Dothistroma Needle Blight of Pines in Japan

By

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Summary : Since 1952, the *Dothistroma* needle blight of pines has been found in several parts of Honsyu and Hokkaido of Japan. The causal fungus was morphologically identical with *Dothistroma pini* HULBARY var. *pini*. Host plants of the fungus hitherto collected in Japan were as follows : *Pinus densiflora*, *P. thunbergii*, *P. elliottii* var. *elliottii* (*P. caribaea*), *P. montana*, *P. jeffereyi*, *P. ponderosa*, and *P. contorta*. In artificial inoculation with the fungus, the infection occurred more severely on pine needles wounded slightly than on those unwounded. Artificial inoculations with the fungus isolated from *Pinus thunbergii* were made to the following pine species : *Pinus densiflora*, *P. thunbergii*, *P. taeda*, *P. elliottii* var. *elliottii*, *P. pinaster*, and *P. radiata*. Results showed that all the species tested were equally susceptible, and the incubation period of the disease was $2\sim6$ months. In Japan, the disease has been generally considered to be a minor obstacle to forest trees, because its damage to the native pine species is still not serious.

Introduction

A needle spotting fungus, *Dothistroma pini* Hulbary (1941)²²⁾, was first described on *Pinus* nigra var. austriaca collected at DeKalb county, Illinois, U. S. A. The distribution of the fungus is now world-wide, and it has been recovered from Canada (PARKER & Collins 1966³³⁾), Chile (Dubin & Staley 1966⁶⁾), Brazil (Figueiredo & NAMEKATA 1969⁸⁾), Argentina (Fresa 1968⁹⁾), Uruguay (Peterson 1969³⁶⁾), Kenya (Gibson 1964¹³⁾, Christensen & Gibson 1964³⁾, Gibson et al. 1964¹⁴⁾, Shaw 1964³⁹⁾), Tanganyika (Gill 1963¹⁶⁾), Tanzania (Etheridge 1965⁷⁾, Hocking 1966²⁰⁾, Hocking & Etheridge 1967²¹⁾, Griffin 1968¹⁹), Yugoslavia (Krstic 1958²⁹), Britain (Murray & Batko 1962³²⁾, Shaw 1964³⁹⁾), France (Morelet 1967³⁰⁾), Roumania (Gremmen 1968¹⁸⁾), New Zealand (Gilmour & Noorderhaven 1966¹⁶⁾, Gilmour 1967¹⁷⁾, Gadcil 1967¹¹⁾), and India (Bakshi & Singh 1968¹⁾, Bakshi et al. 1973²⁾).

The pine disease caused by *Dothistroma pini* commonly goes under the name of *Dothistroma* needle blight or red band needle blight, and it causes serious losses of *Pinus radiata*, an exotic pine species, in East Africa, New Zealand, and South America. The disease continues to be a major obstacle to the satisfactory plantation of radiata pine in these countries.

Since 1952, the present authors have frequently observed a foliage disease of several pine

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species, and they have considered that a fungus associated with this disease might be a member of the genus *Dothistroma* or the allied genera.

Fortunately, examinations were made of HULBARY'S type specimen of *Dothistroma pini* and other specimens of the same fungus on various pine species kindly sent by foreign mycologists and pathologists. Results showed that the fungus found by the authors in Japan was quite identical with *Dothistroma pini* HULBARY.

In Japan, the distribution of the fungus is still limited to several parts of Honshu and Hokkaido, where the disease occurs usually sporadically and rarely in epidemic form.

The *Dothistroma* needle blight of pines has been considered to be one of the most important destructive tree diseases internationally. In this paper, the authors deal with the present status of the disease in Japan, with special references to morphologic characters of the causal fungus, its pathogenicity to the native pine species, and damage of the disease. An outline of the study was already published in the preliminary reports (Irô & ZINNO 1972²³⁾, Irô 1973²⁴⁾, Surô 1974⁴¹⁾).

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Symptoms and signs of the disease

The first obvious symptom of the disease is the appearance of small chlorotic areas on infected needles during the autumn, followed by large areas of necrosis. Small resin drops are frequently produced on necrotic parts. In the spring, bright red bands, $1\sim3$ mm in length, are apparent in the necrotic areas, and shortly thereafter small black bodies, stromata of the causal fungus, rupture the dead epidermis. Infected needles are usually cast in the summer and autumn. Needles of all ages are susceptible, although the older needles are usually more severely infected (Plates 1, 3, 4).

Tamage of the disease

In November, 1960, a severe attack by the disease first called the authors' attention in a young natural stand of *Pinus densiflora* at Narusawa, Yamanashi Prefecture. In the next year, a 3 ha 10-year-old plantation of *P. densiflora*, having a total of about 1,500 trees, was found to be badly spotted and severely defoliated by the disease. Another severe damage of the disease was found in 1962, in a 3.3 ha 7-year-old stand of the same pine species, totalling about 1,200 trees, at Asahi, Gifu Prefecture.

Since about 1965, the disease has commonly occurred on *Pinus thunbergii* planted in gardens and parks in Oki Islands and the eastern parts of Shimane Prefecture.

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Morphology and geographic distribution of the causal fungus

Morphologic characters of the fungus

Stromata linear, solitary or gregarious, subepidermal, erumpent, dark brown to black, 100 $\sim 500 \mu$ in width, $110 \sim 300 \mu$ in height. Locules separate, one to several in the upper part of the stroma, without a distinct wall, the entire inner face sporiferous. Conidiophores numerous, simple, arising directly from the stromatic hyphae, unbranched, hyaline $8 \sim 10 \times 1.5 \sim 2 \mu$, producing conidia at their tips. Conidia hyaline, scoleciform, 1- to 4- but usually 3 septate, blunt at the ends, straight, slightly curved, or bent, $12 \sim 36 \times 1.5 \sim 3.5 \mu$ (Plate 5, Figs. 1, 2).

On hosts, conidia of the fungus are usually found in April to July, and abundantly in May to June.

Dimensions of the fungus in the conidial stage collected in Japan as well as in some foreign countries measured by the present authors are given in Tables $1\sim3$ (Plates 5, 6, Figs. 2, 3).

Three varieties of the fungus have been distinguished by the differences in the conidial dimensions and in the stromata as follws:

> Dothistroma pini HULBARY (1941) var. pini, Dothistroma pini HULBARY var. linearis THYR & SHAW (1964)⁴²⁾, and Dothistroma pini HULBARY var. keniensis IVORY (1967)²⁵⁾.

The fungus in Japan is quite identical with *Dothistroma pini* HULBARY var. *pini* on the basis of conidial size, though there may be some differences in conidial and stromatical characters by either climatological factors or



Fig. 1. A part of a stroma of *Dothistroma* pini on Pinus densiflora collected in Japan $(1-2) = 20\mu$

Pine species	Locality	Size of	stroma	Size of con	Number of	
r me species	Locality	Height	Width	Range	Averaged	septum of a conidium
io of the working	THE PART SOLUTION STOLEN	μ	μ	μ	μ	0.1444606.00
P. densiflora	Narusawa, Yamanashi	130~260	130~430	22~34×2~2.5	27.6×2.2	1~4
do.	do.	110~200	130~500*	18~33×2.5	24.3×2.5	1~3
do.	Komoro, Nagano	160~230	200~390	17~29×1.5~2.3	21.2×1.6	2~3
do.	do.	120~260	120~330	_		
do.	Asahi, Gifu			17~29×1.5~2.3	21.2×1.6	2~3
P. montana	Yamabe, Hokkaido	150~280	200~450*	13~25×2~2.5	19.7×2.2	1~3
P. jeffereyi	do.	190~300	210~480*	15~33×2~3	22.7×2.4	1~3
P. ponderosa	do.	180~230	200~400*	15~30×1.5~2.5	20.0×1.9	1~3
P. contorta	do.	140~190	100~500*	13~25×1.5~2.3	20.5×1.7	1~3

Table 1. Dimension of Dothistroma pini collected in Japan

Note : * Some stromata continued

T 114	Size of con	Number of septum		
Locality	Range	Averaged	of a conidium	
Matsue City	$12 \sim 27 \times 2.0 \sim 3.0^{\mu}$	23×2.3	0~3	
do.	8~36×2.0~3.5	20×2.4	0010000~3	
Yoshida V.	14~34×1.5~3.5	24×2.1	s tuoliti $1\sim3$ and an	
Fuse, Oki Isl.	15~32×2.0~3.0	23×2.3	0~3	
Ota City	12~36×2.0~3.0	25×2.1	0~3	

 Table 2.
 Size of conidia of Dothistroma pini on Pinus thunbergii

 collected in Shimane Pref. Japan
 collected in Shimane Pref. Japan

 Table 3. Dimension of Dothistroma pini collected in several foreign countries

D	T 114	Size of	stroma	Size of	conidium 👘	Number of
Pine species	Locality	Height	Width	Length	Width	a conidium
ANA REPORT		μ	μ	μ	1	ı
P. nigra*	Waterman, DeKalb County, U. S. A.	180~400	260~750 **	20~30	2.5	1~3
P. contorta	Sooke, B. C. Canada	120~230	100~290**	20~38	2.5	1~3
do.	Kispiox, B. C. Canada	100~160	110~270**	20~35	2~2.8	1~3
P. radiata***	Ucluelet, B. C. Canada	50~130	80~320**	35~70	1.8~2.5	2~6
do.	Tokoroa, N. Island, New Zealand	80~150	100~250	18~38	1.8~2.5	1~3
P. muricata	Humboldt County, Calif. U. S. A.	110~230	120~500**	40~58	1.8~2.3	1~5

Note: * Type specimen

** Some stromata continued

*** Dothistroma pini var. linearis

different substrata.

Host plants affected naturally by the fungus in Japan are as follows: Pinus densiflora, P. thunbergii, P. elliottii var. elliottii (P. caribaea), P. montana, P. jeffereyi, P. ponderosa, and P. contorta.

Distribution of the fungus in Japan

No opportunity has been afforded to make a systematic survey of the pine growing region of Japan to determine the distribution of the fungus; however, studies to date show it to be limited to the following prefectures: Hokkaido, Yamanashi, Nagano, Gifu, Fukui, Hiroshima, and Shimane (Fig. 4). 日本におけるマツ赤斑葉枯病の研究(伊藤・陳野・周藤)



Fig. 3. Conidia of *Dothistroma pini* collected in foreign countries $(1-1)^{\mu}=10\mu$

- a. on Pinus contorta collected in Canada.
- b. on Pinus contorta collected in Canada.
- c. on Pinus radiata collected in Canada.
- d. on Pinus radiata collected in New Zealand.

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Fig. 4. Map of distribution of Dothistroma pini in Japan

1. Hokkaido 2. Yamanashi Pref. 3. Nagano Pref.

4. Gifu Pref. 5. Fukui Pref. 6. Hiroshima Pref.

7. Shimane Pref.

Overwintering of the causal fungus

In early November, 1960, the diseased needles of *Pinus densiflora* collected at Narusawa, Yamanashi Prefecture were placed in wire baskets out of doors in Tokyo. Every two weeks, some needles were brought into the laboratory, sectioned and examined under the microscope for the presence of new and old fruit-bodies of the fungus.

In about mid-March of the following year, young conidia of the fungus were produced on old stromata, and the conidia were formed in great abundance in late April.

Diseased needles of Pinus thunbergii gathered in Matsue, Shimane Prefecture were placed

out of doors in early December in Tokyo. On second-year needles as well as current-year needles, fresh young conidia of the fungus were found on stromata as early as the first week of March of the following year. Conidia of the fungus were abundantly produced from mid-April to mid-May, and disappered at the end of June.

In New Zealand, GADGIL (1970)¹²⁾ reported that stromata of *Dothistroma pini* on some needles of *Pinus radiata* exposed over the winter months as well as those on needles exposed over the summer months showed a similar survival period, e. g. needles laid out in May and December each showed a survival period of $4\sim 6$ months.

The authors' observations in Japan showed that stromata of the fungus on needles laying on the ground from winter to spring produced viable conidia after 6 months' exposure.

The ascigerous stage of the fungus was discovered in Canada, and was named as *Scirrhia pini* FUNK et PARKER (1966)¹⁰⁾. In the next year, the existence of this stage was reported from France by MoreLet (1967)³¹⁾.

In Japan, no perfect stage has been found for *Dothistroma pini*, and the dispersal of the fungus appears to be entirely by conidia.

Germination of conidia of the causal fungus

Conidia of the fungus (C-1)* which had been produced on SAITO'S SOY agar were used for germination tests. Conidial suspensions were smeared on the surface of water agar plate (pH 5.4) in Petri dishes. Besides the readings on germination, the lengths of the germ tubes were measured.

Relation between germination and time passed

Results obtained at 20°C are given in Table 4. As shown in Table 4, the conidia begin to germinate within about 16 hours, and germination is over 90 per cent in 24 hours.

Usually, a germ tube grows from the end of each terminal cell, and later from the intermediate cells. In 32 hours, they become septate and branch (Plate 7, Fig. 5).

Relation between germination and temperature

Results of the experiments obtained at the end of 24 hours are presented in Table 5.

From Table 5, it may be said that the range of temperature within which germination takes place is from 10 to 30°C, with an optimum at 20°C.

Relation between germination and H-ion concentration

A range of pH value was obtained by additions of regulated amounts of HCl or NaOH. Results of the experiments at the end of 24 hours at 20°C are briefly noted in Table 6.



^{*} Host : Pinus thunbergii Locality : Matsue, Shimane Pref. Isolation : March 26, 1968

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	Expe	riment-1	Experiment-2			
Time lapsed -	Germination	Maximum length of germ-tube	Germination	Maximum length of germ-tube		
hrs.	%	μ	%	μ		
8	0	Partian ep en rusin to	0	direct structures, conselve		
16	80.8	83	78.8	66		
24	90.7	125	92.1	132		
32	91.2	188	94.1	198		

Table 4. Relation between lapsed time and germination of conidia of *Dothistroma pini* on water agar at 20°C

 Table 5. Effect of temperatures on germination of conidia of

 Dothistroma pini.
 After 24 hours on water agar

- m	Exper	riment-1	Experiment-2			
Temperature	Germination percentage	Maximum length of germ-tube	Germination	Maximum length of germ-tube		
°C	%	μ	%	μ		
3	0		0			
10	3.3	8 8	5.2	8		
15	73.4	89	87.6	99		
20	90.2	109	92.1	149		
25	83.1	79	89.9	83		
30	11.8	46	16.9	30		
35	0	and many and many income	0	INCL. INTERNAL DISC. OF STATE		

Table 6. Effect of H-ion concentrations on germination of conidia of Dothistroma pini. After 24 hours at 20°C

	pH 00 m	2.8	4.0	4.6	6.0	6.8	8.4	9.0
Max	Germination percentage %	4	70	87	94	92	90	88
	Maximum length of germ-tube μ	12	41	68	102	73	56	54
Experiment-2	pH	3.0	4.0	4.6	5.6	6.8	8.2	9.0
	Germination percentage %	6	89	98	99	95	93	91
	Maximum length of germ-tube μ	10	49	80	90	68	54	41

esuits of the experiments obtained at the and

From Table 6, it is to be inferred that germination of conidia is not strikingly effected by the change of H-ion concentration excepting an extreme acid side.

IVORY $(1967)^{26}$ reported that conidial germination of *Dothistroma pini* var. *keniensis* in solution occurred within the temperature range $8\sim25$ °C and the pH range 2.2~5.5, with optimum of 18 °C and pH 3.5, respectively. Peterson $(1967)^{25}$ who studied the fungus on Austrian and ponderosa pines in U. S. A., stated that the conidia germinated over a wide temperature range $(12\sim28$ °C). Sheridan & Yen $(1970)^{40}$, on a New Zealand isolate of the fungus, noted that the conidia germinated at $5\sim30$ °C with the optimum at 17 °C. These characters of the fungus in germination are very similar to those of the fungus in Japan.

Effect of temperatures on the mycelial growth of the causal fungus

The relation of temperature to the growth of the fungus was tested by slant culture method using SAITO'S soy agar (pH 5.4). For inocula, bits of the colonies originated from conidia were transplanted to the center of each agar in test tube. Diameter and height of the colonies at each temperature averaged after 30 days are given in Table 7.

It is known from Table 7 that the fungus grows favorably at the temperatures ranging from 10 to 25°C with an optimum at 20°C, and the maximum and minimum temperatures for the growth are $3\sim5$ °C and 30°C, respectively.

According to Ivorv's $(1967)^{26}$ paper on *Dothistroma pini* var. *keniensis*, growth of the mycelium in liquid culture was very slow, and occurred within the temperature range $6\sim 28$ °C, with the optimum of about 15 °C. The optimal temperature for the mycelial growth of Japanese isolate is somewhat higher than that of Kenyan isolate.

	Tamanak		1	Mycelia	l colony		Conidial formation	
	Temperature		Diameter		Height		Comular formation	
ot ai line	3~5	°C	3	mm	2	mm	territor bio salo contra los	
	10		10		5		me (relate / <u>- c</u>).	
	150 15 8	no bu	13		au gand 5 7		ano 8 944 ni avoie	
	20		20		8		none thee+++ abund	
	25	i sala	7		5		+	
	30	a Strug	3		2		Supra Supra	
	35		0		0		orden) 170 re manual	

Table 7. Effect of temperatures on mycelial growth of Dothistroma pini. After 30 days on SAITO'S soy agar

Mycelial growth of the causal fungus on various agar media

Three isolates from conidia were cultured on potato sucrose agar, and small pieces $(1 \sim 1.5 \text{ mm in diameter})$ of the colony were used as inocula. The fungus was cultured on the following agar media: Potato sucrose agar, pine-needle decoction agar, SAITO'S soy agar, WAKSMAN'S solution agar, RICHARDS' solution agar, and CZAPEK'S solution agar.

On potato sucrose agar, cultures grow slowly. Colonies sometimes spread with appressed growth, usually hemispherical and stromatoid. Mycelial mats are white at first, soon turning dark and pink in patches.

In macroscopic appearances of the colonies on various agar media, there are no remarkable differences among the isolates.

Diameter and height of colonies of the fungus on various agar media were measured after 30 days at 20°C.

As presented in Table 8, the fungus grows well on SAITO'S soy agar, potato sucrose agar, pine-needle decoction agar, WAKSMAN'S solution agar, and RICHARDS' solution agar, but sparsely on CZAPEK'S solution agar (Plate 8).

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Stock No.		C-1*1		C) 9556	C14-1*2			C14-2*8	
at by stor culture	Mycelia	l colony	Degree	Mycelia	l colony	Degree	Mycelia	l colony	Degree of
Agar medium	Diame- ter	Height	conidial forma- tion	Diame- ter	Height	conidial forma- tion	Diame- ter	Height	conidial forma- tion
	mm	mm		mm	mm		mm	mm	
Potato-sucrose agar	14	4	+	15	4	05101610	12	6	1996 <u>0</u> , 011
Pine-needle decoction agar*4	14	1	++	18	5	<u>de 7 eb</u>	13	4	ak <u>11.</u>
SAITO'S SOY agar	20	8	+++	25	5	and the state	16	10	01_0101
WAKSMAN'S sol. agar	12	4	++	20	5	(0° <u>02</u> 6)	15	5	101+ 50
RICHARDS' sol. agar	15	5	+	15	5	(es an	13	3	mail
CZAPEK'S sol. agar	10	4	+	3	2	-	12	3	

Table 8. Mycelial colonies of *Dothistroma pini* on various agar-media. After 30 days at 20°C

Notes : *1 Host : Pinus thunbergii Loc. : Matsue, Shimane Pref. Isol. : March 26, 1968

*2 Host : Pinus densiflora Loc. : Sakamachi, Hiroshima Pref. Isol. : July 10, 1958

*3 Host : Pinus densiflora Loc. : Narusawa, Yamanashi Pref. Isol. : Nov. 24, 1960

*4 Pine needles 100g, sucrose 20g, dist. water 1l, agar-agar 20g

Sporulation of the causal fungus on agar media

Conidia of the fungus were abundantly produced on the young culture, but none of them were found on the old cultures which had been transplanted repeatedly on agar media for a long time (Plate 7, C).

As shown in Table 8, conidia of the fungus (C-1) are produced on all agar media used, and, among them, very abundantly on SAITO'S soy agar (Fig. 6).

On SAITO'S soy agar, conidia are produced at temperatures ranging from 15 to 25° C, with the optimum at 20° C (Table 7).

Relation between sporulation and H-ion concentration was tested on water agar medium regulated by HCl or NaOH at various pH values. Results obtained after 5 days at 20°C showed that conidial production was good at pH $2.8 \sim 4.6$, but did not occur at alkaline side (pH $8.4 \sim 9.0$) (Table 9).

RACK and BUTIN (1973)88) reported that an earlier and more intensified conidia production



Fig. 6. Conidia of *Dothistroma pini* produced on agar media $(1-1)=10\mu$) a. on potato sucrose agar b. on SAITO's soy agar

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$_{\rm pH}$	2.8	4.0	4.6	6.0	6.8	8.4	9.0
Degree of conidial formation	+++	+++	+++	++	+	_	0.4

Table 9. Effect of H-ion concentrations on conidial formation of *Dothistroma pini*. After 5 days at 20°C

might be obtained in the case of *Dothistroma pini* by the following culture condition; Malt extract agar 2% with pH 5.5 as culture medium, high inoculation density $(50\sim100 \text{ spores per mm}^2)$, temperature $20 \pm 1^{\circ}$ C and collection of spores after a culture period of $10\sim12$ days.

The authors' detailed account on sporulation of the fungus will be published in the future.

Pathogenicity of the fungus

Inoculation experiments with the fungus to Pinus densiflora and P. thunbergii

The fungus $(C-1)^*$ was inoculated to potted *Pinus densiflora-* and *P. thunbergii*-seedlings. The inoculum was prepared by breaking the cultured mycelium in sterile distilled water, then filtering its fragments and conidia through double sheets of cotton cloth. On June 17, 1968, the fungous suspension was sprayed on needles of 2-year-old seedlings wounded slightly by fine sand paper or unwounded ones. Check plants were similarly treated except that they were atomized with sterile water instead of the fungous suspension. The inoculated and check plants were kept in moist condition by covering with polyethylene bags for 4 days at $19\sim 22^{\circ}C$.

The first appearance of the symptom was observed on the inoculated wounded needles in November, 1968, while no symptom appeared on the unwounded needles as well as on the checks. In January, 1969, many brown lesions with minute black bodies were formed on the inoculated needles. In March, the small black fruit bodies of the causal fungus ruptured the dead epidermis, and abundant conidia were produced.

Results obtained at the end of the experiment on May 1, 1969 are summarized in Table 10.

Pine species	Inoculation or check	Treatment	Number of seedlings tested	Degree of infection
Pinus densiflora	Inoculation	unwounded wounded		+~+++
	Check	unwounded wounded	5	ensiflere—2. Humis Results of the
n susceptibility in svene in wounder	Inoculation	unwounded wounded	5	++~+++
Pinus thunbergii	Check	unwounded wounded	5	eedies: A wi nt more

Table 10. Result of the inoculation experiments with Dothistroma pini to Pinus densiflora and P. thunbergii. June 17, 1968~May 1, 1969

* Host : Pinus thunbergii Locality : Matsue, Shimane Pref. Isolation : March 26, 1968

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As shown in Table 10, *Pinus densiflora* and *P. thunbergii* are equally attacked by the fungus, and the infection takes place more severely on wounded seedlings than on unwounded ones (Plate 2, A, B).

Inoculation experiments with the fungus to several conifers

In October 1, 1970, the inoculations with the fungus were carried out by the same procedures as in the previous experiments to the following tree species: *Pinus densiflora*, *P. thunbergii*, *Cryptomeria japonica*, and *Chamaecyparis obtusa*.

As early as in December, bright brown lesions appeared on the tip of inoculated needles of pines. In March and April, 1971, lesions with small black fruit bodies of the fungus were found abundantly on inoculated pines, while no lesion was produced on each of *Cryptomeria japonica* and *Chamaecyparis obtusa* (Table 11).

Tree species	Inoculation or check	Treatment	Number of seedlings tested	Degree of infection
Pinus dansi fong	Inoculation	unwounded wounded	5	+ +~++
Pinus densiflora	Check	unwounded `wounded	5 5	and paper or
li in south i not	Inoculation	unwounded wounded	1 not three 5 telever at 5	+~++
Pinus thunbergii	Check	unwounded wounded	5	he h at appe
Cryptomeria	Inoculation	unwounded wounded	5 1991 - 5	ss. H Jaouan Inted needlos
japonica	Check	unwounded wounded	5	epid <mark>-mis</mark> , an leants obtain
Chamaecyparis	Inoculation	unwounded wounded	5 di lo 11 <mark>05,61</mark> ,01 al	
obtusa	Check	unwounded wounded	5	_

Table 11. Result of the inoculation experiments with Dothistromapini to several conifers. October 1, 1970~May 1, 1971

Inoculation experiments with the fungus to several species of Pinus

bads anti-

In June 17, 1971, the inoculation experiments were made by the same procedures as in the previous experiments with conidial suspensions as inocula to the following pine species: *Pinus densi flora*, *P. thunbergii*, *P. taeda*, *P. elliottii* var. elliottii (*P. caribaea*), *P. pinaster*, and *P. radiata*.

Results of the experiments tested on May 10, 1972 are presented in Table 12 (Plate 2, C).

It is evident from Table 12 that there are no remarkable differences in susceptibility to the disease among the pine species tested, and the infection is strikingly severe in wounded needles.

A wide range of species of pine has been found to be susceptible to the disease, but none is so severely attacked as *Pinus radiata* (Peterson 1969³⁶⁾, GIBSON 1964¹³⁾, BAKSHI & SINGH 1968¹⁾, COBB & LIBBY 1968⁴⁾, COBB & MILLER 1968⁵⁾, etc.).

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日本におけるマツ赤斑葉枯病の研究(伊藤・陳野・周藤)

Pine species	Inoculation or check	Treatment	Number of seedlings tested	Degree of infection
	Inoculation	unwounded wounded	4 5	+ +~+++
Pinus densiflora –	Check	unwounded wounded		aibal ai <u>k</u> noù
D	Inoculation	unwounded wounded	5 5	(hogo + soln + + +
Pinus thunbergii	Check	unwounded wounded	3	pines caused
the state of the state of the	Inoculation	unwounded wounded	5	+~++ +++
Pinus taeda	Check	unwounded wounded	2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2	Phy on path., S Conc. 8, W
Pinus elliottii var	Inoculation	unwounded wounded	5 *	++~+++
elliottii	Check	unwounded wounded		50, ¹ 55 0, (1966) Printman, D
minur de leave	Inoculation	unwounded wounded	a roll worf 5 Jeise Ar	1200 + d Teel
Pinus pinaster	Check	unwounded wounded	2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2	aolo <mark>nias em 7 (1965),</mark>
i waris Tave de Sux	Inoculation	unwounded wounded		++++
Pinus radiata –	Check	unwounded wounded	3 2 2) F <u>rre</u> , A., a Hus na r, Cau

Table 12. Result of the inoculation experiments with *Dothistroma pini* to several kinds of *Pinus*. June 17, 1971~May 10, 1972

Note : * killed by white grubs.

So far as the present authors' experiments are concerned, *Pinus radiata* is not more susceptible to the disease than the native species, *P. densiflora* and *P. thunbergii*.

JANCARIK (1969)²⁸⁾ found that the incubation period of the disease varied in New Zealand between 5 weeks and 6 months, depending on the time of year when infection took place. Ivory (1972)²⁷⁾ noted that the incubation period was determined as $32\sim107$ days, the length being apparently related to the climate because the shortest period always coincided with warmest months of the year. PARKER (1972)³⁴⁾, studying effect of humidity and temperature on incubation of the disease, reported that the minimum incubation period in the temperature range $13\sim21^{\circ}$ C might be expressed either as 4 weeks at the 73~100% in relative humidity, or as 23 days at 100% RH.

PETERSON (1967)⁸⁵ stated that symptoms of the disease on Austrian and ponderosa pines were first evident 4 months after first infection in eastern Nebraska, U.S.A. He (1973)⁸⁷ has recently noted that the time between initial infection and first appearance of symptom varies

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from 11 to 16 weeks.

The authors' artificial inoculations show that the incubation period of the disease in Japan may be 2 to 6 months.

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Explanation of Plates

Plate 1

A. Pinus thunbergii attacked by Dothistroma pini, Kitsugi, Shimane Prefecture.

B. Needles of Pinus thunbergii attacked by Dothistroma pini. $\times 1.2$

C. Enlargement of the red band lesion on a needle of Pinus thunbergii. \times 50

Plate 2

Result of the inoculation experiments with *Dothistroma pini* to three species of *Pinus*. A. *Pinus densiflora*. B. *Pinus thunbergii*. C. *Pinus taeda*.

Plate 3

Diseased needles of pine trees caused by Dothistroma pini.

A, B. Pinus densiflora. $\times 2$

C. Pinus thunbergii. $\times 2.5$

Plate 4

Diseased needles of pine trees caused by *Dothistroma pini* collected in Japan. A. *Pinus montana.* $\times 2$ B. *Pinus jeffereyi.* $\times 2$ C. *Pinus contorta.* $\times 2$

Plate 5

Stromata of Dothistroma pini collected in Japan.

A, B. on Pinus densiflora. \times 210

C. on Pinus thunbergii. \times 250

Plate 6

Stromata of Dothistroma pini collected in foreign countries.

A. on *Pinus contorta*, Canada. \times 230

B. on *Pinus radiata*, Canada. \times 230

C. on Pinus radiata, New Zealand. \times 320

Plate 7

A. Germinating conidia of Dothistroma pini, after 32 hours at 20°C. × 400

B. A germinating conidium of Dothistroma pini, after 3 days at 20°C. × 400

C. Conidia of Dothistroma pini produced on potato sucrose agar. × 310

Plate 8

- Mycelial colonies of *Dothistroma pini* on various agar-media, after 30 days at 20°C. From left to right: Potato sucrose agar, pine-needle decoction agar, SAITO's soy agar, WAKSMAN's solution agar, RICHARDS' solution agar, and CZAPEK's solution agar.
- A. C-1. Host: Pinus thunbergii Loc.: Matsue, Shimane Pref.
- Isolation: March 26, 1968.
- B. C₁₄-1. Host: *Pinus densi flora* Loc.: Sakamachi, Hiroshima Pref. Isolation: July 10, 1958.
- C. C₁₄-2. Host: *Pinus densi flora* Loc.: Narusawa, Yamanashi Pref. Isolation: November 24, 1960.

(ドシストロマ葉枯病)の研究

伊藤一雄心•陳野好之心•周藤靖雄3

do: 9. ideanaore Y. . owo_摘 to 2. 要

本病病原菌 Dothistroma pini HULBARY (1941) はアメリカ合衆国イリノイ州においてオーストリアマツ(ヨーロッパクロマツ) (Pinus nigra var. austriaca) 上で発見・記載されたものであるが、その後他の多くのマツ類にも寄生することが知られ、大平洋岸北西部を中心に、かなり広域に分布し、その被害も軽視し得ないことが明らかにされている。

一方,十数年前から東部アフリカのケニア,タンガニカおよびタンザニアにおいて,これはとくに導入 種であるラジアタマツ(*Pinus radiata*)に激しい被害を与え,その成林を左右する最も重要な疾病と認め られている。なお,本病はニュージーランド,南米アルゼンチン,ブラジルなどでも、やはりラジアタマ ツに激害を及ぼしている。そして,アフリカ,ニュージーランド,南米諸国ではいずれも,その病原菌は 北アメリカから侵入したものと信じられている。

今や本病病原菌の分布はアメリカ合衆国,カナダ,ブラジル,チリ,アルゼンチン,ウルグワイ,ケニア,タンガニカ,タンザニア,ユーゴスラビア,イギリス,フランス,ルーマニア,インドおよびニュージーランドと全世界的規模にわたり,その寄主として三十数種のマツ類があげられ,本病は国際的に危険な重要林木疾病の一つとして広く知られている。

本病がわが国で著者らによって発見されたのは1952年(昭和27年)のことで、その後局所的には少な からぬ被害が発生してはいるものの、全般的に見て現在のところ重要疾病とは認められない。しかし、諸 外国における過去の実例から見て、将来わが国でも導入種マッ類に対して著しい被害をもたらす可能性が あり、また学術国際協力の立場からも、本病のわが国における現況および著者らが行なった研究成果の概 要を報告することは有意義だと考えられる。

本病病原菌はこれまで北海道,山梨,長野,岐阜,福井,広島および島根の各県に見い出されており, その寄主としてはアカマツ,クロマツ,スラッシュマツ (*Pinus elliottii* var. *elliottii*),モンタナマツ (*P. montana*),ジェフリーマツ (*P. jeffereyi*),ポンデローザマツ (*P. ponderosa*) およびコントルタ マツ (*P. contorta*) が知られている。

本病はかつて山梨・長野両県において小面積ながらアカマツ造林木に大発生して,はなはだしい落葉を 起こしたことがあり,また島根県ではクロマツ庭園樹に著しい被害を与えている。

本病病原菌には Dothistroma pini var. pini, D. pini var. linearis および D. pini var. keniensis の 3 変種があるとされているが, わが国産の菌は D. pini var. pini に一致する。そして, この分生胞子の 発芽, 菌糸の成長および分生胞子の形成には比較的低温の約 20°C を適温とする。

アカマツ,クロマツ,テーダマツ (P. taeda),スラッシュマツ,フランスカイガンショウ (P. pinaster) およびラジアタマツに対する人工接種試験結果は、いずれも感受性で、この実験に関する限り、ラジアタ マツがとくに感受性が高い傾向は認められなかった。そして、接種前に針葉に軽く付傷すれば、無傷の場 合よりも罹病程度がはなはだしくなることが明らかにされ、また本病の潜伏期は2~6か月であることが 知られた。 日本におけるマツ赤斑葉枯病の研究(伊藤・陳野・周藤) --Plate 1-





日本におけるマツ赤斑葉枯病の研究(伊藤・陳野・周藤)

-Plate 3-







日本におけるマツ赤斑葉枯病の研究(伊藤・陳野・周藤) -Plate 5-





日本におけるマツ赤斑葉枯病の研究(伊藤・陳野・周藤) --Plate 7-

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