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Fungal successions on pine needles fallen at different seasons: the succession of surface colonizers*

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Field experiments were carried out to investigate influences of seasonal change on the fungal succession occurring on the surface of decaying pine needles at a moder site in Japan. At different seasons, the needles fallen for a short period were collected and marked, then placed on the surface of the O horizon. The needles were removed at intervals and their fungal communities were examined by using a washing technique. Unlike the successions of interior colonizers studied at the same time, those of surface colonizers observed on the fallen needles at four different times are roughly similar to each other. *Thysanophora penicillioides* was the major first colonizer on the sample needles from the O horizon, and *Trichoderma* species followed it. In an experiment started in late autumn, three dematiaceous fungi, *Chloridium viride* var. *chlamydosporis*, *Sporidesmium omahutaense*, and *Chalara* sp., commonly occurred and contributed to the darkening of colonized needles. Seasonal variation in climate may have a stronger effect on internal colonizers than external colonizers of needles.

Key Words——fungal succession; needle decomposition; *Pinus densiflora*; seasonal change; surface needle colonizers.

Seasonal climatic variations in Japan may bring about changes in the composition of early fungal colonizer on/in freshly fallen pine needles from the O horizon because pines usually shed their needles throughout the year.

I have previously described a fungal succession on/in decaying pine needles based on the data of the vertical distribution of mycoflora in the upper sub-layers of the O horizon in a pine forest established on a moder site in Sugadaira, central Japan (Tokumasu, 1996). This indicated that the species colonizing needles fallen in the summer were distinct from those in other seasons at the site.

To study the effects of season on the fungal succession on/in fallen pine needles, field experiments were conducted at the same site and fungal successions started at different seasons were compared. In a twin article (Tokumasu, 1998), the successions of interior colonizers, which were estimated from the results obtained by a surface sterilization method, were summarized and discussed. The succession of interior colonizers varied from season to season and the seasonal shift in the species combination was discussed in relation to climatic and biotic factors. Temperature at the surface of litter appeared to be one of the cardinal factors contributing to these seasonal changes in the fungal succession of internal colonizers.

In this paper, the successions of surface colonizers that were recorded from washed needles, excluding the interior colonizers, are described and discussed.

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Materials and Methods

Descriptions of the study site Investigations were carried out on the campus of the Sugadaira Montane Research Center, University of Tsukuba, which is situated at latitude 36°31′N and longitude 138°21′E, and at an altitude of about 1,330 m above sea level. Climatic data for the experimental period are shown in Fig. 1.

The study site was covered with a stand of 11–12 m height *Pinus densiflora* Sieb. et Zucc. and the canopy was closed. The ground was mostly open except for sparse herbaceous plants and some shrubs and mosses.

Seasonal fluctuation of litter fall Monthly production of leaf litter at the study stand from October 1977 to December 1979 is shown in Fig. 2. About 40 percent of the annual production was recorded from October to December.

Profiles of the O horizon The L and F layers of the O horizon at the site were recognizable from the second half of October to the first half of June, but the typical L layer became indistinct in the rest of the year. The H layer was very thin and indistinct throughout the year. The O horizon is described in detail in Tokumasu (1996).

Field experiments The starting times of field experiments were: middle autumn (MA) series, 15 October 1977; late autumn (LA) series, 22 November 1978; spring (SP) series, 25 April 1992; summer (SU) series, 15 July 1978. The bases of selection of data for the starting times are described in Tokumasu (1998).

At the start of the MA, LA and SU series, sufficient needles for subsequent sampling during the experimental period were collected from a several-day accumulation of

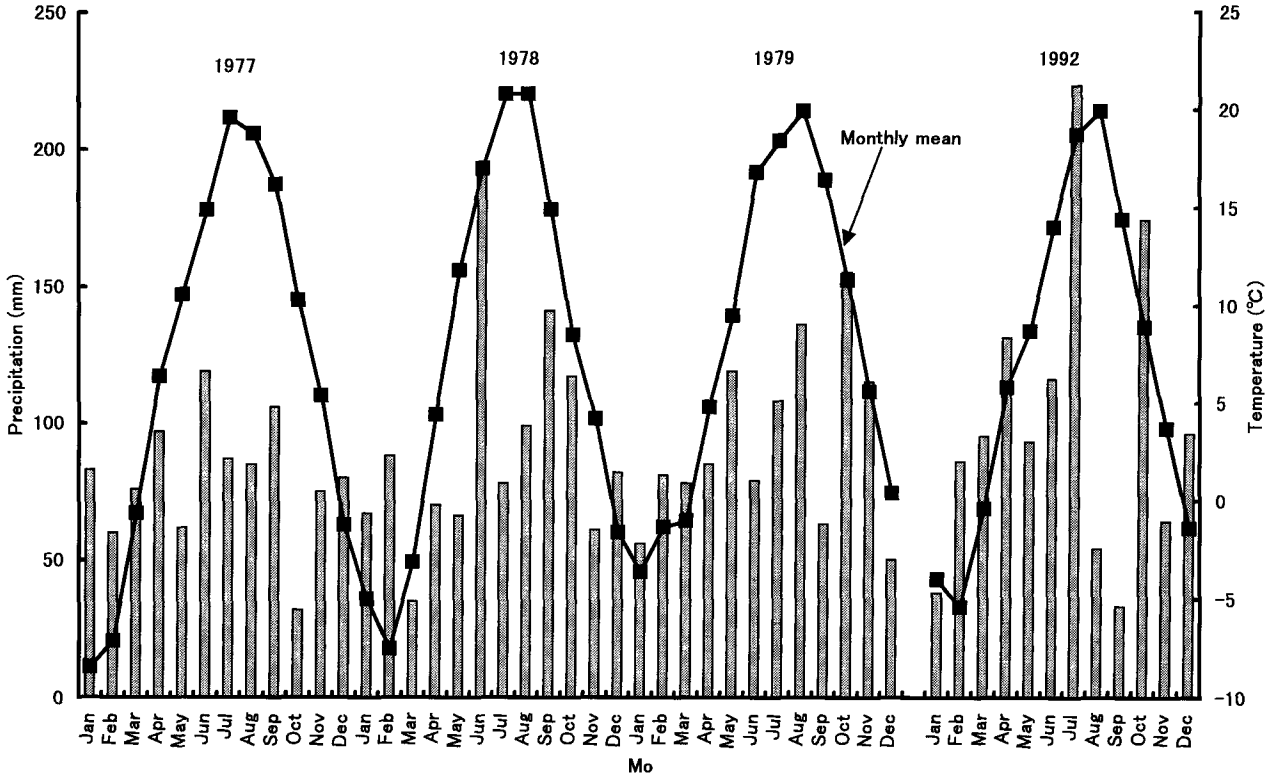

Fig. 1. Monthly mean temperatures and monthly precipitation from 1977 to 1979 and 1992.

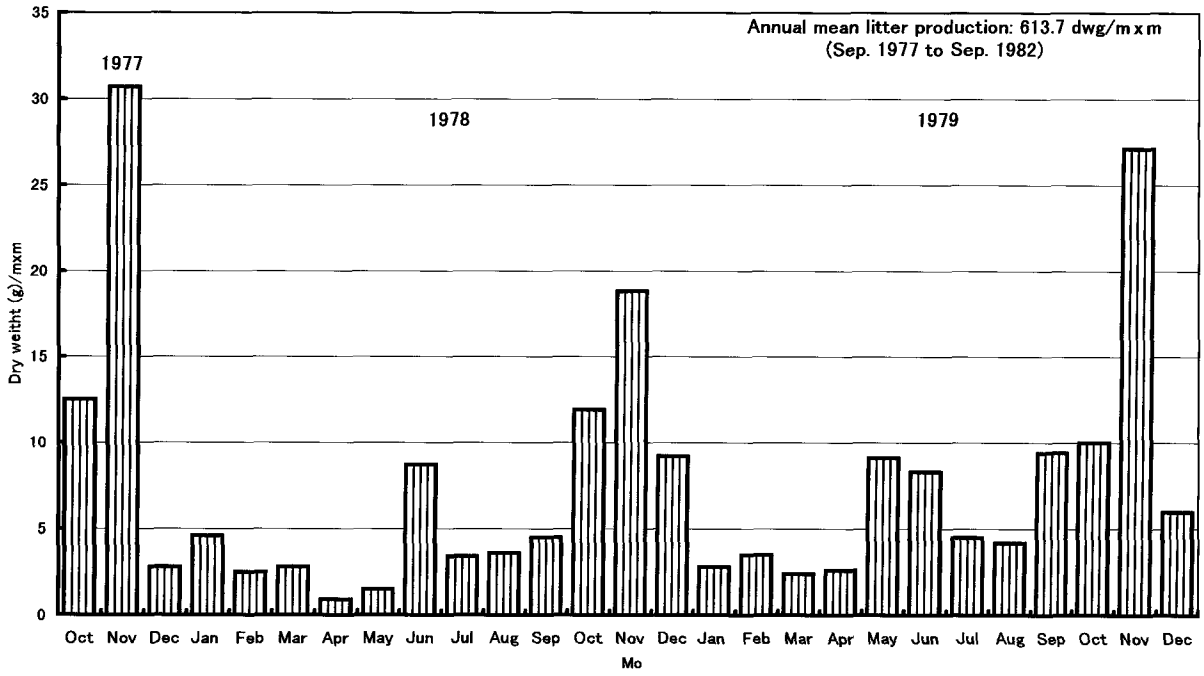

Fig. 2. Monthly amounts of fallen needles from Oct. 1977 to Dec. 1979.

fallen needles in litter traps of 50 cm diam made of butterfly nets. In the case of the SP series the uppermost needles of the L layer were collected. After sorting to remove any obviously damaged needles, twelve needle pairs were attached to a very fine, 1.5 m long polypropylene thread at 10-cm intervals by tying the part of the short branch to the thread. Twenty such threads were prepared for the MA and LA series and 15 for the SP and

SU series. One thread was immediately used to study the mycoflora at the starting point.

For each series, a relatively undisturbed area of 2 m × 2 m was selected in the study site. The threads were placed in parallel at right angles to the slope on the surface of litter at 15-cm intervals and each end was fastened to the base of a thin iron rod 1.5 m high that had been driven into the ground in advance. In this way the needles attached to the threads lay on the litter surface.

In the MA series, samples were collected at short intervals until 2 December 1977, then at irregular longer intervals until August 1978. In the LA series, samples were collected at monthly intervals except when the ground was covered with continuous snow cover. In the SP and SU series, the first two samples were collected at 2-wk intervals and the remainder monthly.

At each collection, a thread was chosen arbitrarily and removed. The material was transported to the laboratory, and 10 of the 12 needle pairs were selected for analysis of fungal community.

Analysis of fungal community To examine fungal colonizers of pine needles, the washing method of Harly and Waid (1955) as modified by Tokumasu (1978, 1980) was adopted. Ten single needles derived from ten needle pairs were washed five times with 10 ml of sterilized 0.005% Aerozol OT solution (di-iso-octyl sodium sulfosuccinate), then rinsed with sterilized water three times in a test-tube using a vortical type shaker. The rinsed needles were transferred to the sterile filter paper in 9-cm Petri dish and dried for 1 d to suppress vigorous bacterial growth after plating (Widden and Parkinson, 1973). Five sets of two needles were placed on the surface of half-strength cornmeal agar (Difco) plates.

All the treatments were performed within 3 h after the removal of sample needles.

The plates were incubated in the laboratory under the fluctuating light and temperature conditions of night and day. The incubated plates were observed microscopically at least four times at 1-wk intervals. Sporulated fungi were isolated onto 2.5% malt extract agar slants. Identification of isolates was attempted to species level. A rhizomorph-forming fungus isolated from aerial strands is here described as a rhizomorph-forming fungus.

Percentage frequency of occurrence for each species at a sampling time was calculated as follows: number of needles bearing a specified fungus/10 × 100.

Results

MP series Eight species occurred on intact needles (Table 1). According to the ecological grouping of fungi by Hudson (1968), the fungi with relatively high frequency values belong to the common primary saprophytes. They are *Cladosporium cladosporioides* (Fres.) de Vries, *Aureobasdium pullulans* (de Bary) Arnaud, and *Alternaria alternata* (Fr.) Keissler. *Cladosporium cladosporioides* declined gradually over a long period, but the other species did so quickly after the sample was laid on the litter surface and had mostly disappeared from the sample needles removed from the following spring.

The first surface colonizer of freshly fallen needles from the O horizon was *Thysanophora penicillioides* (Roum.) Kendrick. The fungus reached the sample needles within 1 wk after setting and established itself, but it did not occur on over-wintered needles. *Trichoderma koningii* Oudem. quickly followed the first colonizer, gradually increased, and finally occurred on all the sample needles in August 1978. The other fungi listed in Table 1 are members of soil fungi, viz., *Verticillium psalliotae* Treschow, *Mortierella isabellina* Oudem., *M. verticillata* Linnem., *M. ramanniana* (Möller) Linnem., and *Penicillium citrinum* Thom. They were major components of the microfungal community after the thaw.

Table 1. Percentage frequencies of the fungi with a frequency of 50% or more at some sampling time in MA series.

Yr	1977							1978			
Sampling mo	Oct	Oct	Oct	Nov	Nov	Nov	Dec	Jan	May	Jul	Aug
Time after exposure of litter (d)	0	10	17	24	31	38	52	89	215	268	307
Fungal species											
Cladosporium cladosporioides	100	100	100	100	100	100	100	80	80	50	50
Alternaria alternata	40	50	60	40	10	20	30	10	10	0	0
Thysanophora penicillioides	0	30	10	10	0	0	70	20	0	0	0
Trichoderma koningii	0	0	0	10	50	20	50	10	50	40	100
Verticillium psalliotae	0	0	0	0	20	0	0	0	30	50	10
Mortierella isabellina	0	0	0	0	10	10	30	30	40	0	50
M. verticillata	0	0	0	0	0	0	20	60	20	50	10
M. ramanniana	0	0	0	0	0	0	10	0	0	0	50
Penicillium citrinum	0	0	0	0	0	0	0	0	10	50	0
Selenosporella curvispora	0	0	0	0	0	0	0	0	30	30	70
Total no. of species recorded	8	7	12	18	18	12	15	15	16	18	15

Selenosporella curvispora MacGarvie, the dominant interior colonizer at the site (Tokumasu, 1996), was recorded from over-wintered needles.

The total number of species recorded in this series was 47.

LA series Table 2 shows the results of the LA series. Two common primary saprophytes with a frequency of 50% or more were recorded, *A. pullulans* and *C. cladosporioides*, while species with moderate or low frequencies included *A. alternata*, *Epicoccum nigrum* Link ex Link and *Nigrospora oryzae* (Berk. et Br.) Petch. The prominent common primary saprophytes gradually decreased in frequency as time passed.

The first surface colonizer from the litter was *Trichoderma longipilis* Bissett in this series. *Thysanophora penicillioides* was first recorded in the following July. Characteristic dematiaceous fungi were recorded from the last quarter of the experimental period. They were *Chloridium viride* var. *chlamydosporis* (van Beyma) W. Gams, *Sporidesmium omahutaense* Matsushima, and *Chalara* sp. They formed a dematiaceous hyphal network on the needle surface and contributed to the darkening of colonized needles. *Trichoderma koningii* first occurred in July as the dominant and maintained a high frequency to the end. This species was also recorded as an interior colonizer in this series (Tokumasu, 1998). *Selenosporella curvispora* and a rhizomoroh-forming fungus, possibly a species of leaf litter decomposing agaric, were superior interior colonizers in this series (Tokumasu, 1998).

The total number of fungi recorded during the experimental period was again 47.

SP series Table 3 shows results of the SP series. This series was begun almost 15 yr after the start of the SU

Table 2. Percentage frequencies of the fungi with a frequency of 50% or more at some sampling time in LA series.

Yr	1978		1979							
Sampling mo	Nov	Dec	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov
Time after exposure of litter (mo)	0	1	5	6	7	8	9	10	11	12
Fungal species										
Aureobasidium pullulans	80	100	0	0	20	20	30	30	0	0
Cladosporium cladosporioides	80	90	60	30	80	70	40	30	40	10
Trichoderma longipilis	0	50	30	20	0	10	0	20	10	0
Thysanophora penicillioides	0	0	0	0	0	20	0	30	60	10
Chloridium viride var. *chlamydosporis*	0	0	0	0	0	0	40	50	70	0
Sporidesmium omahutaense	0	0	0	0	0	0	0	10	90	40
Chalara sp.		0	0	0	0	0	0	0	20	60
Selenosporella curvispora	0	0	70	60	50	80	40	70	20	0
Rhizopmorph-forming fungus	0	0	10	10	10	0	60	20	60	70
Trichoderma koningii	0	0	0	0	0	80	90	80	80	100
Total no. of species isolated	11	11	12	80	13	14	19	14	21	14

Table 3. Percentage frequencies of the fungi with a frequency of 50% or more at some sampling time in SP series.

Yr	1992									
Sampling mo	Apr	May	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec
Time after exposure of litter (mo)	0	0.5	1	2	3	4	5	6	7	8
Fungal species										
Aureobasidium pullulans	100	100	60	60	90	10	10	10	20	10
Cladosporium cladosporioides	80	20	40	70	0	10	70	100	70	70
Thysanophora penicillioides	0	0	10	60	0	20	80	90	70	90
Septonema ochracea	0	0	30	0	0	0	20	60	40	40
Mortierella isabellina	0	0	0	0	30	50	40	50	40	90
M. ramanniana	0	0	0	0	0	30	20	60	10	30
Penicillium brevicompactum	0	0	0	0	0	10	60	20	40	30
Trichoderma polysporum	0	0	0	0	0	0	20	20	40	60
Rhizomorph-forming fungus	0	0	0	10	10	30	0	60	60	80
Total no. of species isolated	14	15	8	12	10	12	19	21	17	23

series.

The major common primary saprophytes of this series were the same as those of the LA series. *Aureobasidium pullulans* declined rapidly and almost disappeared after August. *Cladosporium cladosporioides*, like *A. pullulans* declined temporarily, but revived after September.

Thysanophora penicillioides was the first invader to the sample needles from the O horizon. *Trichoderma polysporum* (Link ex Pers.) Rifai and a dematiaceous fungus, *Septonema ochracea* Matsushima, followed *T. penicillioides*. The common later colonizers were soil fungi, viz., *M. isabellina*, *M. rammnaniana* and *Penicillium brevicompactum* Dierckx.

In this series the rhizomorph-forming fungus and *S. curvispora* were the prevailing interior colonizers (Tokumasu, 1998), but the latter was infrequent as a result of the washing method.

The number of fungi recorded in this series reached 56.

SU series A total of 39 species were recorded in this series. As shown in Table 4, 12 species were recorded from intact needles, but only 5 of those shown in Table 4 were recorded from the sample removed after 2 wk. They were members of the common primary saprophytes and their frequencies dwindled with passage of time, except for *C. cladosporioides*, which showed high frequencies in November and December.

The first surface colonizers on the needles lying on the litter were *T. penicillioides* and two *Trichoderma* species, *T. longipilis* and *T. koningii*. These species were only infrequently recorded from the surface-sterilized needles in this series (Tokumasu, 1998).

Two prevailing interior colonizers of this series, *Chaetopsina fulva* Rambelli and *Verticicladium trifidum* Preuss, continued to occur on the sample needles until the final sampling.

Based on the above results, the successions of surface needle colonizers observed on pine needles fallen at different seasons are summarized in Table 5.

Discussion

The primary fungi colonizing the surface of senescent or dead needles on the tree belonged to the group of "common primary saprophytes" defined by Hudson (1968). He noted that most of the common primary saprophytes are absent from pine needles, but at this site the saprophytes were very diverse, as reported earlier (Tokumasu, 1996). Many of the species occurred with 50% or higher frequency on intact needles at every setting, though the species combination changed. This suggests that there is a set of common primary saprophytes in this area, and the species that occurred on intact needles with high frequencies were those that were better adapted to the climatic conditions immediately before the setting of sample needles. Among them, *C. cladosporioides* was the most frequent species at this site and appeared to be adaptable to a wide range of climatic variation.

In all series *T. penicillioides* was recorded as one of the early colonizers (Table 5). In the previous study (Tokumasu, 1996), this species was reported as mainly colonizing summer fallen needles, but it could colonize freshly fallen needles in various seasons and over-wintered needles (Tables 1–4). It has been suggested that the substratum preference of this fungus is for fir needles rather than pine needles (Ellis and Ellis, 1985). In fact, this species has been recorded from freshly fallen fir needles in high frequency (Gourbière, 1979; Aoki et al., 1990, 1992), although it was also recorded from pine needles (Tokumasu et al., 1994). The results of the present study suggest that the species has as high a preference for freshly fallen pine needles as it does for fir

Table 4. Percentage frequencies of the fungi with a frequency of 50% or more at some sampling time in SU series.

Yr	1979						
Sampling mo	Jul	Jul	Aug	Sep	Oct	Nov	Dec
Time after exposure of litter (mo)	0	0.5	1	2	3	4	5
Fungal species							
Aureobasidium pullulans	70	80	90	30	20	0	30
Cladosporium herbarum	70	20	0	0	60	0	0
C. cladosporioides	50	20	40	80	20	100	60
Alternaria alternata	30	50	60	10	30	10	10
Epicoccum nigrum	30	60	30	20	10	0	0
Thysanophora penicillioides	0	10	20	90	100	90	80
Trichoderma koningii	0	10	20	70	30	60	90
T. longipilis	0	10	0	30	70	40	20
Chaetopsina fulva	0	90	80	80	80	60	70
Verticicladium trifidum	0	30	50	40	30	50	20
Total no. of species isolated	12	13	16	19	19	15	16

Table 5. Successions of surface colonizers observed on pine needles fallen at different seasons.

Season of needle fall	Mid autumn	Late autumn	Spring	Summer
On the tree				
Primary colonizers	*Cladosporium cladosporioides* *Alternaria alternata* *Aureobasidium pullulans*	*A. pullulans* *C. cladosporioides*	*A. pullulans* *C. cladosporioides*	*A. pullulans* *Cladosporium herbarum* *C. cladosporioides* *A. alternata* *Epicoccum nigrum*
On litter				
Secondary colonizers (First wave)	*Thysanophora penicilioides* *Trichoderma koningii*	*Trichoderma longipilis*	*T. penicillioides* *Septonema ochracea*	*T. penicillioides* *T. koningii* *T. longipilis*
In litter				
Secondary colonizers (Second and third waves)	*Verticillium psalliotae* *Mortierella isabellina* *Mortierella verticillata* *Mortierella ramanniana* *Penicillium citrinum*	*T. penicillioides* *T. koningii* *Chloridium viride* var. *chlamydosporis* *Sporidesmium omahutaense* *Chalara* sp.	*M. isabellina* *M. ramanniana* *Penicillium brevicompactum* *Trichoderma polysporum*	
Monthly mean air temperature of the starting mo (°C)	9.4	3.0	5.5	18.1
Mean daily air temperature during 10 d after setting (°C)	7.8	1.3	8.0	16.8

needles.

Three *Trichoderma* species, *T. koningii*, *T. longipilis*, and *T. polysporum*, formed a group of common colonizers from the litter at this site. The former two species occurred in all four series and the latter occurred in all but the SU series.

These occurrence patterns of *Trichoderma* species are compatible with the conclusions drawn from a series of works on the distribution and competitive ability of *Trichoderma* species by Widden and coworkers (Widden 1984; Widden and Hsu, 1987; Widden and Scattolin, 1988). Thus, *T. polysporum* and *T. viride* Pers.: Fr. may colonize newly available substrata such as fallen spruce needles during the cooler months but in the warmer months they may be replaced by better competitive species, viz., *T. hamatum* (Bon.) Bainier and *T. koningii*, and may not colonize such substrata. Expect for the SP series, *T. koningii* was a later colonizer (Tables 1, 2, 4) and seemed to be more aggressive than other *Trichoderma* species. In the SP series, *T. polysporum* was more common in the cooler months (Table 3) and *T. koningii*, which is not listed in Table 3, mainly occurred on 30 or 40% of sample needles from August to November. In addition, *T. viride* was also recorded in November and December in the same series, though its frequency values were small. The occurrence patterns observed in the SP series also appeared to be explained by the conclusions of Widden and coworkers (Widden, 1984; Widden and Hsu, 1987; Widden and Scattolin, 1988). Thus, *T. polysporum* and *T. viride* are relatively less competitive species and prefer lower temperatures, while *T. koningii* is a better competitor and prefers to warmer conditions.

In the LA series, three dematiaceous fungi were found on the sample needles removed in the autumn, which caused darkening of colonized needles by production of a dematiaceous hyphal network on the needle surface. Dematiaceous species that play a similar role have been recorded in the U.K. and Japan: *Sympodiella acicola* Kendrick (Kendrick and Burges, 1962), *Tropospora monospora* (Kendrick) M. B. Ellis (Kendrick and Burges, 1962; Parkinson and Balasooriya, 1967; Lehmann and Hudson, 1977), and *Kriegeriella mirabilis* Höhnel (Mitchell and Millar, 1978) in the U.K., and *Sporidesmium goidanichii* (Rambelli) S. Hughes (Tubaki and Saitô, 1969; Soma and Saitô, 1979), *Anungitea continua* Matsushima (Tokumasu, 1978), and *K. mirabilis* (Soma and Saitô, 1979) in Japan. In the other series, dematiaceous surface colonizers occurred only sporadically and may little contribute to needle darkening. Internal colonizers, however, often caused darkening of the needle surface; at this site *V. trifidum*, as noted by Lehmann and Hudson (1977) and *S. curvispora* caused darkening. Consequently, most fallen needles at this site become dark with 1 yr. Although the role of dark pigmentation of the needle surface in the decomposition is not clear, Lehmann and Hudson (1977) suggested that the dark pigments, melanin, may protect the surface needle dwellers from ultraviolet radiation, microbial lysis, and desiccation.

The successions of surface colonizers observed on the fallen needles at four different times were broadly similar to each other. *Thysanophora penicillioides* and *Trichoderma* species were early colonizers of sample needles in all series. In contrast, the first internal colonizers from the O horizon changed rapidly at the same site

(Tokumasu, 1998). Thus, seasonal variation may have a stronger effect on internal colonizers than on external colonizers of needles.

Many species of common soil fungi were recorded from the sample needles, the major ones being *Mortierella* and *Penicillium* species. On the whole, soil fungi tended to be more abundant on the sample needles removed in the second half of the experimental period than in the first half. This may reflect the fact that mild conditions in the sub-surface layer are suited for the growth of these fungi, since fallen needles buried the sample needles lying on the surface. Their occurrence patterns may also be strongly influenced by a drastic or uneven climatic event, such as disturbance by heavy rainfall or a short drought, which often occur in Japan.

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Literature cited

Aoki, T., Tokumasu, S. and Oberwinkler, F. 1992. Fungal succession on fir needles in Germany. Trans. Mycol. Soc. Japan **33**: 359–374.

Aoki, T., Tokumasu, S. and Tubaki, K. 1990. Fungal succession on momi fir needles. Trans. Mycol. Soc. Japan **31**: 355–374.

Ellis, M. B. and Ellis, J. P. 1985. Microfungi on land plants, p. 76. Croo Helm, London.

Gourbière, F. 1979. Les champignons microscopiques liés aus aiguilles de sapin (*Abies alba* Mill.). 4. Microflore de la littère. Bull. Soc. Mycol. Fr. **91**: 429–441.

Harley, J. L. and Waid, J. S. 1955. A method of studying active mycelia on living roots and other surfaces in the soil. Trans. Br. Mycol. Soc. **38**: 104–118.

Hudson, H. J. 1968. The ecology of fungi on plant remains above the soil. New Phytol. **67**: 837–874.

Kendrick, W. B. and Burges, A. 1962. Biological aspects of the decay of *Pinus sylvestris* leaf litter. Nova Hedwigia **4**: 313–342.

Lehmann, P. F. and Hudson, H. J. 1977. The fungal succession on normal and urea-treated pine needles. Trans. Br. Mycol. Soc. **68**: 221–228.

Mitchell, C. P. and Millar, C. S. 1978. Mycofloral succession on Corsican pine needles colonized on the tree by three different fungi. Trans. Br. Mycol. Soc. **71**: 303–317.

Parkinson, D. and Balasooriya, I. 1967. Studies on fungi in a pine-wood soil. I. Nature and distribution of fungi in the different soil horizons. Rev. Écol. Biol. Sol. **4**: 463–478.

Soma, K. and Saitô, T. 1979. Ecological studies of soil organisms with references to the decomposition of pine needles I. – Soil macrofaunal and mycofloral surveys in coastal pine plantations. Rev. Écol. Biol. Sol. **16**: 337–354.

Tokumasu, S. 1978. Leaf litter fungi of the forests of *Pinus densiflora* and four introduced pines at Sugadaira, central Japan. Trans. Mycol. Soc. Japan **19**: 383–390. (In Japanese.)

Tokumasu, S. 1980. Observations on the fungal flora in the pine leaf litter. In: Biseibutunoseitai, vol. 7, (ed. by Biseibutsuseitaikenkyukai), pp. 129–144. Gakkai Shuppan Center, Tokyo. (In Japanese.)

Tokumasu, S. 1996. Mycofloral succession on *Pinus densiflora* needles on a moder site. Mycoscience **37**: 313–321.

Tokumasu, S. 1998. Fungal successions on pine needles fallen at different seasons: the succession of surface colonizers. Mycoscience **39**: 409–416.

Tokumasu, S., Aoki, T. and Oberwinkler, F. 1994. Fungal succession on pine needles in Germany. Mycoscience **35**: 29–37.

Tubaki, K. and Saitô, T. 1969. *Endophragmia alternata* sp. nov. and other Hyphomycetes on *Pinus* leaves in Japan. Trans. Br. Mycol. Soc. **52**: 477–482.

Widden, P. 1984. The effects of temperature on competition for spruce needles among sympatric species of *Trichoderma*. Mycologia **76**: 873–883.

Widden, P. and Hsu, D. 1987. The effects of temperature and litter types on competition between *Trichoderma* species. Soil Biol. Biochem. **19**: 89–93.

Widden, P. and Parkinson, D. 1973. Fungi from coniferous forest soils. Can. J. Bot. **51**: 2275–2290.

Widden, P. and Scattolin, V. 1988. Competitive interactions and ecological strategies of *Trichoderma* species colonizing spruce litter. Mycologia **80**: 795–803.