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Fast Decline of *Pythium zingiberum* in Soil and Its Recolonization by Cultivating Susceptible Host Plants

Takio ICHITANI and Tokiya SHIMIZU *

Laboratory of Plant Pathology, College of Agriculture

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Abstract

This experiment demonstrates the fast decline of *Pythium zingiberum* in soil and its recolonization by cultivating mioga, susceptible host plant, and discusses growth and survival of the pathogen in the host rhizosphere in cultivated fields.

Introduction

While a population of *Pythium zingiberum* causing rhizome rot of ginger and mioga may decline rapidly in a field soil¹⁾, the fungus can recolonize after the cultivation of host plants²⁾. It becomes detectable after a relatively short rotation period but can not be isolated from the soil when the rotation is longer³⁾.

This experiment was designed to demonstrate the fast decline of *P. zingiberum* in soil and its recolonization by cultivating mioga, *Zingiber mioga* (Thunb.) Roscoe, and to discuss growth and survival of the pathogen in the host rhizosphere in cultivated fields. Part of the work has already been presented elsewhere²⁾.

Materials and Methods

Soils from ginger-growing areas Each 30 kg of naturally infested sandy soil was collected at a depth of 5–30 cm adjacent to plastic house No.1 in Wakayama with a history of ginger rhizome rot⁴⁾. The collections were made before (Oct. 22) and after (Dec. 27) applying a strong drench (9 $\mu\text{g}/\text{m}^2$) of echomezol on Dec. 2, 1979. Similarly, uninfested soil was also collected on Dec. 27 adjacent to plastic house No.15 (near No.1), without a history of the disease⁴⁾. Each soil was divided into 2 pots (30 cm diam.). Two autoclaved soils served as a check. The pots were then placed in a glasshouse.

About 300 kg of heavily infested soil as well as 10 kg of rhizome rotted ginger was collected from the inside of plastic houses No.10 and 17⁴⁾ (near No.1) on June 14, 1977. At this time the disease was unusually prevalent in houses 10 and 17. The soil was mixed thoroughly with small pieces of diseased rhizomes, and then divided into 2 parts: autoclaved and unautoclaved. These were placed in concrete cylinders (85 cm diam., 60 cm high) buried in a field.

Soils from mioga-growing areas Naturally infested sandy loam soil was collected from mioga-growing areas in Nara and Wakayama⁵⁾ on Aug.6, 1978 and 8 samples, each 10 kg, were placed in 30 cm pots, half of which were autoclaved. The pots were then

* Present address: Agricultural Policy Section, Minami-saku Regional Office, Nagano Prefecture, Usuda, Nagano. 384-03.

kept under glasshouse conditions.

Cultivation of disease-free mioga in collected soils Mioga rhizomes were collected from the field without a history of rhizome rot. After random examination of the rhizomes and adhering soil for the pathogen on a selective medium⁵⁾, the disease-free rhizomes were propagated in an isolated growth chamber and checked bimonthly for reinfestation. Disease-free rhizomes thus obtained were cut into 3 cm lengths and 3 or 6 seed pieces were transplanted into each pot or cylinder. Tap water was supplied and any weeds were removed. Corn-meal (30 g/10 kg soil) was introduced once a year in autumn. The same management was applied to non-cultivated pots or cylinders.

Detection methods A small piece (ca. 1 × 1 × 2 mm) of tissue was cut from each separate discolored area of the cultivated mioga. All soil samples were collected from either rhizospheres of mioga or at a corresponding depth (3–10 cm) from containers without mioga. Detection was performed within 1–2 days after sampling by the method reported previously⁶⁾, using soils and/or tissues. Washing and surface sterilization was unnecessary.

Although 10–10⁴ propagules/g oven dry soil could be detected by this method⁷⁾, an identification error of about 10 % might be involved⁸⁾. Percentage detection was calculated from the ratios of samples from which *P. zingiberum* were isolated to 80 assayed samples.

Results

Recolonization of P. zingiberum in soils collected from the surrounding parts of ginger-growing plastic houses As shown in Table 1, no *P. zingiberum* was detected in soils kept under glasshouse conditions for 7 months before amending with ginger extract. Although the fungus was detected slightly in soil No.2, collected before drenching and amended with extract of ginger rhizomes, it was not found later, regardless of heavy introduction of fresh above-ground parts of mioga. On the other hand, a considerable quantity of the fungus was found after 1 year cultivation of mioga in soil No.1 out of 2 soils which were collected before the drench. However, no *P. zingiberum* was detected in another soil of the same experimental series.

Recolonization of P. zingiberum in soils collected from the inside of ginger-growing plastic houses In concrete cylinders under field conditions, no *P. zingiberum* was detected during the non-growing season of mioga (Sept. –May) even after recolonization and no effect of amendment with ginger extract was found in this season. However, the fungus became detectable during the growing season of mioga in the subsequent year (Table 2).

Recolonization of P. zingiberum in soils collected from mioga-growing fields In pots in the glasshouse, no *P. zingiberum* became detectable within 6 months, but the fungus was found in 1 out of 2 pots after 1 year cropping of mioga (Table 3).

Table 1. Detection of *Pythium zingiberum* before and after cultivation of susceptible mioga in soils collected from areas adjacent to ginger-growing plastic houses (glasshouse conditions).

Date detected	Part ¹⁾	% of detection in soils							
		Collected				Uninfested		Autoclaved	
		Before drench ²⁾		After drench		1	2	1	2
		1	2	1	2	1	2	1	2
May 28, '80 ³⁾	Soil	0	0	0	0	0	0	0	0
June 11, '80	Soil	0	1	0	0	0	0	0	0
June 28, '80	Soil	0	0	0	0	0	0	0	0
July 7, '80	Soil	0	0	0	0	0	0	0	0
Oct. 7, '80 ⁴⁾	Soil	0	0	0	0	0	0	0	0
Oct. 20, '80 ⁵⁾	Soil	0	0	0	0	0	0	0	0
Aug. 12, '81 ⁶⁾	Discolored rhizomes	28	0	0	0	0	0	0	0
	Soil	4	0	0	0	0	0	0	0

- 1) Rhizosphere or non-rhizosphere soil, depending upon whether mioga plants are cultivated.
- 2) Estimated 0.3 % average infestation before drench (Dec., 1977–Nov., 1979).
- 3) Kept from Dec. 28, 1979, under glasshouse conditions to the time just before amending with 100 g of ginger extract/pot on May 28, 1980.
- 4) Fresh above-ground parts of mioga introduced, 700 g/pot.
- 5) Mioga seed pieces transplanted, 3/pot.
- 6) All stems diseased in No. 1 soil collected before drench.

Table 2. Detection of *Pythium zingiberum* before and after cultivation of susceptible mioga in heavily infested soil collected from the inside of ginger-growing plastic houses (field conditions).

Date detected	Part ¹⁾	% of detection in soils in concrete cylinders			
		Autoclaved		Unautoclaved	
		1	2	1	2
Aug., '77	Soil	0	0	3	4
Oct., '77	Soil	0	0	0	0
May, '78	Soil	0	0	0	0
June, '78	Soil	0	0	0	0
Sept., '78 ²⁾	Soil	0	0	0	0
Nov., '78	Soil	0	0	0	0
Nov., '78 ³⁾		Yes	No	Yes	No
Aug., '79 ⁴⁾	Soil	0	0	5	0
Nov., '79	Soil	0	0	0	0
June, '80 ⁵⁾	Discolored rhizomes	0		63	
	Soil	0	0	7	0
Nov., '80	Soil	0	0	0	0
May, '81 ⁶⁾	Discolored rhizomes	0		0	
	Soil	0	0	0	0
July, '81 ⁷⁾	Discolored rhizomes	0		46	
	Soil	0	0	53	0
Aug., '81	Soil	0	0	9	0

- 1) As in Table 1.
- 2) Amended with 1 kg ginger extract/cylinder.
- 3) Mioga seed pieces transplanted, 6/cylinder.
- 4) No disease symptoms seen.
- 5) Only 1 stem diseased.
- 6) Apparently healthy (No disease stems remaining due to decomposition in the autumn-winter period).
- 7) Severely diseased (6 out of 11 stems).

Table 3. Percentage detection of *Pythium zingiberum* before and after cultivation of host plants in naturally infested field soils collected from mioga-growing areas (glasshouse conditions)¹⁾.

Date detected	Part ²⁾	Autoclaved soils in pots collected from:				Unautoclaved soils in pots collected from:			
		Nara		Wakayama		Nara		Wakayama	
		1	2	1	2	1	2	1	2
Aug., '78	Soil ³⁾	0	0	0	0	33	45	50	21
Jan., '79	Soil	0	0	0	0	0	0	0	0
Feb., '79	Soil	0	0	0	0	0	0	0	0
Apr., '79 ⁴⁾	Soil	0	0	0	0	0	0	0	0
Mar., '80	Discolored rhizomes	0	0	0	0	0	33	0	13
	Soil	0	0	0	0	0	3	0	3
Aug., '81	Discolored rhizomes	0	0	0	0	0	43	0	4
	Soil	0	0	0	0	0	1	0	0

1) The numerals are % of detection.

2) As in Table 1.

3) *P. zingiberum* with high pathogenicity to young mioga plants detected just after collection.

4) Mioga seed pieces transplanted, 3/pot.

Discussion

The fact that no *P. zingiberum* was detected in 1 out of 2 soils employed (see Tables 1, 3) may indicate either that populations were too low to recolonize or that there were no viable forms in these soils. Although the soils were divided uniformly and experiments were conducted under the same conditions, different results were obtained in recolonization, indicating that the critical factors involved growth and survival of the fungus.

From the results shown in Table 1, the presence of *P. zingiberum* was proved in the soil adjacent to the plastic house with a history of the rhizome rot of ginger. Since the fungus was pathogenic and its population was increased considerably by cultivation of the susceptible mioga, and since the soil inside the plastic house was sterilized before every cropping, the fungus found in the adjacent soil before drenching may have been involved in primary infection in this plastic house. The fact that no *P. zingiberum* was detected in soil collected after drenching, or in soil from an area adjacent to the plastic house without a history of the disease, indicates that the soils were disinfected by the drench or definitely noninfested.

On September 24, 1981, after finishing this experiment, all soils kept in the glasshouse were transferred into concrete cylinders buried in the field. Soils and discolored areas of rhizomes were examined continually and *P. zingiberum* was found in both samples during the growing season of mioga in 1982.

Ginger extract and above-ground parts of mioga had little or no effect on recolonization by *P. zingiberum*, but growing mioga roots were highly effective. Recolonization also occurred around growing ginger roots. On the other hand, the fungus could not be isolated from the rhizosphere soils of various plants or from their roots in both cultivated and uncultivated areas, except for the rhizosphere of ginger and mioga. A few isolations

were made from the roots of some gramineous weeds⁹⁾ but could not be repeated (unpublished data). Thus, the growth and probable survival of *P. zingiberum* in soil may be restricted mainly to the growing host roots. The fact that this fungus can not be isolated from the rhizosphere soils of a wild mioga²⁾ even after cultivating susceptible mioga (unpublished data) may indicate that it is not present in these soils. Other *Pythium* spp. having a high saprophytic ability should be used in comparative studies on *P. zingiberum* and its behavior in soil.

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