very pronounced and by the time three border rows were discarded, such a small area remained that the yield results could not be considered very significant.

The experiment was repeated the following year. The plots were fertilized with the same fertilizers as in the previous year, the rate being reduced by one half in all cases except with the nitrates and manure, which were applied at the same rate as before. The epidemic was induced somewhat earlier than in the previous year and overwintered telial material was scattered over the plots as primary inoculum. The aecial stage developed very vigorously and abundantly and was followed by a heavy epidemic of the uredinial stage.

Again the rust developed with uniform severity on all the plots and the only difference noted was that larger and apparently more vigorous uredinia developed on the plants fertilized with nitrate. Yield data were again obtained. Table VI presents a comparison of the yields of the two years. This table indicates that while the yields are very variable, there seems to be a rather well-marked tendency for nitrate and phosphate fertilization to be associated with high yield.

The most noticeable difference between the various plots was the rate at which the infected leaves dried up on the plants. The percentage of all the leaves developed throughout the season which remained on the plants at harvest time, was determined from counts of twenty typical plants on each plot. The results are given in Table VII.

Marked differences in the percentage of leaves retained are evident. It will be noticed that the plants fertilized with nitrates consistently retained fewer leaves than did the plants fertilized with other fertilizers. This may be correlated with rust development, as the individual pustules are typically larger and more vigorous on plants heavily fertilized with nitrates. More accurately controlled experiments will be necessary to establish the relationship.

TABLE VI

COMPARATIVE YIELDS OF FERTILIZER PLOTS FOR TWO YEARS

Fertilizer	Rate of application Pounds per acre		Yield in lbs. per sq. rod	
	1921	1922	1921	1922
N	600	600	114.5	112.5
PO_4 N	300 600	150 600	112.5	85.0
PO_4	300	150	85.0	53.5
N K PO_4	300 300 300	300 150 150	80.5	114.5
K	300	150	78.5	64.5
K	600	300	76.5	64.5
N PO_4	300 300	300 150	72.5	60.0
PO_4 K	600 300	300 150	70.0	30.5
N PO_4	600 600	600 300	64.5	61.75
K PO_4	600 300	300 150	64.5	76.5
K PO_4	600 600	300 300	64.5	72.5
N	300	300	62.0	59.5
K PO_4	300 300	150 150	60.0	70.0
Manure	40,000	40,000	61.75	78.5
N PO_4	300 600	300 300	59.5	62.0
PO_4	600	300	57.5	53.5
Check			53.5	64.5
Check			53.5	57.5

N = sodium nitrate.
K = Potassium chloride.
PO_4 = Treble superphosphate.

TABLE VII

PERCENTAGE OF LEAVES RETAINED UNTIL HARVEST ON VARIOUSLY FERTILIZED PLOTS

Fertilizer	Rate of application Pounds per acre	Percentage of leaves retained at harvest
N PO_4	600 150	21.7
N PO_4	300 300	22.3
N	300	23.0
N PO_4	600 300	23.5
N	600	24.8
PO_4 K	300 300	25.8
Manure	40,000	27.1
N K PO_4	300 150 150	27.2
N PO_4	300 150	28.5
K PO_4	300 150	28.9
PO_4	300	29.0
PO_4	150	29.1
Check		29.9
K PO_4	150 150	30.9
K	300	31.0
K	150	31.0
K PO_4	150 300	31.1
Check		33.0

N = sodium nitrate.
K = Potassium chloride.
PO_4 = Treble superphosphate.

The experiment, as it has been carried out, obviously can be expected to bring out only large differences, consequently it is unsafe to base conclusions on any but the marked differences. The fact that no fertilizer or combination of fertilizers can be relied upon to control the rust or to influence materially the severity of its attack seems established. There are also strong indications that heavy fertilization with nitrates and phosphates will result in higher yields even under the conditions of a rust epidemic. However, if fertilization with nitrates is associated with excessive defoliation when the plants are attacked by rust, it is questionable whether the loss of succulent leaf tissue, which is of great value for ensilage purposes, might not offset the advantage of increased yield.

SPRAYING AND DUSTING

Preliminary greenhouse trials gave some hope that copper fungicides might effectively control the rust. Plants could not be infected artificially when they had been sprayed previously with 4-6-50 bordeaux mixture. Consequently an attempt was made to control the rust under field conditions by spraying and by dusting. The plots used in the experiment were situated close to those used in the experiment on the influence of fertilizers on the development of rust. As a severe epidemic was induced on the fertilizer plots, the adjoining ones had every opportunity to become rusted naturally and hence were not inoculated artificially. Four square-rod plots were treated and four similarly located ones were maintained as checks. Treatments were begun as soon as rust began to develop on the plots. Plot I was sprayed weekly from July 14 to August 18 with Sherwin and Williams Company's "Fungi Bordo," seven and a half ounces to three gallons of water, which is the equivalent of 4-4-50 bordeaux. Plot II was sprayed every two weeks during the same period with the same fungicide. Plot III was dusted weekly and Plot IV every two weeks during the same period with copper carbonate dust (commercial preparation from the Corona Chemical Division, Pittsburgh Plate Glass Company).

In no case did the spraying or dusting make any appreciable difference in the amount of rust present. The weekly spraying experiment was a fair test, as there was always a satisfactory coating of fungicide on the upper leaf surface at least. At two different times, when copper carbonate dust was applied, weather conditions were unfavorable to efficient dusting, so that here the test is not so convincing. It seems clear, however, that the control of sunflower rust even on a limited scale, as in windbreaks, can not be achieved practically by the use of copper fungicides.

Thus it would seem that in the development of resistant varieties remains the only possibility of controlling the disease. Kaeurpher, a South American variety, is reported (15) to be resistant in Michigan. This appears to be the only variety of cultivated sunflower for which any resistance is claimed. If this variety is resistant to all biologic forms of the rust it might well form the starting point of an attempt to develop improved resistant varieties adapted to those regions where sunflowers are likely to be of considerable economic importance.

DISCUSSION

SIGNIFICANCE OF RESULTS OBTAINED

From the facts obtained in this investigation, it is evident that *Puccinia helianthi* has many characteristics which favor its occurrence in epidemic form.

The urediniospores are produced in great numbers and retain their viability for at least six months. These are supplemented by the teliospores, some of which are capable of germinating without a rest period and originating more urediniospores, and the rest of which germinate over a long period. Thus abundant inoculum is assured.

Temperature relations are not likely to be prohibitive, as the urediniospores and teliospores germinate through a wide range of temperatures (6 to 28° C.).

The fact that urediniospores may germinate within an hour and a half and cause infection within six hours, not only assures the development of rust under normal conditions, but indicates that epidemics can develop even in a normal season. That is, a certain loss must be expected every year and this will be greatly increased in wet years.

Host conditions are also particularly favorable to the development of epidemics. It has been shown that the rust found on four of the most commonly occurring wild varieties may transfer readily to cultivated sunflowers. Thus an abundance of spores will have been produced on wild varieties before the cultivated ones develop. In any year, therefore, only the dissemination of spores from wild varieties and suitable weather conditions for short intervals will be necessary to produce an epidemic.

Sunflower rust is well fortified against unfavorable conditions and it seems particularly well adapted to rapid development when conditions become favorable.

It has been found that the onset of unfavorable conditions is followed quickly by the formation of telia. That is, a resistant spore form, capable of remaining dormant over long periods, is readily developed and insures the perpetuation of the fungus until the return of suitable growth conditions. On the other hand, some telia can germi-

nate any time and thus start rust anew. The urediniospores were found also to retain their viability for at least six months.

Under favorable conditions the rust develops very rapidly. Short-cycling is of significance in this respect, inasmuch as it results in the early production of the repeating urediniospores, thus obviating the necessity of developing aeciospores.

Short periods of high humidity are sufficient to insure the development of the urediniospores thus formed, as they may germinate within an hour and a half and cause infection within six hours. The efficiency of the urediniospores is further increased by their ability to germinate over such a wide range of temperature (6 to 30° C.) and to infect almost any part of the host.

The striking fact that the development of the rust may be inhibited for a month after infection, by unfavorable external conditions, and then proceed normally, may largely account for the sudden appearance of the rust over large areas and may also be an important factor in the development of rust epidemics. Spores can germinate at much lower temperatures than will permit subsequent development of the rust. Infection may therefore occur just before or during periods of cold wet weather, and the rust then develops very rapidly with the return of favorable conditions. The sudden development of an epidemic may be explained thus and this explanation is probably applicable to the similar phenomenon in other rusts.

From the experiments on control it is evident that the development of resistant varieties is the only promising method of control. The fact that *P. helianthi* consists of several biologic forms must be considered in the problem of resistance.

Sunflower rust has offered a fruitful field for preliminary investigation and promises much to more extensive research. Much is still to be learned of the life history of the causal organism, its methods of overwintering, the physiology of spore germination, and development of the various stages. Conditions influencing infection and the exact nature of the injury to the host call for further study. The working out of the histology of infection offers an especially interesting field because of the phenomenon of short-cycling which occurs. The host range and biologic specialization are very imperfectly known and should be investigated because of their practical, as well as their scientific value. Finally, the economic importance of the disease warrants much more extensive investigation of methods of control.

SUMMARY

1. Sunflower rust assumed economic importance when sunflowers began to be grown for ensilage purposes. It is a serious problem in their cultivation.

2. *Puccinia helianthi* was first described by Schweinitz in 1822. It has now attained practically world-wide distribution and occurs throughout the United States.

3. *P. helianthi* occurs on at least sixteen species of *Helianthus* but has not been shown to go to other genera.

4. Sunflower rust is a typical euautoecious rust. All four stages were produced and studied under greenhouse conditions.

5. Sunflower rust has a tendency to short-cycle itself by omitting the aecial stage.

6. Altho most teliospores will not germinate without a rest period, a small percentage may germinate immediately.

7. Teliospores germinate at from 6 to 28° C. They usually germinate by a four-celled promycelium, each cell of which bears an oval, hyaline sporidium. They may also germinate by producing a branched promycelium of indefinite growth.

8. At ordinary temperatures, the viability of aeciospores decreases rapidly. After three weeks only a small percentage of spores were viable. Humidity seems to be the most important factor in determining how long aeciospores retain their viability.

9. Urediniospores may germinate within an hour and a half. Each spore sends out a germ tube from a median germ pore.

10. The optimum temperature for germination of urediniospores is about 18° C., the maximum slightly above 28° C., and the minimum below 6° C.

11. Urediniospores immersed in water germinate very poorly, if at all. They germinate best when floating on water. An oil film on the cover slip of hanging-drop cultures increases the percentage of germination.

12. Uredinial infection develops if inoculated plants are incubated in moist chambers for six hours or longer.

13. Germ tubes from the aeciospores and urediniospores enter the host through the stomata, while those from the sporidia seem to penetrate the epidermis directly.

14. Under optimum conditions pycnia follow inoculation with teliospores within ten or twelve days. Aecia follow pycnia after from eight to ten days. Uredinia usually develop in from five to seven days after inoculation.

15. The incubation period of the uredinial stage is about two days longer in mature plants than in young plants.

16. Light is essential to the development of the rust. Reduced light intensity increased the length of the incubation period from six to eight days and may prolong it indefinitely.

17. Rust will not develop at temperatures below 50° F., but, if infection has already taken place, the mycelium may remain dormant in the leaves for a month at this temperature and may develop quickly with the return of higher temperatures. At 55° F. the rust develops very slowly and the uredinial stage is soon replaced by the telial stage.

18. The uredinial mycelium is largely intercellular and binucleate. Cells are killed only where pustules are being developed.

19. Rust from four of the most commonly occurring wild varieties of sunflower (*H. scaberrimus, H. annuus, H. subrhomboideus, H. maximiliani*) readily infected cultivated sunflowers.

20. The existence of at least three, and probably of four, biologic forms of *P. helianthi* has been demonstrated.

21. Sunflower rust can not be controlled nor can the severity of its attack be modified greatly by the fertilizers tested. There is some indication that the defoliation resulting from a rust attack is more severe on plants fertilized with nitrates.

22. Spraying with bordeaux mixture and dusting with copper carbonate powder were altogether ineffective in controlling the rust in a single year's trial.

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EXPLANATION OF PLATES

Plate I. Graphic presentation of the results of an experiment on the relation of temperature and humidity to the vitality of aeciospores.

Fig. 1. Percentage of germination, plotted against storage time in days, of spores kept at 33° C. (1) spores stored at 0% relative humidity; (2) spores stored at 20% relative humidity: (3) spores stored at 40% relative humidity; (4) spores stored at 60% relative humidity; (5) spores stored at 80% relative humidity; (6) spores stored at 100% relative humidity.

Fig. 2. Storage temperature 10-31° C.; otherwise the same as 1.

Fig. 3. Storage temperature 23.5° C.; otherwise the same as 1.

Fig. 4. Storage temperature 21° C.; otherwise the same as 1.

Fig. 5. Storage temperature 8° C.; otherwise the same as 1.

Fig. 6. Percentage of germination, plotted against storage time in days, of spores kept at 80% relative humidity. Storage temperatures (1) = 33° C.; (2) = 10-31° C. (outside light); (3) = 8° C.; (4) = 8-31° C. (outside dark); (5) = 23.3° C.; (6) = 21° C.

Plate II. (1) Germinating teliospore; (2) tip of promycelium showing sporidia; (3) germinating sporidia; (4) cross-section of leaf showing penetration of uredinial germ tube; (5) inner surface of epidermis three days after inoculation with urediniospores. A substomatal vesicle giving rise to hyphae; (6) aecial penetration. View of inner surface of epidermis three days after inoculation with aeciospores. Substomatal vesicle giving rise to hyphae.

Plate III. (1) The reaction of Mammoth Russian to *Puccinia helianthi* Form 1. The lower (right leaf) and upper (left leaf) leaf surfaces are shown.

(2) The reaction of Mammoth Russian to *Puccinia helianthi* Form II. Lower (left leaf) and upper (right leaf) leaf surfaces are shown.

PLATE I

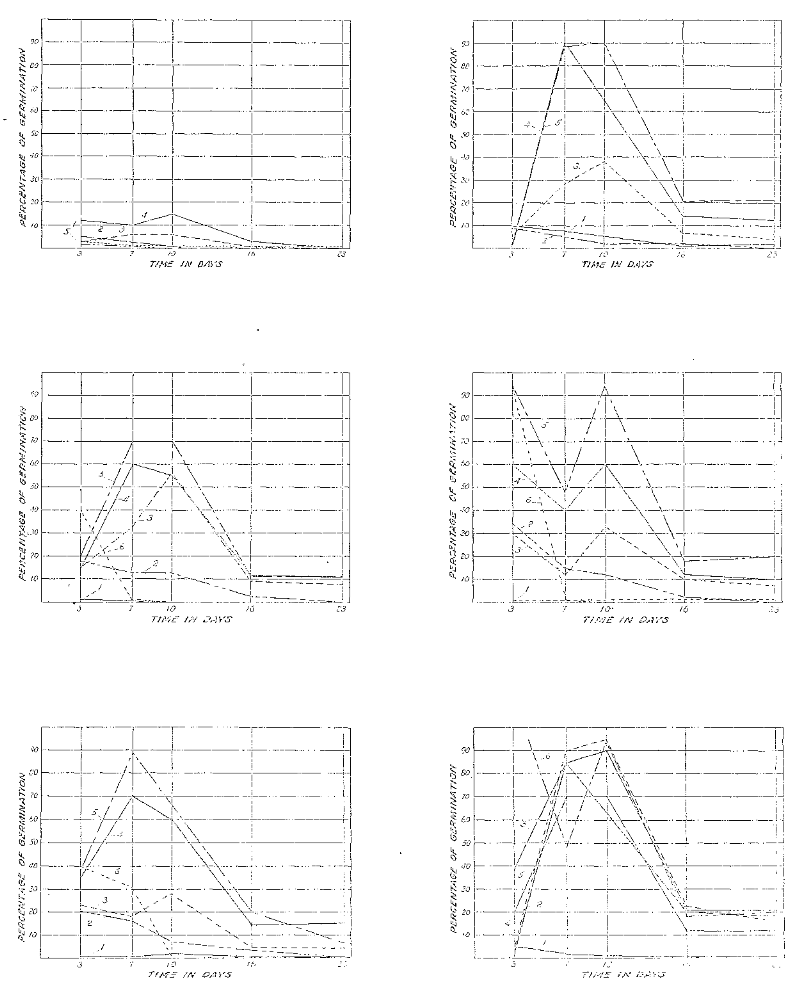

PLATE II

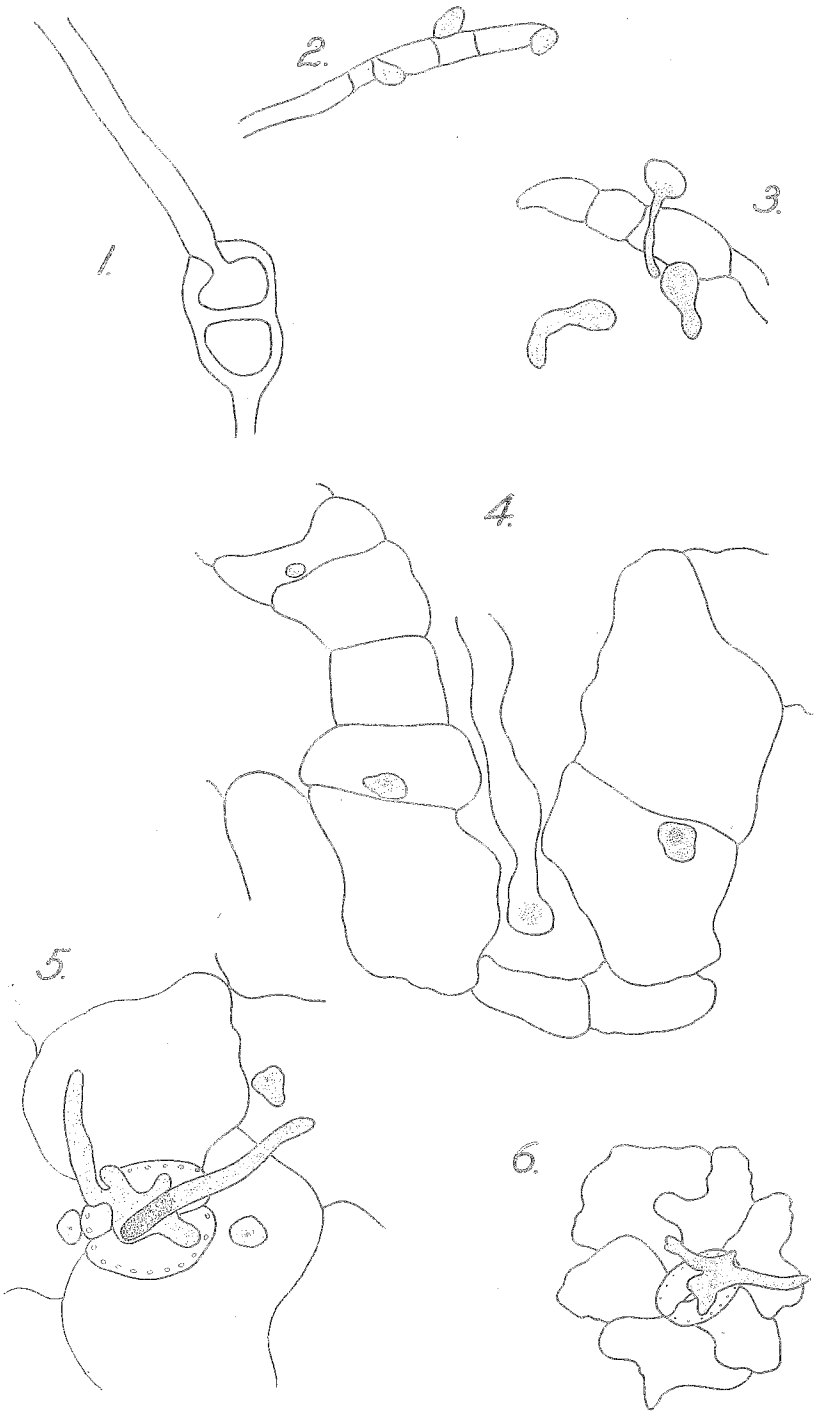

PLATE III

Figure 1

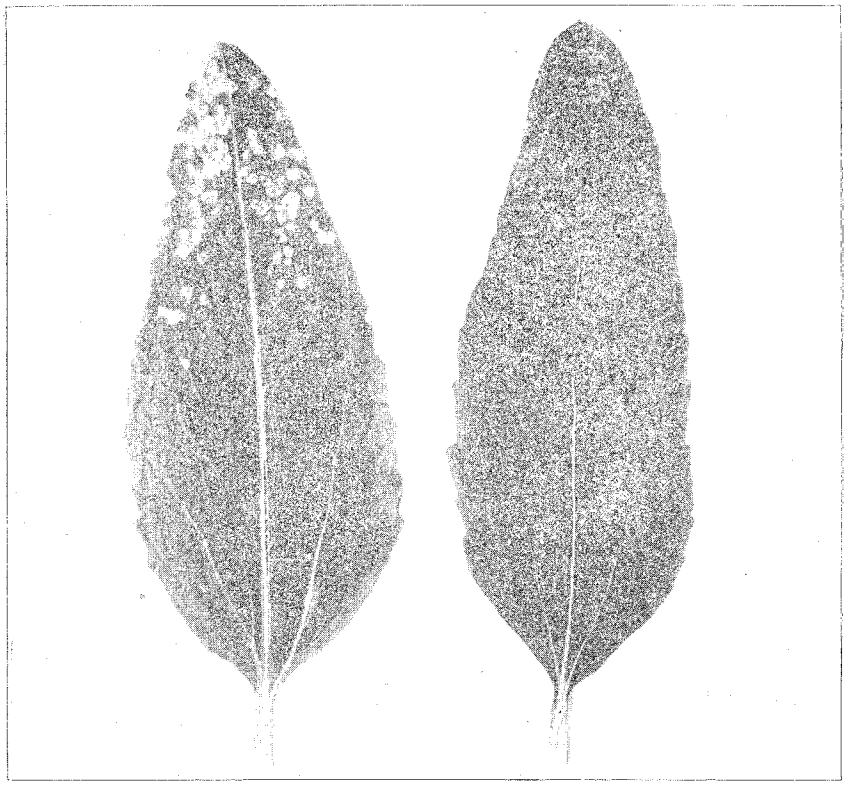

Figure 2

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