Notes on Some Leaf-spot Diseases of Broadleaved Trees- I. Cercospora leaf-spot of plane trees.

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With Plates I—IV

Introduction

The tree species belonging to the genus Platanus havs been used extensively for roadside planting in Japan. Some years ago the senior author observed and collected in Tokyo a leaf-spot disease of the oriental plane tree (*Platanus orientalis* L.) planted for roadside tree caused by a species of Cercospora, but the damage was not serious.

In the summers of 1948~50, a large number of Platanus seedlings were severely attacked by the same fungus in the nurseries of the Government Forest Experiment Station at Meguro in Tokyo.

Since it has already caused considerable loss and bids fair to become an important disease here, it has been made a subject of investigation by the authors.

ISHIZAKA (1914) made a brief note on a foliage disease of *P. occidentalis* L. caused by *Pestalotia funerea* DESM., and some years later NAMBU (1917, 1920) reported an anthracnose of the same plant caused by *Gloeosporium nervisequum* (FUCKEL) SACC. (*Gnomonia veneta* (SACC. et SPEG.) SACC.). *Discosia Artocreas* (TOD.) FR. and *Mycosphaerella maculiformis* (PERS.) AUERSW. were listed by SHIRAI and HARA (1927) as the fungi inhabiting on the leaves of the plane trees.

So far as can be ascertained by the authors, there is no account concerning the occurrence of any other fungi parasitic on the leaves of the Platanus in Japan.

Regarding the important foliage diseases of sycamores some contributions were published in America. APOSTOLIDES (1929) and D.J. SMITH and C.O. SMITH (1939) made some studies on *Stigmina platani* (FUCKEL) SACC. parasitic on *Platanus racemosa*. *Stigmella Platani-racemosae* DEARN. et BARTH., a new species similar to *S. platani*, was described by DEARNESS (1929). SUMSTINE (1936) noted a powdery mildew by *Oidium abductum*, a variety of *Phyllactinia corylea* (PERS.) KARST. WOLF (1938) reported in detail the life histories of two leaf-inhabiting fungi on *P. occidentalis*, *Cercospora platanifolia* ELLIS et EV. and *Stigmina platani* (FUCKEL) SACC., and made clear the ascigerous stage of each of these two fungi.

It is the purpose of the present paper to report a foliage disease of sycamores unreported in Japan and to give a summary of the results of the authors' observations and experiments obtained in the past three years.

The authors are much indebted to Mr. Yasaka HAYASHI, of Government Forest

Experiment Station, for his kindness in identifying the host plants.

Symptom and damage

In the nurseries under the observation in Tokyo, the disease first makes its appearance in the middle of June and is found any time thereafter until the leaves have been shed. There is an evident tendency, toward leaf spot infection to start on the younger leaves. This seems to indicate that much of the infection takes place while the leaves are young.

The lesions are at first pin-point-like, circular, brown spots present on the under surface of the leaf, while light brown on the upper surface. The shape of the lesions is commonly circular, 0.5～3 mm. in diameter, and frequently irregular. They become dark brown with a border of light color, about 1 mm. in width. The lesions on the petioles appear in the form of small deep brown spots which are usually somewhat longer than wide, 0.1—1×1—2 mm. in size, and extend up and down the petiole. The effects of the malady on the petioles is never serious. The necrotic tissues gradually become dry and brittle and when the lesions are abundant they coalesce to form large, irregular, dead areas, sometimes 15～20 mm. in size. In the case of severe infections the diseased leaves are curled and rather serious defoliation results. Under or both leaf surfaces of mature lesions are covered with a number of minute black dots, barely visible to the naked eye.

The disease is frequently observed and often a large percentage of the leaves on the lowest parts of branches of the mature trees is affected. After the rainy weather in the summer, the damage of the disease appears very distinct.

In the nursery plantings the leaf spot is particularly destructive. Very often, by midsummer, the seedling beds are severely defoliated and the growth of the young trees is greatly retarded. When the disease has become established in a block of young seedlings it usually causes considerable damage each year.

All of the three species of Platanus planted commonly in Japan, *P. acerifolia* Willd., *P. orientalis* L. and *P. occidentalis* L., are affected by the disease. Under field conditions it may be seen that *P. acerifolia* is very susceptible, *P. orientalis* modelately and *P. occidentalis* less susceptible (Pl. I, A, B,; Pl. II, A, B, C, D).

Causal fungus

On the lesions numerous fruit-bodies of a Cercospora are usually produced. Occasionally *Stigmina platani* (Fuckel) Sacc.? is found on the old necrotic portions of the leaf, but not on the fresh lesions (Pl. IV. E, F).

(1) Isolation and culture of the Cercospora

Cultures of the fungus were obtained from both of *P. acerifolia* and *P. orientalis*. Monoconidial isolations were made by a modification of Yoshii's (1933) method using two per cent aqueous solution of copper sulphate to avoid the bacterial contamination.

The growth of the fungus on agar media was slow, and aerial mycelium was dirty gray at first, then forming a gnarled, piled-up mat covered with the white mycelium on the surface of media. Conidia were produced abundantly on the surface of the younger gray colonies, but never on the old white mycelia (Pl. Ⅲ. G.).

All of the cultures isolated from the two hosts were the same in the macroscopic and microscopic appearances.

(2) Inoculation experiments

In order to test the pathogenicity of the fungus, the inoculation experiments were performed on the leaves of the potted healthy seedlings (sowed on May 16, 1949) of *P. acerifolia* and *P. orientalis* during the summer of 1949. The fungous culture which had been derived from the monoconidial isolate obtained from the lesion of *P. acerifolia* and cultured for two weeks on potato-glucose agar was used as the inoculum. The organism from the slant was first broken up in sterilized water, then filtered through double sheets of cotton cloth. Potted seedlings were sprayed with the spore suspension containing some fragment mycelia by means of an atomizer, then being covered with bell-jars for two days. The check-plants were sprayed with sterilized water instead of the fungous suspension.

On August 13, the seedlings both of *P. acerifolia* and *P. orientalis* were inoculated with the fungus. The results of the experiments examined on September 3 are summarized in Table 1.

Table 1. Results of the inoculation experiments.

Pot no.	Kind of tree	Inoculated or Control	Number of seedlings	Number of infected seedlings	Maximum number of lesions on a leaf
1	*P. acerifolia*	Inoculated	14	4	24
2	do.	do.	13	2	12
3	do.	Control	12	0	0
4	*P. orientalis*	Inoculated	11	7	28
5	do.	do.	10	4	6
6	do.	Control	9	0	0

Re-isolations were made from the conidia produced on the lesions resulting from the inoculation. The mycelial colonies and conidia obtained were of the same character as the monoconidial culture used as the inoculum. All of the check-seedlings remained healthy (Pl. Ⅲ, A, B).

It is obvious from Table 1 that the pathogenicity of the fungus on the plane trees was proved by the authors and there were no remarkable differences in susceptibility to the disease between the two species of Platanus, *P. acerifolia* and *P. orientalis*.

Another inoculation test was made by the use of Petri dishes noted by Clinton and McCormick (1924). The inoculum consisted of conidial suspensions in sterilized distilled water was smeared on the upper or lower leaf surface of

P. acerifolia by means of a platinum loope. At the end of ten days the results of the test showed that the lower-surface inoculation resulted in heavy infection, while, the upper-surface inoculation resulted either in no infection at all, or in very light infection.

The stomata of the mature healthy leaves of *P. acerifolia* are distributed on both surfaces, but the number of the stomata per unit square of the upper surface is very small comparing with that of the lower surface (5.3 : 100).

To know the mode of infection the preliminary experiments were done by the surficial observations of the inoculated leaves. In all the material examined, no indication that the fungus entered by penetration of either the upper or lower epidermis was ever observed. On the other hand entrance through the stomata was observed very frequently.

(3) Identification of the Cercospora

The morphological characters of the Cercospora are as follows: Fruit-bodies of the fungus occur on both leaf surface on the lesions. The tuberculate or subglobular stromata from which the conidiophores arise are pseudoparenchymatous, olivish brown and occupy the stomatal opening. Each fascicle is composed of 10～30 laxly-spreading conidiophores. Conidiophores are non-septate, no-branching, light brown, 9～20 μ in length and 2～5 μ in width. The conidia are hyaline or very light brown, curved, clavate or helminthosporioid, commonly 4-6 septate, rarely 3- or 7-septate; range from 30—80×4—6 μ (Pl. Ⅳ.A, B.).

Chupp (1937) listed two species of Cercospora parasitic on the Platanus, namely *C. platani* and *C. platanicola*, without any description and comparison.

The authors' Cercospora is closely related in many respects to *C. platanifolia* Ellis et Ev., which was first described by Ellis and Everhart (1887) and then studied in detail by Wolf (1938).

There are seen some differences in the dimension of the conidia between *C. platanifolia* and the present authors' fungus. This discrepancy in size may probably due to the environmental conditions here and there under which they inhabit, as pointed out already by Nakata et *al.* (1922), Welles (1924) and etc. on other species of Cercospora. The present authors' fungus causing the leaf-spot disease of plane trees in Japan may be the same as *Cercospora platanifolia* Ellis et Ev., though no direct comparison has been conducted.

By Wolf (l. c.) it was reported that *Mycosphaerella platanifolia* Cooke (syn. *Sphaerella platanifolia* Cooke) found on the old fallen leaves of *P. occidentalis* was the ascigerous stage of *C. platanifolia*. The authors have observed in Japan the formation of spermogonia and young perithecia on the fallen diseased leaves of plane trees, but never mature perithecia and asci (Pl. Ⅲ. D, E. F).

Germination of conidia

(1) Germination of fresh conidia

The conidia germinate readily in distilled water, and the germination occurs

within several hours at summer room temperature.

To know the time required for the germination and the detailed germination processes, the authors carried out the following experiments. Conidia used for this experiments were collected from the seedling of *P. orientalis* on August 31, 1949. The conidia were sown on two per cent glucose agar in Petri dishes and kept at 25°C.

Results of the experiments are shown in Table 2.

Table 2. Germination of conidia (1).

Time passed (hours)	5	7	9	18	26
Number of conidia observed	—	188	101	177	193
Number of germinating conidia	0	4	34	150	184
Germination percentage (%)	0	2.1	33.6	84.7	95.3
Maximum length of germ-tube (μ)	—	—	—	175	229

From Table 2, it is clear that the conidia of the fungus germinate after about seven hours at 25°C., and almost all of the viable conidia have germinated within 26 hours at the same temperature. The germination starts most frequently from both of the end cells, less commonly from either end, and very rarely from the intermediate cells (Pl. Ⅳ. C).

Two supplementary germination tests were done using the other materials, and the results of these experiments after 20 hours on two per cent glucose agar will be shown in Table 3.

Table 3. Germination of conidia (2).

Experiment no.	Number of conidia observed	Number of germinating conidia	Germination percentage (%)
I	199	128	64.3
II	140	83	59.2

The effect of the temperatures upon the germination of the fresh conidia collected from *P. acerifolia* was tested by Van Tieghem cell method using sterilized distilled water. Results of the experiment at the end of 24 hours are summarized in Table 4.

Table 4. Effects of the temperature upon the conidial germination.

Temperature (°C)	1~7	23	R.T. (26~29)	33	37
Number of conidia observed	873	1078	1075	803	830
Number of germinating conidia	0	824	935	454	18
Germination percentage (%)	0	76.4	86.9	56.6	2.2
Range of germ-tube length (μ)	—	162~250	175~250	37~75	6~25

From the data shown in Table 4, it may be said that the optimum temperature for the germination of the conidia lies at the temperature between 23° and 29°C.

(2) **Germination of conidia on the dried specimen**

The diseased leaves of *P. orientalis* having many lesions were collected on July 12, 1949 and placed in the laboratory.

On November 24, 1949, about four months after the collection, germination of the conidia on the dried specimen was examined by the same procedure mentioned above, and the high percentage of germination (about 60 per cent) was gained.

While, on the contrary, on July 8, 1950, the germination percentage became extremely small (0.8 per cent).

From these results it may be said that the longevity for survival of the conidia under such condition is about a year. Data of the experiments will be given in Table 5.

Table 5. Germination tests of the conidia on the dried specimen.

Period of storage (months)	Temperature tested (°C)	Time incubated (hours)	Number of conidia observed	Number of germinating conidia	Germination percentage (%)	Maximum length of germ-tube (μ)
ca. 4 (XI—24—49)*	25	18	625	292	46.7	72.5
		25	932	571	61.2	143.0
		42	765	506	66.1	328.5
ca. 6 (I—13—50)	25	24	1780	57	3.2	—
		40	1455	136	9.3	416.0
ca. 12 (VII—8—50)	26~32	30	720	6	0.8	—

* XI—24—49 (Date of observation) means November 24th, 1949.

Conidial production of the fungus in pure culture

WOLF (l. c., p. 59), working with *C. platanifolia* on *P. occidentalis*, stated that "conidial production was not noted in any of the cultures, and for this reason artificial inoculations with pure cultures were not attempted". The present authors, however, obtained conidia in culture with the same fungus isolated in Japan.

Conidia of the organism were produced abundantly on the young culture. As the developing mycelia progressively covered the surface of the medium, few and fewer conidia were produced, and later none of them were found at all on the old mycelial colonies.

The effect of different media and a procedure for maintaining cultures in a sporulating condition studied by the authors are briefly described as follows:

(1) **Conidial production on agar media**

The cultures from conidia were obtained by making a suspension of fresh

conidia collected from the diseased leaves of the host and streaking loopfuls of the suspension over the surface of Petri dishes containing hardened two per cent glucose agar. After about 24 hours the germinating conidia were transplanted to agar slant and kept at desirable temperatures. To avoid the bacterial contamination the modified Yoshii's (l. c.) isolation method was employed.

The conidial production on agar media was examined under the microscope at various intervals. Data obtained on both of potato-glucose and two per cent glucose agar at 24°—26°C will be shown in Tables 6—8, and those at the lower temperatures in Tables 9—10.

Table 6. Conidial production on potato-glucose and 2 per cent glucose agar (1).

Experiment-1. Host: *P. orientalis*,
Temperature tested: 24°~26°C.

Period of culture (days)	Degree of conidial production	
	Potato agar	2% glucose agar
6	−	−
7	+	+
10	+	+
12	+	+
15	+	+
19	++++	++++
23	+++	++++
26	++	++++
35	+	++++
45	+	++++
68	+	++++
77	+	++

Notes: +……Conidial production is present,
−……conidial production, absent.

Table 7. Conidial production on potato-glucose and 2 per cent glucose agar (2).

Experiment-2. Host: *P. acerifolia*,
Temperature tested: 24°~26°C.

Period of culture (days)	Degree of conidial production	
	Potato agar	2% glucose agar
7	−	−
8	−	−
9	−	−
12	+++++	+++++
16	+++	+++++
19	++	++++
28	+	+++
38	+	+++
61	+	++
70	−	++

Table 8. Conidial production on potato-glucose and 2 per cent glucose agar (3).
Experiment-3. Host: *P. orientalis*,
Temperature tested: 24°～26°C.

Period of culture (days)	Degree of conidial production	
	Potato agar	2% glucose agar
6	−	−
11	+++	+++
14	+++	+++
24	++	+++
34	+	+++
56	+	+++
65	+	+++
87	+	++
97	+	++
107	+	++
117	+	++

Table 9. Conidial production on potato-glucose and 2 per cent glucose agar (4).
Experiment-4. Host: *P. acerifolia*,
Temperature tested: 18°C.

Period of culture (days)	Degree of conidial production	
	Potato agar	2% glucose agar
8	−	−
13	+	+
16	+	+
19	+	+
28	+	+
36	+	+
68	+	+
106	+	+

Table 10. Conidial production on potato-glucose and 2 per cent glucose agar (5).
Experiment-5. Host: *P. acerifolia*,
Temperature tested: 15°C.

Period of culture (days)	Degree of conidial production	
	Potato agar	2% glucose agar
9	−	−
12	−	−
21	−	−
29	−	−
61	−	−
99	+	+

As shown in Tables 6—8, the conidial production was found on both agar media after about a week and maintained for more than one hundred days. On potato-glucose agar the conidia were observed most abundantly after 10 to 20 days and later they decreased in number gradually. While, on two per cent glucose agar, a large number of conidia has been seen for longer.

It is evident, from the data in Tables 9—10, that the conidial production was not good at the lower temperatures (15° and 18°C.).

There was considerable variation in the amount of sporulation among the different isolates; some produced conidia profusely, others less abundantly, and some sparingly.

The same experiments were made on the following agar media: 0.5 per cent glucose agar, one per cent glucose agar, two per cent peptone agar, peptone-glucose agar[1], Platanus-decoction agar[2] and Platanus-decoction with one per cent glucose agar.

Results obtained will be summarized in Tables 11—12.

Table 11. Conidial production on various agar media (1).

Period of culture (days)	Degree of conidial production					
	0.5% glucose agar	1% glucose agar	2% peptone agar	Peptone-glucose agar	Platanus-decoction agar	Platanus-decoction plus 1% glucose agar
7	−	−	−	−	−	−
9	−	−	−	−	−	−
11	−	+	−	−	++	+
17	+++++	+++++	+	−	+++++	+++++
26	+++++	+++++	+	−	+++++	+++
36	+++++	+++++	+	+++	+++	++
46	+++++	+++++	+	++	+++	++
56	+++	+++++	+	++	++	++
66	+++	++++	+	++	++	++
76	++	++++	+	++	++	++
86	++	+++	+	++	++	++

Table 12. Conidial production on various agar media (2).

Period of culture (days)	Degree of conidial production					
	0.5% glucose agar	2% glucose agar	2% peptone agar	Peptone-glucose agar	Platanus-decoction agar	Platanus-decoction plus 1% glucose agar
6	−	−	−	−	−	−
17	++++	++++	−	−	+++++	+++++
27	+++++	+++++	+	−	+++++	+++++
37	+++++	+++++	+	−	+++++	++++
47	+++++	++	++	−	++++	++
57	+++++	++	++	−	+++	++
67	+++	++	++	+	++	++
77	++	++	+	+	++	++

1) Distilled water 1,000 cc., peptone 20 g., glucose 20 g., agar-agar 30 g.

2) Distilled water 1,000 cc., young succulent leaves of *P. acerifolia* 100 g., agar-agar 30 g.

From Tables 11—12, it may be said that the conidia are produced on all agar media used and there are no remarkable differences in the amount of conidia produced among them, except two agar-media, namely two per cent peptone and peptone-glucose agar.

(2) Maintaining cultures in a sporulating condition

The Cercosporae as a group have been found to produce few typical conidia in pure culture. Various workers have reported many results of their studies on the conidial production of Cercosporae in pure culture, because considerable difficulty was encountered in obtaining and maintaining conidial production in artificial media in many species of Cercospora (NAKATA et *al.* 1922, NAGEL 1934, LEWIS 1940, DIACHUN and VALLEAU 1941, IKATA 1942, etc.). NAGEL (l. c.) gave a résumé of the studies on this subject up to that time and stated the results of his own experiments.

As noted already the present authors have obtained conidia with *Cercospora platanifolia* in the younger culture isolated from the conidia in nature for about four months, but could not gain conidia in the old culture being more than five months in age. Small pieces of the old mycelial colony having no conidia were transplanted to new substrata and tried by various treatments, but the conidial production was never observed, although the fungus made a good vegetative growth.

By a modification of NAGEL's (l. c.) "newly-made isolation method" the authors have been able to keep cultures of the fungus in a sporulating condition for a long time.

On September 12, 1949, isolations were made from conidia obtained from diseased leaves of *P. acerifolia* by the modified YOSHII's (l.c.) method. These isolates were transferred to potato-glucose agar on September 30, 1949, and then once more on October 8 in the same year. Microscopic observation showed that abundant conidial production had taken place in the mycelial colonies. Transfers were made from these mycelial colonies to two per cent glucose and potato-glucose slants, and continued for about 10 months at the intervals of about 10 days.

The cultures were held at a temperature of approximately 25°C over a period of 10 months keeping from rapid desiccation of the media, and during this time 28 consecutive transfers were made.

On potato-glucose agar, abundant conidia were produced throughout the entire period, and the amount of conidial production at the end of this time was not less than at the initial culture. Comparing with the former case, however, the duration of conidial production on glucose agar was remarkably short.

Data of the authors' experiments will be summarized in Table 13.

Table 13. Conidial production on consecutive transfers of culture.

Consecutive transfers					Control (no-transferred)	
Date of transplanting	Number of generation	Conidial production			Conidial production	
		Date of observation	2% glucose agar	Potato agar	Date of observation	Potato agar
X—14—49	3	X—31—49	+++	+++		
XI—2—49	4	XI—18—49	++++	++	XI—16—49	++
XI—18—49	5	XI—28—49	+++	+++++		
XI—28—49	6	XII—8—49	+	+++	XII—8—49	++
XII—8—49	7	XII—18—49	+	+++	XII—18—49	+
XII—18—49	8	XII—28—49	+	++++	XII—28—49	+
XII—28—49	9	I—7—50	+	+++++	I—7—50	+
I—7—50	10	I—17—50	+	+++++	I—17—50	+
I—17—50	11	I—27—50	+	++++	I—27—50	—
I—27—50	12	II—6—50	+	+++++	II—6—50	—
II—6—50	13	II—16—50	—	+++++	II—16—50	—
II—16—50	14	II—26—50	—	+++++	II—26—50	—
II—26—50	15	III—8—50	—	+++++	III—8—50	—
III—8—50	16	III—18—50	—	+++++	III—18—50	—
III—18—50	17	III—30—50	—	++++	III—30—50	—
III—30—50	18	IV—12—50	—	++++	IV—12—50	—
IV—12—50	19	IV—22—50	—	++++	IV—22—50	—
IV—22—50	20	V—2—50	—	++++	V—2—50	—
V—2—50	21	V—12—50	—	++++	V—12—50	—
V—12—50	22	V—22—50	—	++++	V—22—50	—
V—22—50	23	VI—1—50	—	++++	VI—1—50	—
VI—1—50	24	VI—12—50	—	++++	VI—12—50	—
VI—12—50	25	VI—21—50	—	++++	VI—21—50	—
VI—21—50	26	VII—1—50	—	++++	VII—1—50	—
VII—1—50	27	VII—11—50	—	++++	VII—11—50	—
VII—11—50	28	VII—21—50	—	++++	VII—21—50	—

Conidia produced in the young culture were similar in shape and size to those produced on natural leaf spots. However, after many consecutive transfers, they were deep brown in color, distinctly constricted at the septata, very granular in the cell content and often abnormal in the form.

To test the influence of the consecutive transfers upon the pathogenicity of the fungus, inoculation experiments were conducted by the authors. On June 12, 1950, the leaves of the potted seedlings of *P. acerifolia* were sprayed with the conidial suspension taken from the fungous culture. The culture used had been held at about 25°C. over a period of nine months and during this time 25 consecutive transfers had been made at the intervals of about 10 days.

On June 19, 1950, a week later, the incipient stage of the symptom of this disease was observed on the inoculated plants. On July 4, 1950, about three weeks after inoculation, the inoculated seedlings were diseased very severely.

The lesions resulting from inoculum bore conidiophores and conidia of *C. platanifolia*. (Pl. Ⅲ. C).

From the results of the experiment it was shown that the pathogenicity of the fungus was never diminished in virulence by many consecutive transfers, at least 25 transfers during nine months.

Summary

In the present paper the authors deal with the results of some studies on a Cercospora leaf-spot disease of tree species of Platanus unreported in Japan.

The fungus was identified by the authors as *Cercospora platanifolia* Ellis et Ev. (*Mycosphaerella platanifolia* Cooke), and the pathogenicity of the fungus was proved by inoculation experiments. The incubation period of this disease was 7～10 days in the summer.

All of the three species of Platanus observed, *P. acerifolia*, *P. orientalis* and *P. occidentalis*, were attacked by the fungus, but *P. occidentalis* was less susceptible.

The conidia on the dried specimen placed in the laboratory germinated well after four months, while, however, the germination percentage was very small at the end of a year.

Conidia were produced abundantly on younger cultures, but few or no conidia were found on 4-month-old culture. By the method of consecutive conidial transfers at about 10-day intervals, the authors were able to maintain the cultures in a sporulating condition for 10 months and obtained abundant conidia even at the end of this experimental period. Conidia produced in pure culture which had been maintained in a sporulating condition by 25 consecutive transfers were deep brown in color, granular and often abnormal in form. Pathogenicity of these conidia was proved by the inoculation experiment.

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Explanation of plates

Plate I.

A. Seedlings of *P. acerifolia* attacked by *C. platanifolia* in the nursery. Photographed on August 31, 1948.

B. Young seedlings of *P. acerifolia* attacked by *C. platanifolia.* x 1.

Plate II.

A. Leaf-spot of *P. acerifolia* caused by *C. platanifolia* (the upper surface). x 2/3.

B. Leaf-spot of *P. orientalis* caused by *C. platanifolia* (the upper surface). x 2/3.

C. Leaf-spot of *P. occidentalis* caused by *C. platanifolia* (the under surface). x 2/3.

D. Magnified lesions of *P. acerifolia* caused by *C. platanifolia* (the under surface). x 1.5.

Plate Ⅲ.

A. Diseased leaves of *P. acerifolia* caused by artificial inoculation with pure culture of *C. platanifolia.* x 1.

B. Diseased leaves of *P. orientalis* caused by artificial inoculation with pure culture of *C. platanifolia.* x 1.

C. Diseased leaves of *P. acerifolia* caused by artificial inoculation with the conidia of *C. platanifolia* produced in pure culture, which had been maintained by 25 consecutive transfers at the intervals of 10 days. x 5/7.

D. Spermogonia and young perithecia of *C. platanifolia* formed on the diseased fallen leaf of *P. acerifolia.* x 2.

E～F. Sections of spermogonia and young perithecia of *C. platanifolia* formed on the diseased fallen leaves of *P. acerifolia.* x 120.

G. Mycelial colonies of *C. platanifolia* on agar media. x 5/6.
(From left to right)

a. After 13 days at 25°—30°C. on 2 per cent glucose agar. Conidia are observed abundantly.

b. After 13 days at 25°—30°C. on potato-glucose agar. Conidia are observed abundantly.

c. After 30 days at 25°—30°C. on potato-glucose agar. Surface of the colony is covered with the white mycelium. A few conidia are observed beneath the white mycelium.

Plate Ⅳ.

A. Conidiophores of *C. platanifolia.*

B. Conidia of *C. platanifolia* produced on the diseased leaf of *P. acerifolia.*

C. Germinating conidia of *C. platanifolia.*

D. Conidia of *C. platanifolia* produced in pure culture made by 25 consecutive transfers at 10-day intervals.

E. Conidia of *Stigmina platani*? associated with *C. platanifolia* on the old lesions.

F. Germinating conidia of *S. platani*?.

廣葉樹の斑點性病害に關する研究-I.

スズカケノキの褐點病 *,**

（摘　要）

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農林技官　保坂義行

本論文は日本に於てはこれまで記載されたことのないスズカケノキの斑点性病害について述べたもので，病原菌を *Cercospora platanifolia* Ellis et Ev.（*Mycosphaerella platanifolia* Cooke）と同定し，病名を新にスズカケノキの褐点病と称する。

本病害はスズカケノキ（*Platanus orientalis*），モミヂバスズカケノキ（*P. acerifolia*），及びアメリカスズカケノキ（*P. occidentalis*）のすべてに認められるが，これらのうちアメリカスズカケノキはこの病害に対してやや強い傾向がある。

成木の葉が侵された場合はさしたることはないが，幼苗が本病に罹ると病状はかなり烈しくその被害は軽視しえない程大きい。

(1) 病　　徴

本病は東京では6月中旬から晩秋まで認められ高溫期の雨後には急速な伝播をみる。初め病斑は褐色小点状を呈し直径 0.5～3 mm であるが，やや不規則な形状をとることもある。病斑の周縁部は淡緑～黄色の暈帯でとりまかれる。病斑は漸次拡大するとともに形状もまた不規則となり，各病斑が融合して時に 15×20 mm 以上に達することがある。病斑は往々葉柄にも形成される。

幼若多汁な葉が侵されると斑点は急速に拡大し，更に融合して巨大な病斑となり，壊死部は黒褐色，しばしば畸形を呈して葉表に捲く。

病斑上には暗濃緑色の病原菌子実体が形成され，病葉は早期に落葉する。

(2) 分生胞子の発芽

本菌の新鮮な分生胞子は蒸溜水中で僅か7時間内外（25°C）で発芽し，26 時間後には大部

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*　病徴からみて褐斑病とするのがより適切と思われる場合もあるが既に石坂 (1914) は *Pestalotia funerea* Desm. によるスズカケノキの斑点性病害に対してこの病名を使用している。それでこれと混同するのを避けるため著者等は本病の病名として新に褐点病を選んだ。

** 本病の防除法　(1) 病葉及び落葉を完全に焼却すること。(2) 連作は絶対に避けること。(3) 密植にならぬよう注意すること。(4) 6斗式ボルドウ合剤を撒布すること。春季新葉開舒後間もなく（東京では6月上旬）第1回を行い，9月下旬まで毎月2回実施する。特に梅雨前後には回数を増加し，又撒布方法は地面に近い処から上部に向けて葉裏に薬剤を噴霧するようつとめる。

分の胞子が発芽する。発芽に対する最適温度は 23°C から 29°C の間にある。

乾燥標本上の胞子は室内保存4ヶ月後には発芽が極めて良好であるが6ヶ月後には甚しく不良となり，約1ヶ年後には極めて少数のものが発芽するにすぎない。すなわち乾燥標本上の胞子の生存期間は約1ヶ年である。

(3) 培養基上に於ける分生胞子の生成

Wolf (1938) の研究によれば本菌は人工培養基上に分生胞子を形成しないと言うことであるが，著者等はこれを認めた。しかし著者等の実験でも培養基上の胞子は次第に数を減じ，培養約5ヶ月後には全然認めることが出来なかつた。

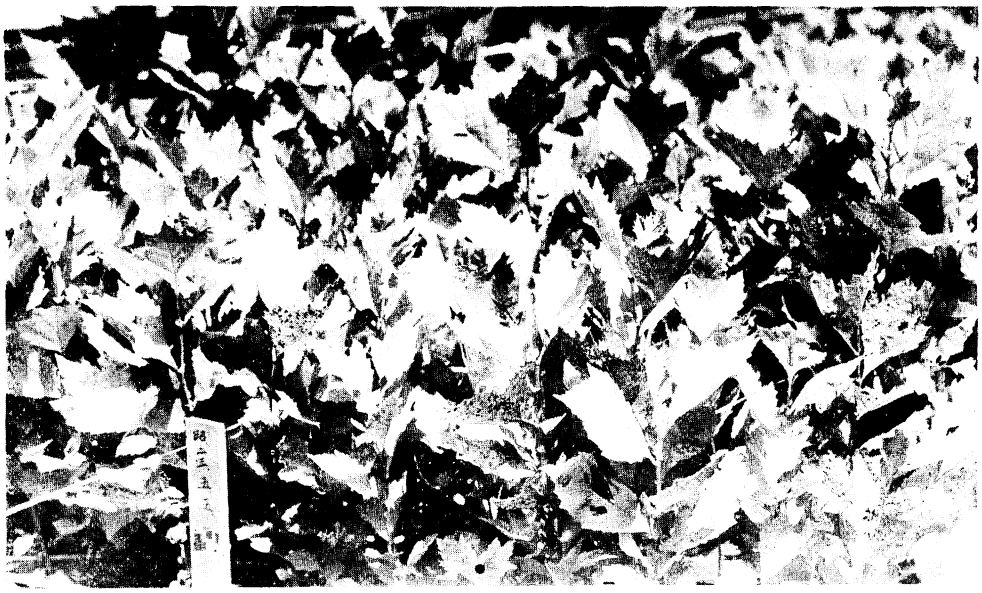
由来 *Cercospora* 属菌の分生胞子を培養基上で生成させ，かつこの状態を永く保持することは一般に困難とされているものである。著者等は「継続移植法」により培養基上に常に多量の胞子を生成させ，かつこの状態を 10 ヶ月以上保持することが出来た。

なお継続移植 25 回（約 10 日間隔）の分生胞子は自然のものに比べて濃色，空胞多く，しばしば畸形を呈するが，接種試験の結果病原性はいささかも減退していないことを確めた。

Plate I

Bull. Gov. Forest Exp. Sta. No. 46.

A



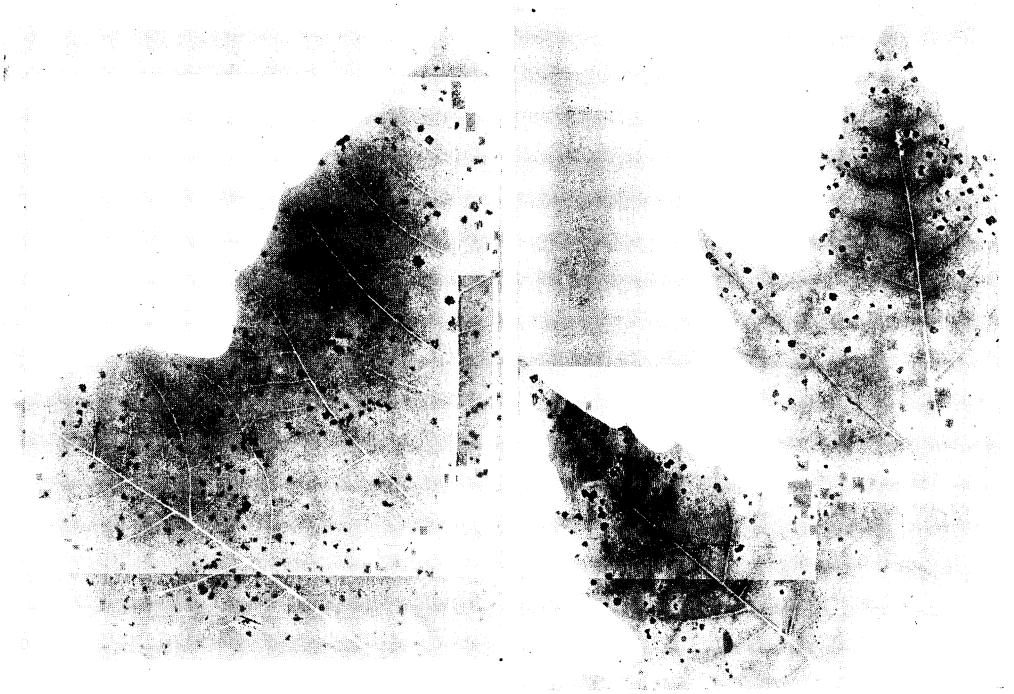
B

ITÔ, K., & HOSAKA, Y.: Leaf-spot disease — I.

Plate II

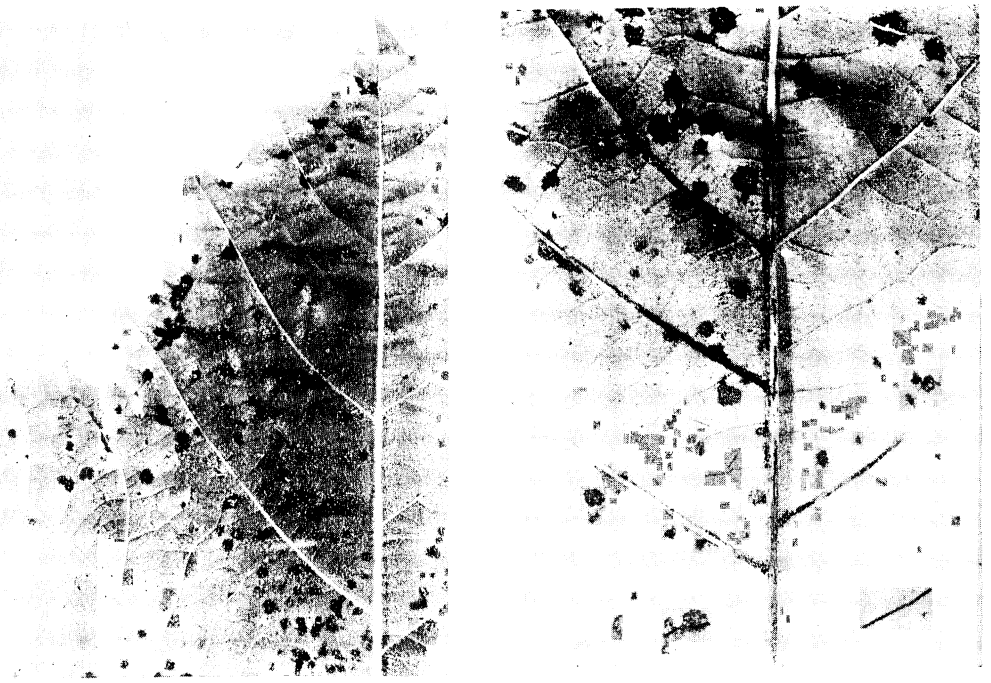
A

B



C

D

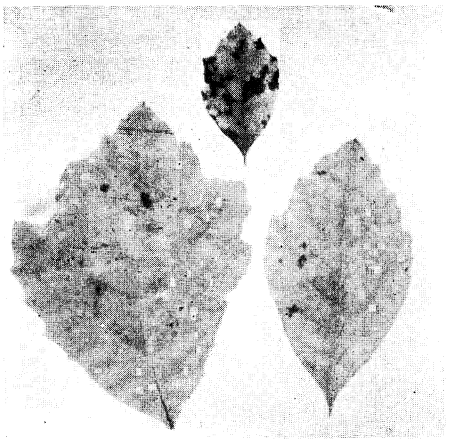


ITÔ, K., & HOSAKA, Y.: Leaf-spot disease — I.

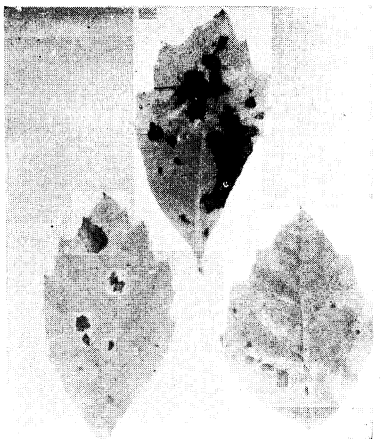
Plate III

Bull. Gov. Forest Exp. Sta. No. 46.

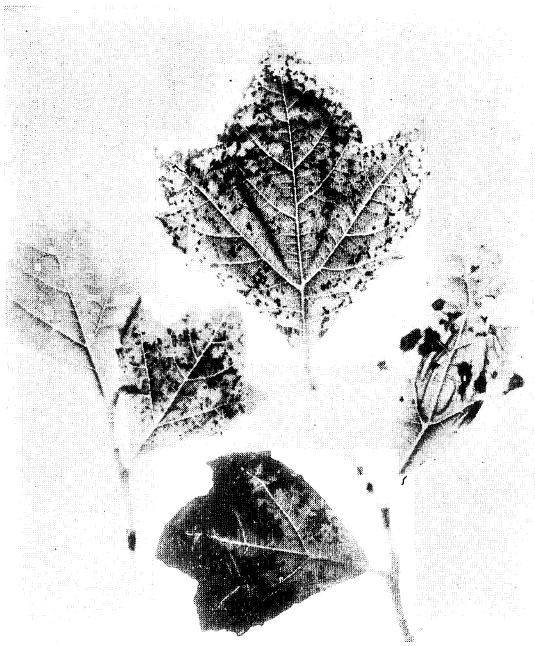
A



B

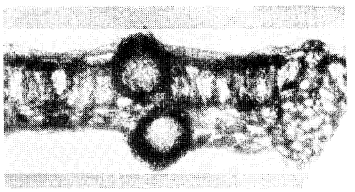


C

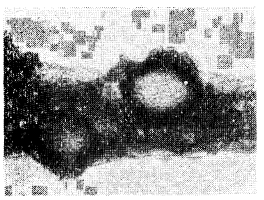


D

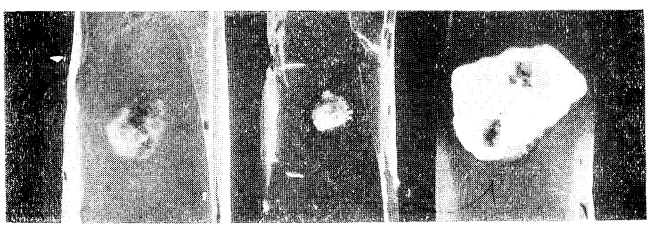
E



F

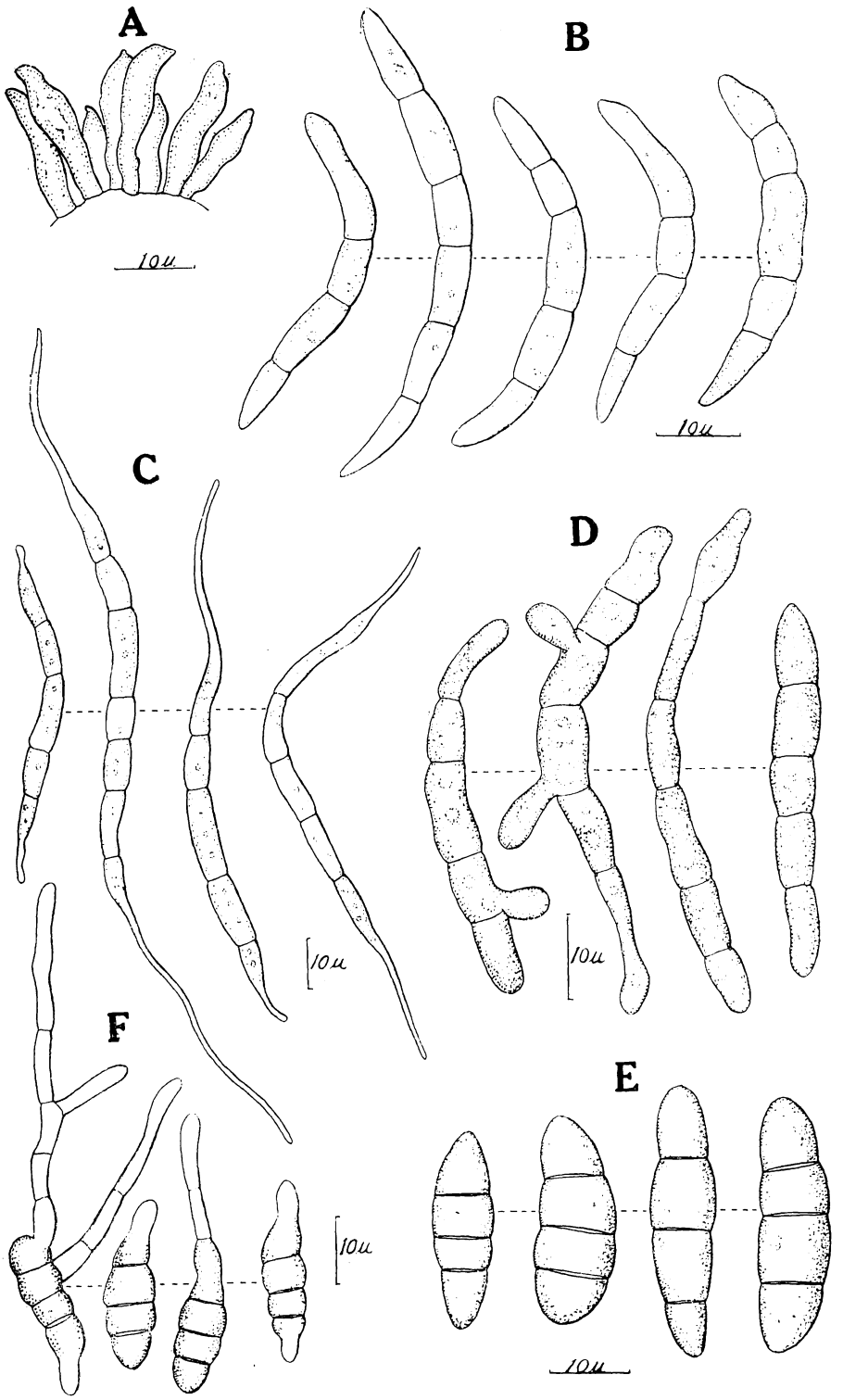


G



ITÒ, K., & HOSAKA, Y.: Leaf-spot disease — I.

Plate IV　　Bull. Gov. Forest Exp. Sta. No. 46.



ITÔ, K, &. HOSAKA, Y.: Leaf-spot disease — I.