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**Disease of poplar caused by *Ceratocystis fimbriata* Ell. et Halst.
I. Isolation of *C. fimbriata*, symptoms of the disease and evaluation of
resistance of poplar clones resulting from artificial
infection*.**

INTRODUCTION

Ceratocystis fimbriata is known to be a pathogen on a variety of hosts (Olson and Martin 1949, Feazeli and Martin 1950, Pontis 1951, De Vay and others 1968, Webster and Butler 1967, Cristinzio and others 1973, Panconesi 1973, Ferrari and Pichenot 1978, Muchovej and others 1978). Hyphae of the pathogen are capable of invading roots, stems, fruits only through fresh wounds.

C. fimbriata canker of aspen (*Populus tremuloides* Michx.) called "target canker" was described by Wood and French (1963), Zalasky (1965) and Hinds (1972). The first paper about the presence of the disease on poplars in Poland was done by Gremmen and de Kam (1977). Afterwards Przybył (1980 a, b) described symptoms on the trunks of poplars and presented a list of poplar clones from which *C. fimbriata* was isolated.

In this paper I would like to present additional studies on the distribution of *C. fimbriata* fungus in Poland, observations of the disease development after inoculations and evaluation of the degree of infection on many studied poplar clones.

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MATERIAL AND METHODS

ISOLATION OF *C. FIMBRIATA*

For the purpose of isolation of the causal agent of the target canker disease additional survey of poplar plantations was carried out in 1979 (Table 1). Samples of bark and phloem from the border line between healthy and necrotic tissues were collected from poplar clones, with careful maintenance of sterile conditions.

Inocula were portioned in the laboratory into smaller pieces which were put into a moist chamber and incubated at 29–30°C. Type of

Distribution of *Ceratocystis fimbriata*

Table 1

Clones of poplar	Place of isolation 1. Forest district ó. Forest Range	No of strain
Isolation in May 1979		
<i>P.</i> 'Kórník 1' (<i>P. maximowiczii</i> × <i>P. nigra</i> 'Italica' T × A)	1. Radomsko	2 I 79
<i>P.</i> 'Kórník 6°' (<i>P. maximowiczii</i> × <i>P. trichocarpa</i> ?* T × T)	2. Masłowice 1. Radomsko	3 I 79
<i>P.</i> 'Kórník 6°' (<i>P. maximowiczii</i> × <i>P. trichocarpa</i> ?* T × T)	2. Masłowice 1. Tomaszów Lub.	7 I 79
<i>P.</i> 'Kórník 41°' (<i>P. deltoides angulata</i> Cordata × <i>P. berolinensis</i> A × T)	2. Grodysławice 1. Radomsko	1 I 79
<i>P.</i> 'NE-42°' ('Hybrida 275' = <i>P. maximowiczii</i> × <i>P. trichocarpa</i> T × T)	2. Masłowice 1. Krasnystaw	5 I 79
<i>P.</i> 'NE-42°' ('Hybrida 275°' = <i>P. maximowiczii</i> × <i>P. trichocarpa</i> T × T)	2. Namule 1. Oleszyce	8 I 79
<i>P.</i> 'NE-44°' ('Hybrida 277°' = <i>P. maximowiczii</i> × <i>P. berolinensis</i> T × T)	2. Stybna 1. Ruda Naleniecka	4 I 79
<i>P.</i> 'NE-44°' ('Hybrida 277°' = <i>P. maximowiczii</i> × <i>P. berolinensis</i> T × T)	2. Czapla 1. Strzelce	6 I 79
<i>P.</i> 'NE 44°' ('Hybrida 277°' = <i>P. maximowiczii</i> × <i>P. berolinensis</i> T × T)	2. Jarosławiec 1. Oleszyce	9 I 79
<i>P.</i> 'NE 44°' ('Hybrida 277°' = <i>P. maximowiczii</i> × <i>P. berolinensis</i> T × T)	2. Stybna 1. Kańczuga	10 I 79
<i>P.</i> 'NE-44°' ('Hybrida 277°' = <i>P. maximowiczii</i> × <i>P. berolinensis</i> T × T)	2. Mokra 1. Pińczów	11 I 79
Isolation in July 1979		
<i>P.</i> 'Kórník 6°' (<i>P. maximowiczii</i> × <i>P. trichocarpa</i> ?* T × T)	2. Woszczowice 1. Miechów	2 II 79
<i>P.</i> 'Kórník 21°' (<i>P. nigra</i> Italica × <i>P. berolinensis</i> A × T)	2. Tunel 1. Brynek	6 II 79
<i>P.</i> 'Kórník 36°' (<i>P. maximowiczii</i> × <i>P. nigra</i> Italica T × A)	2. Bezchlebie 1. Brynek	7 II 79
<i>P.</i> 'NE-42°' ('Hybrida 275°' = <i>P. maximowiczii</i> × <i>P. trichocarpa</i> T × T)	2. Bezchlebie 1. Miechów	1 II 79
<i>P.</i> 'NE-42°' (Hybrida 275°' = <i>P. maximowiczii</i> × <i>P. trichocarpa</i> T × T)	2. Tunel 1. Miechów	4 II 79
<i>P.</i> 'NE-42°' ('Hybrida 275°' = <i>P. maximowiczii</i> × <i>P. trichocarpa</i> T × T)	2. Goszcza 1. Miechów	3 II 79
	2. Goszcza	

* Bugała, Stecki – 1961

Symbols of sections and intersectional hybrids:

A – *Aigeiros*

T × T – *Tacamahaca* × *Tacamahaca*

A × A – *Aigeiros* × *Aigeiros*

T × A – *Tacamahaca* × *Aigeiros*

T – *Tacamahaca*

A × T – *Aigeiros* × *Tacamahaca*

sporulation was studied in a suspension of sterile water after 4-5 days of incubation.

In order to obtain pure cultures of the *C. fimbriata* fungus a method described by Király and others (1977) was used. The suspension with endoconidial spores was streaked out on solid media: maltose (Difco) pH 5,5 and potato dextrose agar (Difco) pH 5,6 (PDA).

More over the fragments of bark and phloem which were taken from the infected trunks of poplar clones were immediately placed on the above mentioned media.

ARTIFICIAL INFECTIONS WITH *C. FIMBRIATA*

Pathogenicity test. Fourteen days old pure cultures of all isolated fungi together with the PDA medium, 0,5×0,5 mm in size, with three types of conidia (cylindrical, barrel-shaped and thick walled) have been

Table 2

Evaluation of the degree of infection of poplar clones. Experiment carried out on several years old shoots in 1979

Poplar clones	Observations after inoculation	
	2 months	12 months
<i>AIGEIROS</i>		
<i>P. deltoides</i> B- 18	2	2
<i>P. nigra</i> PW-3	2	2
<i>AIGEIROS</i> × <i>AIGEIROS</i>		
<i>P. deltoides</i> × <i>P. nigra</i> 490-2	1	2
<i>P. 'Marilandica'</i>	2	2
<i>P. PK-137</i> (8) (<i>P. nigra Italica</i> × <i>P. nigra</i>)	2	2
<i>P. Serotina</i> de Poitou'	2	2
<i>TACAMAHACA</i>		
<i>P. balsamifera</i>	3	2
<i>P. tacamahaca</i>	3	3
<i>TACAMAHACA</i> × <i>TACAMAHACA</i>		
<i>P. 'NE-46'</i> - <i>P. 'Geneva'</i> (<i>P. maximowiczii</i> × <i>P. × berolinensis</i>)	1	2
<i>P. 'NE-42'</i> , 'Hybrida 275' (<i>P. maximowiczii</i> × <i>P. trichocarpa</i>)	4	5
<i>P. maximowiczii</i> × <i>P. trichocarpa</i>	3	3/2
<i>P. 'PK-127'</i> (13) (<i>P. maximowiczii</i> × <i>P. laurifolia</i>)	3	3
<i>P. 'PK-127'</i> (15) (<i>P. maximowiczii</i> × <i>P. laurifolia</i>)	3/4	3
<i>P. 'PK-127'</i> (16) (<i>P. maximowiczii</i> × <i>P. laurifolia</i>)	3/4	3
<i>P. 'PK-127'</i> (18) (<i>P. maximowiczii</i> × <i>P. laurifolia</i>)	3	2
<i>TACAMAHACA</i> × <i>AIGEIROS</i>		
<i>P. 'PK-124'</i> (19) (<i>P. maximowiczii</i> × <i>P. nigra</i>)	3	2
<i>P. 'PK-124'</i> (20) (<i>P. maximowiczii</i> × <i>P. nigra</i>)	3	2
<i>P. 'PK-124'</i> (21) (<i>P. maximowiczii</i> × <i>P. nigra</i>)	4	3
<i>P. 'PK-124'</i> (22) (<i>P. maximowiczii</i> × <i>P. nigra</i>)	3	3
<i>P. 'PK-124'</i> (24) (<i>P. maximowiczii</i> × <i>P. nigra</i>)	4	3
<i>AIGEIROS</i> × <i>TACAMAHACA</i>		
<i>P. 'Kórník 23'</i> (<i>P. deltoides angulata</i> 'Cordata' × <i>P. × berolinensis</i>)	4	5
<i>P. 'NE-33'</i> 'Hybrida 283' (<i>P. deltoides angulata</i> 'Cordata' × <i>P. × berolinensis</i>)	3	5
<i>P. 'PK-136'</i> (2) (<i>P. nigra Italica</i> × <i>P. laurifolia</i>)	3	2
<i>P. 'PK-136'</i> (3) (<i>P. nigra Italica</i> × <i>P. laurifolia</i>)	4/3	2
<i>P. 'PK-136'</i> (4) (<i>P. nigra Italica</i> × <i>P. laurifolia</i>)	3	3
<i>P. 'PK-136'</i> (5) (<i>P. nigra Italica</i> × <i>P. laurifolia</i>)	3	3
<i>P. × petrowskyana</i> (<i>P. deltoides</i> × <i>P. laurifolia</i>)	3	2

Table 3

Evaluation of the degree of infection of poplar clones. Experiment carried out on four months old shoots in 1979

Poplar clones	Observations after inoculation			
	2 months		12 months	
	A	B	A	B
<i>AIGEIROIS</i>				
<i>P. × deltooides</i> 121	1	1	1	1
<i>AIGEIROIS × AIGEIROIS</i>				
<i>P. × deltooides × P. nigra</i>	2	2	2	2
<i>P. 'Marilandica'</i>	1	1	1	1
<i>P. 'Robusta'</i>	2	1	2	1
<i>P. 'Serotina'</i>	1	2	1	2
<i>TACAMAHAHA</i>				
<i>P. balsamifera</i>	3	3	2	2
<i>P. tacamahaca</i>	3	3	3	3
<i>TACAMAHAHA × TACAMAHAHA</i>				
<i>P. 'Kórnik 5'</i> (<i>P. maximowiczii × P. trichocarpa?</i> *)	4	4	4/5	4
<i>P. 'Kórnik 6'</i> (<i>P. maximowiczii × P. trichocarpa?</i> *)	4	4	4	4
<i>P. 'Kórnik 7'</i> (<i>P. maximowiczii × P. trichocarpa?</i> *)	3	3	3	3
<i>P. 'NE-44'</i> ('Hybrida 277' <i>P. maximowiczii × P. × berolinensis</i>)	3	3	3	3
<i>P. 'NE-49'</i> ('Hybrida 194' <i>P. maximowiczii × P. × berolinensis</i>)	3	3	3/5	3
<i>P. 'PK-127'</i> (13) (<i>P. maximowiczii × P. laurifolia</i>)	3	2	3	2
<i>P. 'PK-127'</i> (15) (<i>P. maximowiczii × P. laurifolia</i>)	3	2	3	2
<i>TACAMAHAHA × AIGEIROIS</i>				
<i>P. × berolinensis</i> (<i>P. laurifolia × P. plantierensis</i>)	2	2	2	2
<i>P. 'PK-124'</i> (21) (<i>P. maximowiczii × P. nigra</i>)	3	3	3	3
<i>P. 'PK-124'</i> (22) (<i>P. maximowiczii × P. nigra</i>)	3	3	3	3
<i>P. 'PK-124'</i> (24) (<i>P. maximowiczii × P. nigra</i>)	3	3	3	3
<i>AIGEIROIS × TACAMAHAHA</i>				
<i>P. 'Kórnik 23'</i> (<i>P. deltooides angulata 'Cordata' × P. × berolinensis</i>)	3	4	5	4
<i>P. 'NE-33'</i> ('Hybrida 283' <i>P. deltooides angulata 'Cordata' × P. × berolinensis</i>)	3	4	3	3
<i>P. 'PK-136'</i> (5) (<i>P. nigra Italica × P. laurifolia</i>)	3	3	3	3
<i>P. 'PK-136'</i> (6) (<i>P. nigra Italica × P. laurifolia</i>)	3	3	3	3
<i>P. × petrowskyana</i> (<i>P. deltooides × P. laurifolia</i>)	3	2	2	2

A - mycelium of fungus placed on leaf wound

B - suspension of spores sprayed on leaf wound

* Bugała, Stecki - 1961.

set onto wound immediately after detachment of leaf of four months old shoots of NE-42 (*Populus maximowiczii × P. trichocarpa*). The shoots of this poplar clone were growing in a greenhouse.

The disease symptoms were observed 48 hours, 7, 14, 30, 60 days after inoculations.

Invasion of the pathogen into host. Study of the invasion of *C. fimbriata* into the host was carried out on four months old poplar clones NE-42 (*P. maximowiczii × P. trichocarpa*), NE-44 (*P. maximowiczii × P. × berolinensis*), *P. 'Kórnik 6'* (*P. maximowiczii × P. trichocarpa*) and *P. 'Kórnik 13'* (*P. maximowiczii × P. nigra Italica*). The poplar clones have been infected in the greenhouse. Inoculum of *C. fimbriata* 0,5×0,5 mm in size, with three types of asexual conidia cylindrical, barrel-shaped, thick-walled and the sexual ascospores has been set: a) onto a leaf wound of the shoots, b) into a 3 cm length wound

Table 4

Evaluation of the degree of infection of poplar clones. Experiment carried out on four months old shoots in 1979

Poplar clones	Observations after inoculations			
	2 months		12 months	
	A	B	A	B
<i>AIGEIRO</i> S				
<i>P. nigra</i> 68/2	1	1/2	1	2
<i>P. nigra</i> 189	1	1/2	1	2
<i>P. nigra</i> 198	2	2	1	2
<i>P. nigra</i> 199	1	2	2	2
<i>P. nigra</i> Janinów	1	2	2	2
<i>P. nigra</i> Krzepice	2	2	2	2
<i>P. nigra</i> Ostromecko	2	1/2	2	2
<i>P. nigra</i> Placzkowo	2/1	2	2	2
<i>P. nigra</i> Tryńcza 2	2	2	2	2
<i>AIGEIRO</i> S × <i>AIGEIRO</i> S				
<i>P. Gelrica</i>	2	2	2	2
<i>P. 'Grandis'</i>	2	1/2	2	2
<i>P. 'Heidemij'</i>	1/2	1/2	2	2
<i>P. 'I-154'</i>	1/2	1/2	2	2
<i>P. 'I-214'</i>	1	2	2	2
<i>P. 'I-488'</i>	2	2	2	2
<i>P. 'Robusta'</i>	1/2	2	2	2
<i>P. 'Serotina'</i>	2	2	2	2
<i>TACAMAHA</i> CA				
<i>P. maximowiczii</i>	3	2	3	2
<i>P. trichocarpa</i> Łódź 1/73	1/2	2	2	2
<i>P. trichocarpa</i> Łódź 2/73*	3	3	3	3
<i>P. trichocarpa</i> 28/34	3	3	3	3
<i>P. trichocarpa</i> 105	1	1	2	2
<i>P. trichocarpa</i> 630/71	2/3	3	2/3	2/3
<i>P. trichocarpa</i> 631/71	2/3	3	2/3	2/3
<i>P. trichocarpa</i> 910	1	1	2	2
<i>P. trichocarpa</i> 916	1	1	2	2
<i>P. trichocarpa</i> 1153	1/2	2	2	2
<i>P. trichocarpa</i> 1199	1/2	1/2	2	2
<i>P. trichocarpa</i> 1200	2/3	1/2	2/3	2
<i>P. trichocarpa</i> BC 6209	1	2	2	2
<i>P. trichocarpa</i> 6209	3	3	3	2/3
<i>P. trichocarpa</i> OB Poznań	1	2	2	2
<i>P. trichocarpa</i> var <i>hostata</i>	2	1/2	2	2
<i>TACAMAHA</i> CA × <i>TACAMAHA</i> CA				
<i>P. 'Kórnik 6'</i> (<i>P. maximowiczii</i> × <i>P. trichocarpa</i> ?)	4	4	5	5
<i>AIGEIRO</i> S × <i>TACAMAHA</i> CA				
<i>P. 'Kórnik 41'</i> (<i>P. deltoides angulata</i> 'Cordata' × <i>P. × berolinensis</i>)	3	3	3/2	2

A - mycelium of fungus placed on leaf wound;

B - suspension of spores sprayed on leaf wound

* Bugała, Stecki - 1961

made with a scalpel, c) onto surface of leaves from which the epidermis was removed, d) onto intact surfaces of shoots and leaves.

Observations on the development of the disease were carried out 48 hours, 7, 14, 30, 60 days after inoculations.

Development of disease on inoculated shoots of poplar clones. One to three years old shoots were used from

27 poplar clones represented by different numbers (10 - 20) of trees planted in rows in a field trial (Experimental Forest — Zwierzyniec) (Table 2). The plants have been infected with *C. fimbriata* at one height on the shoot. Mycelium of the fungus with asexual and sexual spores together with the PGA medium has been set into a $0,5 \times 1$ cm wound made by the removal of the epidermis. The inoculum was fasten to the shoots using poloplast.

In June 1979 four months old shoots of 23 poplar clones growing in pots in the greenhouse were also infected with *C. fimbriata* (Table 3). In this case, the following methods of inoculation were used: a) the culture of the fungus together with the PDA medium has been set onto wound immediately after detachment of leaf, b) the shoots have been sprayed after detachment of leaves with a spores suspension (5×10^8 per 1 ml sterile water). Density of spores in sterile water was determined by counting using a Thom's chamber (Bieszkiewicz 1971).

In both variants of the experiment the plants were inoculated at three levels along the height of the shoots. Each variant consists of five replications. On the other hand each studied poplar clone was represented by four plants (three infected plants+control).

In all experiments the observations of the development of the disease was performed 48 hours, 4, 7, 14, 30, 60 days and 6, 12, 20 months after inoculation.

A new experiments with artificial infection were begun in July 1980 on four months old shoots of 35 poplar clones (Table 4). Also in this case, the two infection methods mentioned above were applied.

The experiment had three replications. Each was represented by four plants (three infected plants+control).

Observations of development of the disease were carried out 48 hours, 7, 14, 30, 60, days and 6, 12 months after inoculation.

In experiments performed both in 1979 and 1980 the shoots of plants which were used as controls, were treated in the same way, as those of the inoculated plants.

Reisolations of *C. fimbriata* fungus from the inoculated plants were made two months after inoculation. The same method of isolation as described above was used.

EVALUATION OF THE DEGREE OF INFECTION

In order to select poplar clones more or less resistant to *C. fimbriata* infection a 6 degrees scale evaluating the intensity of infection of the studied poplar clones was used. The scale is based on the development of disease on the shoots of poplar clones after artificial infection with *C. fimbriata*. The evaluating scale was as follows:

	degree of infection
no symptoms on the shoots at all	0
presence of a brown spot and a small depression in the point of infection on the shoots	1
cicatrization of the wound on the shoot by callus tissue	2
presence of wound on shoots with a more or less developed callus tissue	3
presence of a deep wound on the shoots non covered by callus tissue	4
presence of the disease stage called 'target canker', or death of plants in the case of four months old shoots	5

RESULTS

SYMPTOMS OF THE DISEASE OBSERVED ON NATURALLY INFECTED POPLAR TREES FROM WHICH *C. FIMBRIATA* WAS ISOLATED

Samples of bark and phloem were collected from trunks of 17 trees. They belonged to 7 poplar clones (Table 1). On trunks of trees from which the *C. fimbriata* was isolated three stages of the disease were observed: — early stage (Fig. 1), characterized by the presence of superficial wounds, — moderate stage (Fig. 2) characterized by the presence of deep wounds with irregular edges. On the edges of it, layers of callus tissue were formed. Advanced stage (Fig. 3) characterized by the presence of concentrically displaced crumbling away callus rings around on open deep wound (Przybył 1980 b).

Irrespective of the disease stage, after bark removal there was a dark brown coloured layer observed on the border line between healthy and diseased tissue (Fig. 4).

ARTIFICIAL INFECTION WITH *C. FIMBRIATA*

Pathogenicity test. On shoots of NE-42 a dark brown spot at the point of infection was seen within four days after inoculation. The elongation of the brown spot (1-1,5 cm) was observed two weeks after inoculation. Also during that time the first narrow wounds, on the brownish surface of shoots were observed, resembling those after incision. A month after inoculation a wide, deep wound with a first layer of callus tissue on the edges was observed.

The shorter superficial wounds on both sides of a deep wound appeared two months after inoculation.

All isolated strains of *C. fimbriata* fungus were capable of causing disease symptoms on four months old shoots of NE-42. The differences

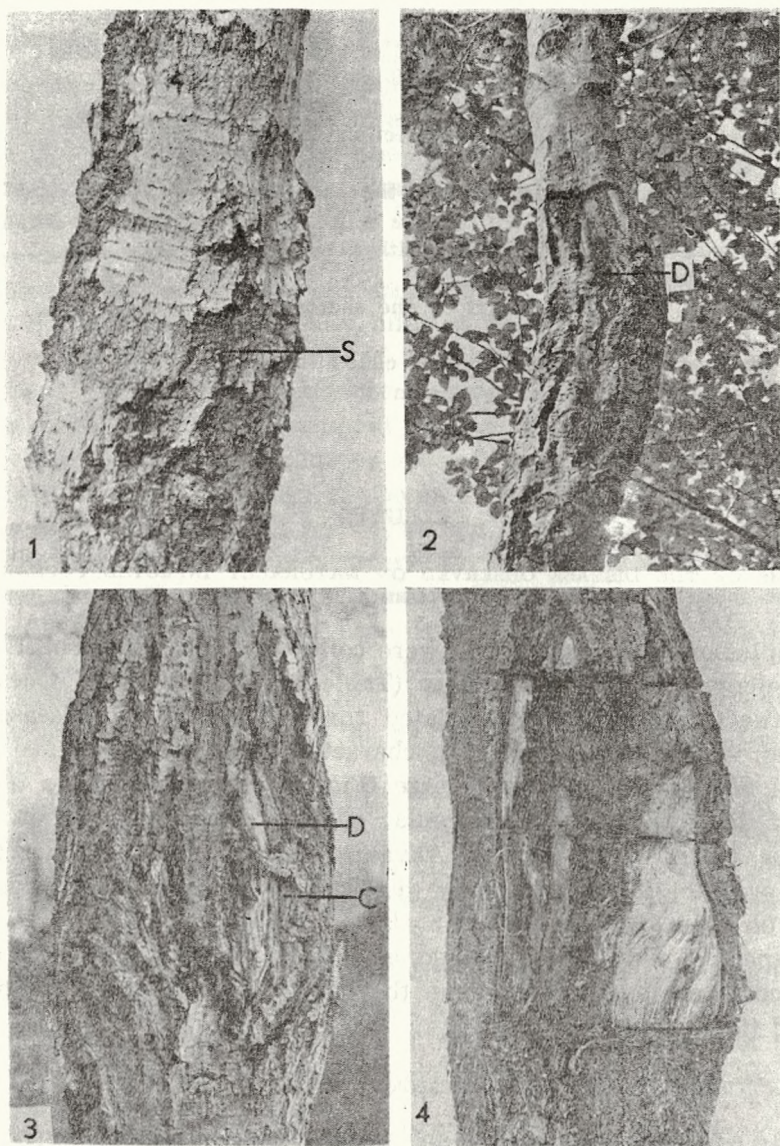


Fig. 1-4: 1 — Early stage of disease caused by *C. fimbriata* on the trunk of 'NE-49'. Superficial wound — S. 2 — Moderate stage of disease caused by *C. fimbriata* on the trunk of 'NE-49'. Deep wound — D. 3 — Advanced stage of disease caused by *C. fimbriata* on the trunk of 'NE-42'. Deep wound — D, callus — C. 4 — A dark brown layer on the border line between the healthy and diseased tissues infected by *C. fimbriata* on the trunk of 'NE-42'. Photo: 1-4 by K. Jakusz

Isolation of the pathogen *C. fimbriata* from trunks of poplar clones, demonstrating the stages of disease on figures 1-3 was defined by Przybył (1980a)

in pathogenicity of all the studied *C. fimbriata* strains refer only to the length of the brown spot.

Invasion of the pathogen into the host. When the inoculum *C. fimbriata* has been set onto a wound after detachment of leaf the above mentioned symptoms of disease were visible on four month old shoots poplar clones after two months of observations.

On the other hand, when the inoculum has been put into 3 cm long superficial wounds, the clones reacted with a 10-12 cm long brown spot of infection with *C. fimbriata* within two weeks after inoculation. The first layer of callus tissue on the both sides of the deep wound appeared one month after inoculation.

On the leaves which have been inoculated by putting the culture of *C. fimbriata* onto an injured surface the following symptoms were observed: a) a brown spot at the point of inoculation, b) bleaching of a part of the leaf blade, which surrounds the point of inoculation.

Within two weeks after inoculation the clear fragment of the blade began to darken.

A month or more after inoculation no further development of the disease was observed.

No symptoms of the disease were seen when inoculation was carried out on an undamaged surface of the leaf blade.

Development of the disease. Observations on 1-3 years old shoots of poplar clones. A brownish decay of the leaf scar (place of infection) was observed on all poplar clones within four days after inoculation. Growth of a white mycelium without perithecia was seen only on the shoots of 'NE-42' and *P. 'Kórnik 6'*. Seven days after inoculation the first differences between the clones were observed in the length of the brown spot.

Two weeks after inoculation on all studied poplar shoots an elongation of the brown spot and a depression of a dark surface were seen. Also after that time, an abundant growth of the mycelium together with perithecia containing ascospores characteristic for *C. fimbriata* were visible on the dark surface of shoot (Fig. 5). Deep tissues of shoots, for example in 'PK-135' (1) and 'PK 124' (21) crumbled away. As a consequence a superficial wounds appeared.

Within one month from the moment of inoculation the following symptoms were observed on the investigated shoots:

- presence of a non-elongated, wet and dark spot and narrow superficial wounds e.g. on shoots of clones *P. deltoides* × *P. nigra* (490-2) and *P. 'Geneva'* (Fig. 6),
- cicatrization of the point of infection with help of an abundantly formed callus tissue e.g. on *P. deltoides* B 218, *P. 'Marilandica'*, *P. nigra* PW-3 and *P. 'Serotina de Poitou'* (Fig. 7),
- presence of a wide, deep wound with callus tissue formed more er

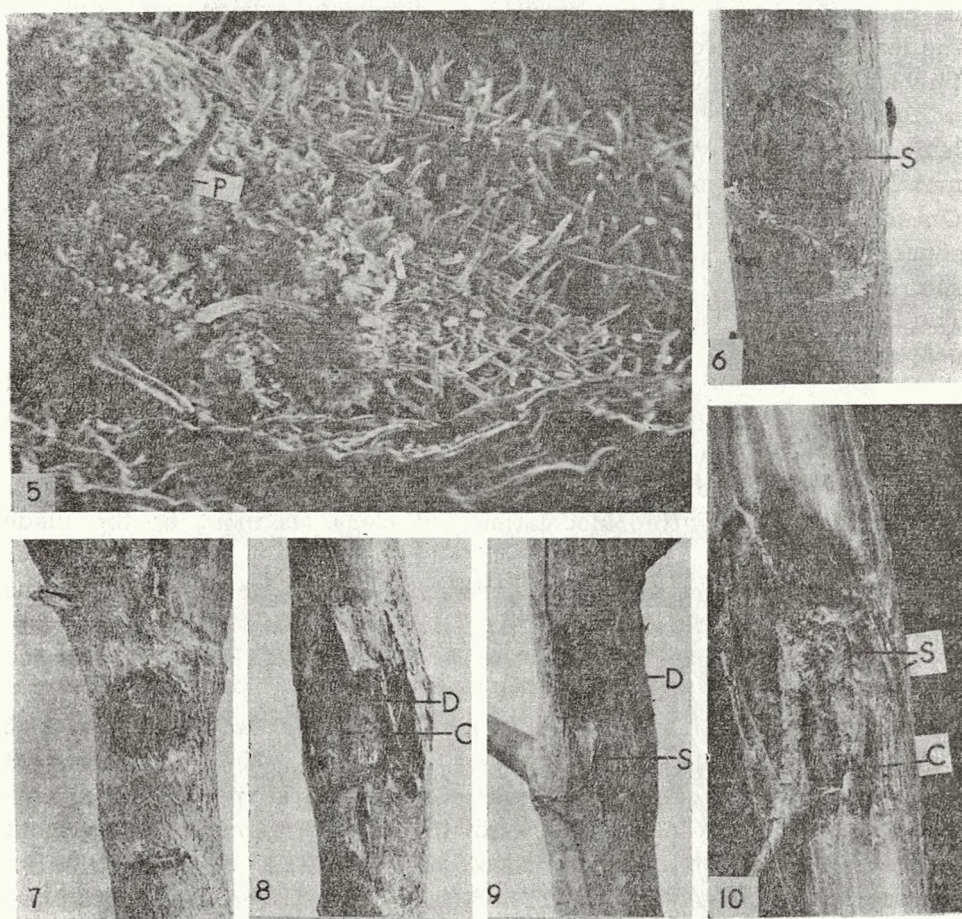


Fig. 5 - 10: 5 — Mycelium with perithecia (P) of *C. fimbriata* on the dark surface of shoots two weeks after inoculation. Scanning electron microscope, $\times 140$. 6 — A narrow superficial wounds (S) at the point of inoculation on the shoot of *P. 'Geneva'* one month after inoculation. 7 — Cicatrization of the point of infection on the shoot of *P. nigra* PW-3 one month after inoculation. 8 — A wide, deep wound (D) with callus tissue (C) on the shoot of *P. maximowiczii* \times *P. trichocarpa* one month after inoculation. 9 — A wide, deep wound (D) and shorter superficial wounds (S) with no formation of callus tissue, on the shoot of *P. 'Kórnik 23'* one month after inoculation. 10 — Deep wound with callus tissue (C) and with concentrically dislocated superficial wounds (S) on the shoot of 'NE-33' twelve months after inoculation. Photo: 5 by F. Młodzianowski, 6 - 10 by E. Szubert

less on edges and superficial wounds beyond eg. on *P. balsamifera*, *P. maximowiczii* \times *P. trichocarpa*, 'NE-33', *P. petrowskyana*, 'PK-124' (22), 'PK-127' (18), 'PK-136' (2, 4, 5 and 6), *P. tacamahaca* (Fig. 8), — presence of a wide, deep wound and shorter superficial wounds without any formation of callus tissue e.g. *P. 'Kórnik 23'* 'NE-42' PK-124 (21, 24), 'PK-127' (16), 'PK-137' (8) (Fig. 9).

The deep wounds which were visible one month after infection e.g. on 'PK-124' (19) and (20), *P. ×petrowskyana* were cicatrized by means of an intensive development of the callus tissue two or six months after inoculation.

On the other hand an elongation of the deep wounds took place and the superficial wounds deepened on shoots of those poplar clones, where cicatrization was not observed. Wide and deep wounds with callus tissue and with concentrically dislocated superficial wounds were observed on the shoots of *P.* 'Kórnik 23', 'NE-33' and 'NE-42'. During observations carried out over six months, the elongation of the dark spots and than wounds progressed in the basipetal direction. Twelve months after inoculation the new superficial and deepened wounds were formed around and above the point of infection (Fig. 10). The phenomenon was seen on shoots representing the 'target canker' stage of the disease and on shoots on which the point of inoculation cicatrized totally twenty months after inoculation.

Observations on four months old shoots. Experiments carried out in 1979 and 1980. The same symptoms described above were observed on four months old shoots inoculated by the spraying method and by putting a mycelium of *C. fimbriata* on the wound after detachment of leaf. In the experiments carried out in 1979 and 1980, the stage of disease resembling a 'target canker' on four months old shoots was not seen. The final picture in the course of the disease development was the characteristic breakage or by death of shoots observed twelve months after inoculation e.g. on *P.* 'Kórnik 6' and *P. trichocarpa* 28.

EVALUATION OF THE DEGREE OF INFECTION

The shoots presenting a low degree of infection (1 and 2 on the evaluation scale) were those, on which the point of inoculation was cicatrized two months after inoculation (Tables 2, 3 and 4). Clones with shoots reacting in this way were considered resistant to the infection with *C. fimbriata*. The moderately resistant poplar clones were those, on which shoot recover wounds being overgrown by an abundantly produced callus tissue observed six or twelve months after inoculation (degree 2 on the inoculation scale) e.g. *P. ×balsamifera*, *P. ×petrowskyana*. In the case of susceptible clones the 'target canker' or the death of shoots were the final pictures of disease development (degree 5 on the evaluation scale). Among the susceptible clones were also those on which new superficial and deep wounds were formed in spite of a cicatrization of the point of infection. The wounds were observed twelve months after inoculation around the point of infection e.g. *P.* 'Kórnik 23' (Table 2).

The degrees of infection of all the investigated poplar clones are demonstrated in tables 2, 3, and 4.

REISOLATION OF THE PATHOGEN

The reisolation of *C. fimbriata* was carried out from inoculated shoots of poplar clones selected at random. The shoots of these clones presented various pictures of the disease. In all cases the isolated fungus showed the perfect and imperfect stages typical for *C. fimbriata*.

DISCUSSION

Ceratocystis fimbriata has been isolated from trunks of poplar clones being hybrids within section *Tacamahaca* (T×T) or intersectionally *Aigeiros*×*Tacamahaca* (A×T) and *Tacamahaca*×*Aigeiros* (T×A). During the observations carried out on the poplar plantations in nature, among all infected trees it was visible most often on clones 'NE-42', 'NE-44' and P. 'Kórnik 6'.

In this paper a method was presented of the isolation of *C. fimbriata* from poplar clones as used by Gremmen and de Kam (1977). In this case the samples of bark and phloem were put into a moist chamber and incubated at 29 - 30° C. The same method only with a different temperature of incubation, was applied by Vigoroux (1979) for the isolation of *C. fimbriata* f. *platani* and by Pontis (1951) for the isolation of *C. fimbriata* from coffee trees.

In Polish climatic conditions the three stages of the disease were recognized: early stage, moderate stage and an advanced stage (Przybył 1980 b). Symptoms which were characteristic for the advanced stage of poplar disease were similar to the 'target canker' observed on trunks of aspen by Wood and French (1963) and Zalasky (1965).

In experiments carried out on four months old shoots of a susceptible, poplar clone 'NE-42' it was shown that the fungus invaded the host through wound after detachment of leaf. Establishment of this was the basis for the use of the leaf wound also as the point of infection in experiments on the selection of poplar clones for resistance to infection with *C. fimbriata*. Another method of inoculation was described by DeVay and others (1968). In this method agar with mycelium of the fungus was put beneath bark of the trees.

Two months after inoculation the point of infection was cicatrized on shoots of clones from section *Aigeiros* and hybrids within it (*Aigeiros*×*Aigeiros*). No symptoms were seen on the clones in a further observations (four, six, twelve months after inoculation). Clones reacting in this way, were evaluated as resistant to infection with *C. fimbriata*. Susceptible clones were those, on the shoots of which the 'target canker' or deep wound without callus formation or death resulted on four months old shoots observed twelve months after inoculation. Among the

group of moderately resistant clones were those, which were classified with second a third degree of infection. In this case the disease can to be inhibited or it may deepened. Independently of the possible recovery of the infected trees, the opened wound provides a convenient point for reinfection with the same pathogen or with others for a certain time.

In the tables 2, 3 and 4 sometimes two degrees of infection with *C. fimbriata* of poplar are shown the some poplar for clones. It means that not all trees of the some clone reacted in the same way to the infection with the pathogen.

The paper shows, that the used scale of evaluation of the degree of infection based on many criteria can to be used in the selection of poplar clones for resistance to attack by *C. fimbriata*.

Is not always, sufficient to evaluate resistance only on the basis of the length of necrotic spots or of the wound. The length of wounds is not always proportional to the intensity of infection.

SUMMARY

The fungus *Ceratocystis fimbriata* — causative agent of the disease of poplars was isolated from seventeen trees of seven poplar clones hybrids with in section *Tacamahaca* (T×T) and intersectional *Aigeiros*×*Tacamahaca* (A×T) and *Tacamahaca*×*Aigeiros* (T×A).

In the paper the development of the disease on artificially infected four months old and one to three years old shoots was described. Also lists of many poplar clones with the evaluation of the degree of their infection are presented. It was confirmed that poplars of section *Tacamahaca* (T) or intersectional hybrids (*Tacamahaca*×*Tacamahaca*) as well as intersectional *Aigeiros*×*Tacamahaca* (A×T) and *Tacamahaca*×*Aigeiros* (T×A) are more susceptible to infection with *C. fimbriata* than clones belonging to the group of black poplars (*Aigeiros*).

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*Choroba topoli wywołana przez grzyb Ceratocystis fimbriata
Ell. et Halst.*

I. Izolacja *C. fimbriata*, symptomy chorobowe oraz ocena stopnia porażenia sztucznie zakażonych klonów topoli

Streszczenie

W roku 1979 wyizolowano grzyb *Ceratocystis fimbriata* Ell. et Halst. z siedmiu klonów topoli (siedemnastu drzew) będących mieszańcami w obrębie sekcji *Tacamahaca* (X T) oraz mieszańcami międzysekcyjnymi: *Tacamahaca* × *Aigeiros* (T × A) i *Aigeiros* × *Tacamahaca* (A × T).

W przeprowadzonym na pędach podatnego na infekcję *C. fimbriata* klonu P. 'NE-42' teście na patogeniczność wykazano, że wszystkie wyizolowane szczepy grzyba były w równym stopniu zdolne do wywołania choroby. Rozwój choroby

obserwowano na sztucznie zakażonych losowo wybranymi szczepami patogena 4-miesięcznych i 1-3-letnich pędach kilkudziesięciu klonów topoli. Ocena stopnia porażenia klonów topoli przedstawiono za pomocą 6-stopniowej skali. W wyniku tej oceny stwierdzono, że topole z sekcji *Tacamahaca* oraz będące mieszańcami w obrębie tej sekcji i międzysekcyjnymi (A×T i T×A) są bardziej podatne na infekcję *C. fimbriata*, niż klony należące do grupy topoli czarnej.

Zabolewanie topolej вызванное грибом Ceratocystis fimbriata
Ell. et Halst. I. *Изоляция C. fimbriata, симптомы болезни и оценка степени поражения искусственно зараженных клонов топелей*

Резюме

В 1979 году был изолирован гриб *Ceratocystis fimbriata* Ell. et Halst. с семи клонов топелей (семнадцать деревьев) являющихся гибридами в рамках секции *Tacamahaca* (T×T) и межсекционными гибридами: *Tacamahaca*×*Aigeiros* (T×A) и *Aigeiros*×*Tacamahaca* (A×T).

В проведенном на побегах чувствительного к заражению *C. fimbriata* клона Р. 'NE-42' тестировании патогенности оказалось, что все изолированные штаммы гриба были в равной степени способны вызывать заболевание. Развитие болезни наблюдалось на искусственно зараженных, случайно выбранными штаммами патогена 4-месячных и 1-3-летних побегах нескольких десятков клонов топелей. Оценка степени поражения клонов топелей представлена с помощью 6-бальной шкалы. В результате этой оценки отмечено, что тополя с секции *Tacamahaca* и гибриды в рамках этой секции и между секциями (A×T и T×A) являются более восприимчивыми к инфекции вызванной *C. fimbriata*, нежели клоны принадлежащие к группе черных топелей.

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