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Note

Long-term changes in frequencies of *Pythium iwayamai* and *P. paddicum* in upland- and paddy-field soils*

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The long-term changes in the frequencies of *Pythium iwayamai* and *P. paddicum* in upland- and flooded paddy-field soils were compared. The frequencies of both species fell in both soils within the first 6 months after they had been buried. After 36 and 48 months, *P. iwayamai* showed higher frequency than *P. paddicum* in upland-field soil, while *P. paddicum* showed higher frequency than *P. iwayamai* in paddy-field soil. These findings indicate respectively higher colonization ability to organic matters of *P. iwayamai* in upland-field soil and *P. paddicum* in paddy-field soil.

Key Words——detection; paddy-field soil; *Pythium iwayamai*; *Pythium paddicum*; upland-field soil.

In a study of *Pythium* snow rot of wheat and barley, the prevailing pathogens in upland- and poorly drained paddy-field soils were found to be *Pythium iwayamai* S. Ito and *Pythium paddicum* Hirane, respectively (Takamatsu and Ichitani, 1987a, b, c). Factors involved in this distribution pattern have been analyzed from the aspects of differences in virulence (Takamatsu, 1989), pH ranges for mycelial growth (Inoue and Ichitani, 1990), pH ranges for oospore germination (Ge and Ichitani, 1992), tolerance of mycelium to CO_2 (Takamatsu et al., 1987) and tolerance of oospore germination to osmotic potentials (Ge and Ichitani, 1992).

This study deals with the long-term changes in the frequencies of *P. iwayamai* and *P. paddicum* in upland- and paddy-field soils. Part of the work has been reported elsewhere (Ichitani and Takamatsu, 1988; Hiraoka et al., 1992).

Fungal isolates *P. iwayamai* (UOP 381) and *P. paddicum* (UOP 405) were used. These two species were obtained from wheat-growing soils in Fukui Prefecture in 1982. After assaying their pathogenicity to wheat and barley leaves (Takamatsu, 1983), these species were identified by referring to *P. iwayamai* UOP 8 (=IFO 31990) and *P. paddicum* UOP 6 (=IFO 31993), following the methods reported elsewhere (Ichitani et al., 1986). The fungi were maintained as described previously (Ichitani and Kang, 1988).

Soils and burying of oospores Upland-field (loam) and paddy-field (sandy clay) soils were collected at the University Farm, University of Osaka Prefecture (Sakai, Osaka), in April, 1988. The soils were assumed to be free from *P. iwayamai* and *P. paddicum* because neither species had been isolated in attempts made since 1984 by a baiting method with wheat cultivar 「Norin 1」 leaf pieces (Takamatsu and Ichitani, 1984), which is considered to be the most sensitive method in the present experiment. Both soils were placed in duplicate concrete cylinders (75 cm diam × 60 cm height), kept under field conditions for the first 6 months and then characterized as upland- and paddy-field soils by determining the three-phase composition by the actual-volumetric method (DIK-100, Daiki Rika Co., Ltd., Tokyo); pH; oxidation-reduction potential (Toa Denpa Kogyo Co., Ltd., Tokyo); CN (CN Corder-MTN-500, Yanaco Co., Ltd., Kyoto) and humus contents (Schollenberger, 1945); vegetation and microbiota. Tap water was supplied at regular intervals to upland-field soil. Paddy-field soil was kept constantly flooded. Weeds growing in the soils were identified and removed. During the experiment, no cultivation or fertilization was done.

Oospores of *P. iwayamai* and *P. paddicum* were obtained by culturing in 10% V8-juice supplemented with 500 μg/ml wheat germ oil (Japan Impex Co., Ltd., Tokyo) at 15°C for 3 weeks. Oospores free from all viable propagules were obtained by a freezing method (Takamatsu, 1989). About 5×10^4 oospores were sandwiched between nylon screens (10 μm pore size, square with 20-mm sides, NBC Ind. Co., Ltd., Tokyo) and buried at a depth of 3 cm in both soils in October 1988, just after the soils had been characterized as upland- and paddy-field soils. The same number of nylon screens with killed oospores or without oospores were similarly buried in both soils in duplicate cylinders to serve as controls.

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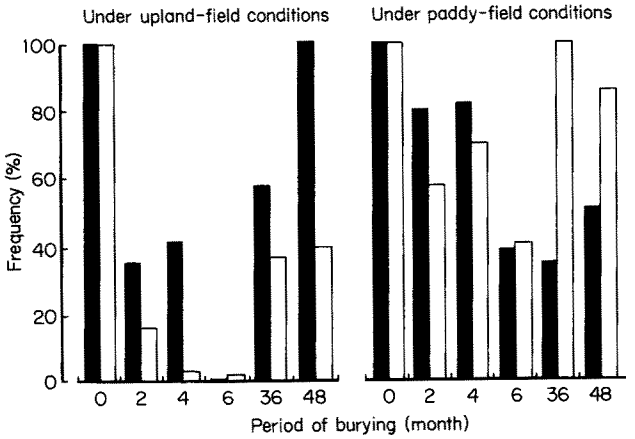

Fig. 1. Frequency of *P. iwayamai* (■) and *P. paddicum* (□) in soils where their oospores had been buried for 48 months under upland- and paddy-field conditions.

Differences in frequency of detection *P. iwayamai* and *P. paddicum* were detected by the baiting method (Takamatsu and Ichitani, 1984). Frequency of detection was expressed as the percentage of leaf pieces from which *P. iwayamai* or *P. paddicum* was isolated to all assayed (20-40) leaf pieces. The results are listed in Fig. 1. The frequencies of both species fell in both soils in the first 6 months after they had been buried. After 36 and 48 months, *P. iwayamai* showed higher frequency than *P. paddicum* in upland-field soil, while *P. paddicum* showed higher frequency than *P. iwayamai* in paddy-field soil. Neither *P. iwayamai* nor *P. paddicum* was isolated from control cylinders during the experiment.

Possible significance of differences in frequency Takamatsu et al. (1987) and Takamatsu (1989) examined mycelial tolerance of CO_2 in the two fungi and found that *P. paddicum*, a paddy-field inhabitant, is more tolerant of CO_2 than *P. iwayamai*, an upland-field inhabitant. Inoue and Ichitani (1990) and Ge and Ichitani (1992) also showed that the optimum pH for mycelial growth and oospore germination of the two fungi differed but was consistent with the soil pH at which each existed. Ge and Ichitani (1992) showed that, in oospore germination, *P. iwayamai* was more tolerant of lower osmotic potential than *P. paddicum*, which ceased to germinate at this low level, indicating adaptation of the fungi to their respective environments. Since *P. paddicum* was less virulent than *P. iwayamai* to wheat, barley and grasses in upland-field soil (Takamatsu, 1989), its population may gradually decrease in this soil as compared with that of *P. iwayamai*.

The higher frequency of *P. iwayamai* than *P. paddicum* in upland-field soil after burying for 36 and 48 months (Fig. 1) indicates that *P. iwayamai* will dominate in upland-field soil, where *P. paddicum* will remain at relatively low level. On the contrary, the higher frequency of *P. paddicum* than *P. iwayamai* in paddy-field soil indicates that *P. paddicum* will dominate in this soil, while *P. iwayamai* will remain at lewer lovel. These indications are also supported by a separate experiment (Ichitani and Takamatsu, unpublished).

The differences in the long-term frequencies of *P. iwayamai* and *P. paddicum* may thus be involved in explaining their different patterns of distribution. Further studies are needed to examine the responses of these two fungi to other factors in natural soil environments.

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