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New Facts Regarding the Period of Ascopore Discharge of the Apple Scab Fungus

BY

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*On leave of absence.

NEW FACTS REGARDING THE PERIOD OF ASCOSPORE DISCHARGE OF THE APPLE SCAB FUNGUS

By LEROY CHILDS

Investigations for the control of apple scab, which have been carried on at Hood River, Oregon, during the past three years, have demonstrated that, unlike control measures for this disease in eastern United States, an application of a fungicide has to be made earlier than the "pink" or cluster bud spray in order successfully to control the fungus. Jackson and Winston; (1) in their experimental work in 1914 at Hood River for the control of scab, began their experimental work with the pink spray. From the standpoint of scab control the results obtained were very poor. A very large percentage of the fruits were found to possess primary or ascospore infections at the time the experiments were checked at harvest time.

Investigations were continued by J. R. Winston (2) and the writer in 1915. One of the most important objects of the work was to determine the value for scab control of what is often called the "delayed-dormant" application given previous to the "pink" spray. The experimental plots were each divided into two parts. On one part the applications were begun with the delayed-dormant spray; on the other part this was omitted, and the applications were begun with the pink spray. In every experiment better control of scab was obtained where the extra-early spray was used, averaging about 12 percent more clean fruit on all plots. These results convincingly demonstrated the value of the delayed-dormant application for controlling the disease in this region.

The Investigation

Up to the present time practically no work of an investigational nature, directed toward the study of the life-history of the disease, has been undertaken in the West. For the most part, control measures preceding 1915 have been based on studies of the disease carried on in several sections of the East. The need for a spray earlier than that required in other sections, however, seemed to point out that some marked differences in the early development of the fungus existed.

In order to determine the time at which primary infection could be expected and the bearing that it would have on the first application of a fungicide, a study of the ascospore activity of the fungus was made during 1916. The investigation was productive of several interesting facts; including the correlation of spore ejection with the prevailing weather conditions, the number of periods of serious ascospore discharge, and the length of ascospore activity during the season.

In 1916 the first field observations were made by the writer on February 20. Perithecia at that time were found to be numerous on the old leaves in nearly every orchard examined. Material was brought into the laboratory at this time, individual perithecium were teased out of the leaf

(1) Jackson, H. S., and Winston, J. R. Report of the Hood River Branch Exp. Sta. 1913-14.

(2) Winston, J R., and Childs, Leroy. Report of the Hood River Branch Exp. Sta. 1914-15.

tissues, mounted on slides, and examined with the microscope. The contents were observed to be composed of undifferentiated protoplasm. Cross sections of perithecia were also made with a razor, but no cellular divisions were found in the contents of these bodies.

On March 7 another examination was made. Development had occurred to the extent that the general form of the asci were distinguishable; no differentiation or visible spore formation was observed within the asci. From material which was gathered on February 20, and kept in a moist chamber in the laboratory, developing spores were observed within the asci on this date. Spore discharge occurred from this material on March 14.

The first spores were observed "shooting" in the field on March 20. The spores were caught by placing ordinary miscroscope slides over old scab-infected leaves in much the same manner as described by Wallace (3). Instead of coating the sides with glue, however, a very thin film

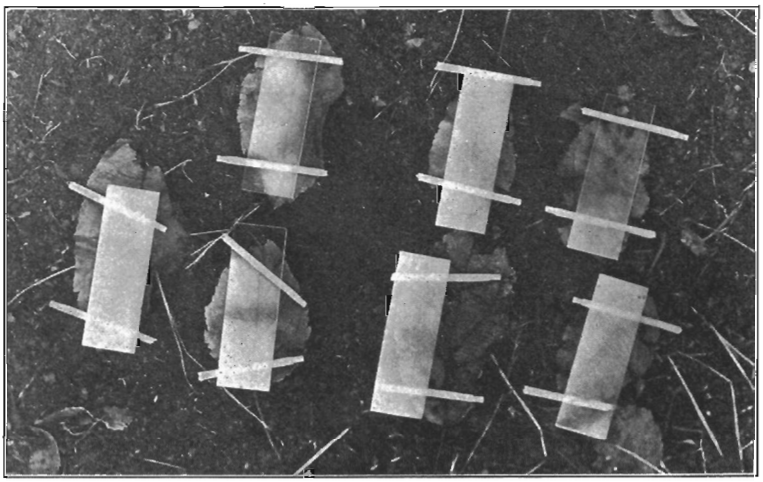

Fig. 1. Method used in catching ejected scab spores in the field.

of glycerine jelly was rubbed on the surfaces that were to be placed next to the leaves. The spores were found to stick readily to the glass itself, but with the glycerine jelly present there was much less tendency for washing away the spores by the heavy rains that occurred. At each end of the glass slide a match, split lengthwise, was placed to prevent the slides from exerting any pressure upon the leaf surface. See Figure 1. Quite often the leaves became swollen with long continued rains, at which times they came into contact with the surface of the glass. Seven leaves were used in the experiment, these same leaves being employed throughout the entire season. The scabby leaves and slides were given corresponding letters so that a complete record of activity was obtained from each leaf studied. During the first two months the slides were examined daily and replaced by clean slides; later, during periods when

(3) Wallace, Errett. Scab Disease of Apple. Cornell Univ. Bul. 335.

a great deal of fair weather occurred with no ejection, the slides were examined but every other day.

It was thought advisable to record, as accurately as was possible with the limited time for the work, the severity of daily discharge. Owing to the fact that it was impracticable to attempt the counting of ejected spores over the entire slide, the numbers were determined on twenty-five or thirty fields of the low power of the microscope.

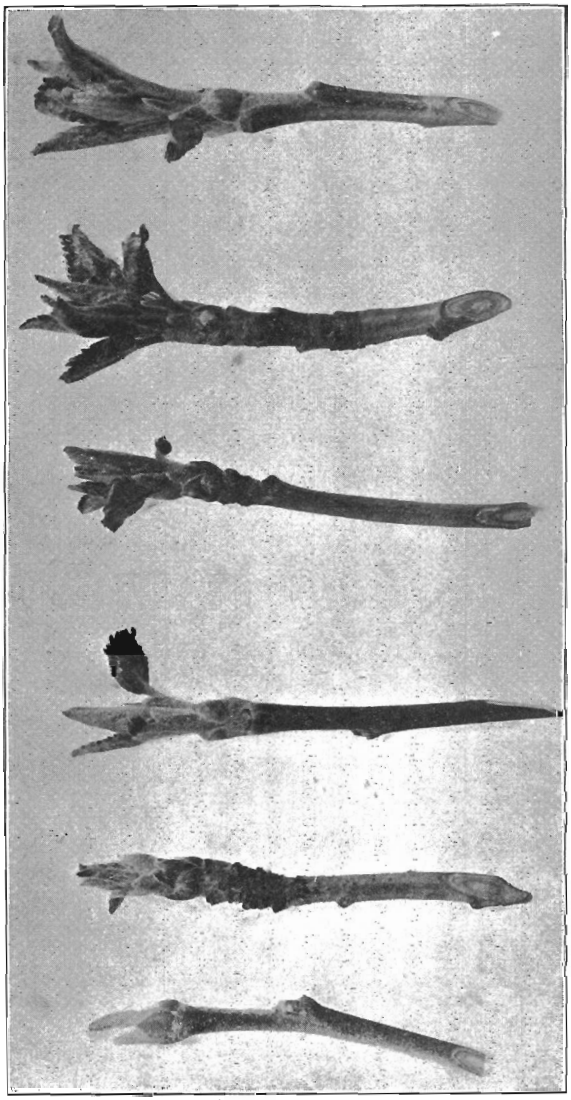

Fig. 2. Average foliage development at time of second period of heavy spore discharge in 1916. The third and fourth twigs show approximately the ideal development for the delayed-dormant spray.

Table I. Record of Ascospore Ejection of Apple Scab Fungus, 1916.

Key to Symbols used:

— No spore ejection.
(x) Not more than 10 spores found on slide.
x Average of 2 to 8 spores to low power field of microscope.
xx Average of 8 to 15 spores to low power field of microscope.
xxx Average of 50 to 70 spores to low power field of microscope.
xxxx Average of more than 70 spores to low power field of microscope.
1 Record not obtained. T. Trace of rain.

	—MARCH—													—APRIL—																				
Slide	20	21	22	23	24	25	26	27	28	29	30	31		1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21
A	xxx	1	1	1	1	xxx	xx	x	1	—	—	—		—	—	—	—	—	—	—	—	—	(x)	x	(x)	—		xx	xxx	(x)	x	(x)	(x)	—
B	1	1	1	1	1	1	1	x	xxx	—	—	—		—	—	—	—	(x)	—	—	—	—	xx	x	xx	—		xxx	xx	x	x	(x)	x	(x)
C	1	1	1	1	1	1	1	1	xx	—	—	—		—	—	—	—	—	—	—	—	—	xx	xxx	x	—		xxx	xx	x	x	xx	xx	(x)
D	1	1	1	1	1	1	1	1	xx	—	—	—		—	—	—	—	(x)	—	—	—	—	(x)	xx	x	—		xx	x	(x)	(x)	x	x	(x)
E	1	1	1	1	1	1	1	1	xx	—	—	—		—	—	—	—	(x)	—	—	—	—	x	x	xxx	—		xx	xx	xx	xx	xx	xx	x
F	1	1	1	1	1	1	1	1	xx	—	—	—		—	—	—	—	(x)	—	—	—	—	x	(x)	(x)	—		x	x	(x)	(x)	x	x	—
G	1	1	1	1	1	1	1	1	xx	—	—	—		—	—	—	—	(x)	(x)	—	—	—	—	—	(x)	—		x	(x)	(x)	x	—	(x)	(x)
Weather Conditions	Rain	Not noted	Not noted	Not noted	Not noted	Rain	Rain	Showers	Showers	Warm with frost in A. M.	Clear frost in A. M.	Part Cloudy		Clear	Clear	Clear	Clear	Showers in A. M.	Clear	Cloudy	Part cloudy	Light showers	Showers	Cloudy	Showers	Clear, warm	Rain	Showers	Showers	Rain	Rain in P. M.	Light Showers	Showers	Showers
*Precipitation	.35	1.05	.28	—	.14	1.8	.27	.04	T	—	—	—		—	—	—	—	T	—	—	—	—	T	.13	—	—	.07	T	.18	.10	.55	.05	.24	.38

*Record of precipitation kindly supplied by Mr. E. W. Birge, U. S. Cooperative Observer.

Table I. Record of Ascospore Ejection of Apple Scab Fungus, 1916—Continued.

Key to Symbols used:

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(x) Not more than 10 spores found on slide.
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xx Average of 8 to 15 spores to low power field of microscope.
xxx Average of 50 to 70 spores to low power field of microscope.
xxxx Average of more than 70 spores to low power field of microscope.
1 Record not obtained. T. Trace of rain.

	—APRIL—Continued.									—MAY—																					
Slide	22	23	24	25	26	27	28	29	30	1	2	3	4	5	6	7	8	9–10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25
A	—	—	—	—	(x)	x	x	—	—	—	—	—	—	(x)	x	x	(x)	x	x	(x)	(x)	—	—	—	—	xx	—	—	x	x	xx	x	x
B	(x)	—	—	—	—	x	xx	(x)	—	—	(x)	—	—	(x)	xx	xx	x	x	xx	x	(x)	—	—	—	—	(x)	—	—	(x)	(x)	xxx	x	x
C	(x)	—	—	—	—	xx	xx	—	—	—	—	—	—	(x)	xxx	xxx	xx	xxx	x	x	(x)	—	—	—	—	x	—	—	(x)	(x)	xx	xx	xx
D	—	—	—	—	—	x	(x)	—	—	—	(x)	—	—	—	xxxx	xx	xx	xxx	x	x	—	—	—	—	—	—	—	—	—	—	x	(x)	x
E	(x)	—	—	—	—	xxx	xxx	(x)	—	—	—	—	—	x	xxxx	xxx	xxx	xxx	xx	x	x	(x)	—	—	—	(x)	—	(x)	(x)	(x)	x	x	xx
F	(x)	—	—	—	—	(x)	xxx	—	—	—	—	—	—	(x)	xxxx	xx	xxx	xxx	xx	xx	x	—	—	—	—	(x)	—	—	xx	xx	xxx	xx	xxx
G	—	—	—	—	—	(x)	xx	—	—	—	—	—	—	(x)	x	x	x	xx	x	x	—	(x)	—	—	—	—	—	—	x	x	(x)	—	x
Weather Conditions	Clear	Clear	Part Cloudy	Part Cloudy	Rain in P. M.	Rain in P. M.	Rain	Clear	Clear	Clear	Clear	Clear	Clear	Showers	Heavy showers	Showers	Showers	Showers	Thunder showers	Showers	Clear	Clear	Clear	Clear	Clear	Rain	Clear	Cloudy	Rain	Heavy Showers	Showers	Thunder Showers	Showers
Precipitation	—	—	—	—	T	.85	.17	—	—	—	—	—	—	T	.10	.08	.23	.48	.07	T	—	—	—	—	—	.05	—	—	.38	.55	.29	.04	T

Table I. Record of Ascospore Ejection of Apple Scab Fungus, 1916—Continued.

Key to Symbols used:

— No spore ejection.
(x) Not more than 10 spores found on slide.
x Average of 2 to 8 spores to low power field of microscope.
xx Average of 8 to 15 spores to low power field of microscope.
xxx Average of 50 to 70 spores to low power field of microscope
xxxx Average of more than 70 spores to low power field of microscope.
1 Record not obtained. T. Trace of rain.

	—MAY—Continued.					—JUNE—																				—JULY—					
Slide	26	27	28	29	30	1	2	3	4	5	6–7	8	9	10	11	12	13	14	15–16	17–18	19	20–21	22–23	24	25–26	27–28	29–30	1–3	4–5	6–11	12–16	
A.....	(x)	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	(x)	(x)	—	—	—	—	—	—	—	—	—	—	
B.....	(x)	—	—	—	—	—	—	—	—	—	(x)	—	—	—	—	—	—	—	—	x	—	—	—	—	—	—	—	—	—	—	
C.....	x	—	—	(x)	x	—	—	—	—	—	—	—	—	—	—	—	—	—	—	(x)	(x)	—	—	—	—	—	—	—	—	—	
D.....	(x)	—	—	—	(x)	—	—	—	—	—	—	—	—	—	—	—	—	—	—	(x)	—	(x)[2]	—	—	—	—	—	—	—	—	
E.....	(x)	—	(x)	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	(x)	—	—	—	—	—	(x)[3]	—	—	—	—	
F.....	(x)	—	(x)	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	xx	(x)	—	—	—	—	—	—	—	—	—	
G.....	—	—	—	—	x	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	
Weather Conditions	Part Cloudy	Clear	Clear	Light showers	Not recorded	Part Cloudy	Clear	Clear	Part cloudy	Clear	Clear	Clear	Clear	Clear	Clear	Clear	Clear	Clear	Clear	Rain night of 17th	Showers	Showers	Showers	Showers	Showers	Showers	Showers	Showers	Clear	Clear	Clear	Observations discontinued.
Precipitation.	—	—	—	T	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	.26	.08	—	.15	T	.42	.21	.30	.47	—	—	—	

2. 8 spores from one ascus found; all found to have germinated.
3. 8 spores from one ascus; none germinated when found.

The earliest spore ejection noted at Hood River occurred on March 20, 1916; the last dicharge was observed on June 27 or more than three months later. In the case of leaf "E," spores were ejected on 38 different days during this time. The perithecia on leaf "G," ejected spores on but 25 days. The average for the seven leaves studied show that spore ejection occurred on more than 30 days.

During the spring of 1916 there were five distinct periods of abundant ascospore discharge, during which time the weather was ideal for general scab infection. There were also four periods of limited ejection with weather conditions less favorable for infection. The longest continuous discharge recorded began on May 5 extending to May 14, inclusive, or 10 days. No rain fell on the 13th and 14th; moisture, however, from prevailing heavy dews was sufficient to wet the slides and leaves, causing spore ejection on these days. This would probably not occur to any great extent under normal conditions. The glass slides often collected moisture from heavy dews. This excess moisture probably accounts for occasional light discharges of spores on days upon which no precipitation occurred.

The first period of ejection, occurring between March 20 and 28, took place before the winter buds on the trees had expanded, with the possible exception of a few very early varieties, and on this account practically no infection resulted. Newtowns and Spitzenburgs, the most widely grown varieties in the valley, were largely dormant. By referring to the record, it will be found that the second period of ejection began on April 10. With the exception of the thirteenth, a relatively abundant discharge of spores occurred until the twenty-first; the weather conditions were ideal for the germination of scab spores. Fortunately, the delayed-dormant lime-sulfur application had been made by most of the orchardists between the sixth and the tenth of the month. On the writer's experimental plots this spray was applied on the eighth. Figure 2 shows the condition of the trees at the time of this heavy spore discharge.

The trees had not sufficiently developed to receive the pink or cluster bud application of spray until the twentieth to the twenty-fifth. Applications made during this period were again advantageously applied, since many days of abundant spore ejection immediately followed. Scab infections were first observed on unsprayed trees on May 8. This conidial growth was undoubtedly the result of infections which took place between April 10 and 21, as the limited time elapsing following the last period of discharge would hardly warrant drawing any other conclusion.

It was observed that as a rule the heaviest spore ejection occurred during the first two or three days of a rainy period, gradually becoming less towards the end of the wet weather. It would appear then that the store of spores, which was ripened during the dry weather, are released abundantly during wet weather and that discharge during the latter part of the wet periods occurs as the spores reach maturity. The spore ejection which began on May 21 and extended to May 26, was the last of importance. On some of the slides (e. g., C and F) large numbers of spores were obtained daily during this period of rainy weather. Following this activity, settled weather prevailed until June 17, at which time a light discharge of spores was obtained on all but one of the slides. With but one exception, ejection ceased (slide E) even though ideal weather conditions prevailed until July 4. Severe conidial infection took

place during this period in poorly sprayed orchards. Scab infections developed on some of the writer's unsprayed check trees from 30.3 percent (primary or ascospore infections) to a total of 97.04 percent.

Comparison of Local Ascospore Ejection With That Reported in Other Localities

As Table III indicates, ascospore ejection began not only more than a month earlier than has been previously recorded but extended to a much later date than has been supposed. Clinton (4) reports that the ascospore reaches maturity during April and May in Illinois. He gives no specific date. Wallace (1908) (4) has made observations in New York relative to initial ejection; he states that, "In nature the ascospores usually begin to mature at or about the time when the blossoms are ready to open. In 1908 mature spores were found on May 4, and in 1910 on May 1. In each case the blossoms were about to open." Morse (1915) (5) reports the following observations in the State of Maine: "On May 3 only immature asci or spore sacs could be found. These leaves were moistened and then placed in covered porous flower pot saucers, the latter being partly sunk in the soil out of doors to prevent drying out and to keep the leaves as near under normal conditions as was possible. Asci were still immature at the time of the first application of the fungicidal spray on May 14 and did not reach maturity until 4 or 5 days thereafter."

Seasonal development was much earlier at Hood River in both 1914 and 1915 than during the past year. On March 21, 1915, the following is quoted from the notes of J. R. Winston relative to the condition of scab on the fallen leaves: "Scab on the old leaves is quite abundant and the perithecia appear somewhat larger than usual. Found ascospores of **Venturia inaequalis** in all stages of development from mature ascospores to masses of protoplasm without differentiation. There is an abundance of spores ready for ejection with the occurence of favorable circumstances." In this year Winston found the primary infections appearing on the foliage on April 20, or 18 days earlier than was the case in 1916.

Professor H. P. Barss, Plant Pathologist, and G. B. Posey, Research Assistant, department of Plant Pathology, Oregon Agrcultural Experiment Station, have also made observations relative to the ascospore discharge of this fungus in the Willamette Valley at Corvallis, Oregon. General climatic conditions are somewhat different there than at Hood River; the winters being usually milder, with a greater rainfall.

The first observations on ascospore development were made by Professor Barss during the early part of February, 1915. At that time perithecia were noted to be plentiful and leaves were brought into the laboratory. On February 19, mature ascospores were discharged from the leaves. No further observations were made in that year.

On February 25, 1916, Professor Barss placed glass slides for 24 hours over six old scabby apple leaves and four scabby pear leaves in the

(4) Clinton, G. P. Apple Scab. Univ. of Ill. Exp. Sta. 1901. Bul. 67.
(5) Morse, W. J. Spraying Experiments and Apple Diseases. Maine Exp. Sta. 1915, Bul. 252.

field and found that ascospores were ejected in all cases, although not in abundance.

On April 13, 1916, Mr. Posey tested several leaves of both apple and pear, finding abundant ejection of ascospores from all.

On May 20, 1916, Mr. Posey again gathered apple and pear leaves and reported that a great number of ascospores were being shot from the apple leaves, although none could be observed in the case of pear leaves. No later observations were made at Corvallis during the remainder of the season. These investigators, however, demonstrated that beginning with the end of February ascospore ejection extended over a period of at least three months with a probability that it continued even longer.

CONCLUSIONS

Mature ascospores were first ejected at Hood River on March 20, 1916. At Corvallis, Oregon, this activity was first noted on February 25. In both localities apple trees were largely dormant.

The last ejection of ascospores occurred at Hood River on June 27. At Corvallis abundant discharge was observed as late as May 20, after which date no examinations were made.

The study of ascospore discharge demonstrates beyond a doubt that the delayed-dormant application given while the leaves are small and undeveloped (See Fig. 2) cannot be safely dispensed with in the Hood River and Willamette Valleys, at least since the ejection of ascospores begins before the foliage has even started. This would probably also hold true in all sections of Western Oregon and Washington, including the White Salmon Valley. It is not at all unlikely that other sections could also be included except for our present limited knowledge.

Owing to the great length of ascospore activity, orchardists are not safe in omitting any one of the later spring sprays even though conidial infections were not present, for fear of infections from the primary source. In 1916 a long period existed during which infections could originate from both ascospores and conidia.

It is the belief of the writer that much can be learned in other sections of the country relative to early ascospore discharge, and that if an annual study were made to determine the earliest period of ejection, recommendations might often be issued by plant pathologists which would prevent some of the serious outbreaks that now occasionally occur in some sections, in spite of the usually recommended applications of spray.

The author is indebted to Professor Barss, Plant Pathologist; Mr. Posey, former Research Assistant of the Oregon Experiment Station; and to Mr. J. R. Winston, former Pathologist of the Hood River Station, for the use of their notes relative to the ascospore activity of this fungus. Professor Barss has also offered many suggestions that have assisted in the investigation as well as the preparation of this paper.