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Changes in organic matter composition of forest soil treated with a large amount of urea to promote ammonia fungi and the abilities of these fungi to decompose organic matter

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Organic matter composition (lignin, holocellulose, 50% (v/v) methanol extract, water-soluble carbohydrate (WSC) and phenolics (WSP), petroleum ether extract, and ash) of A_0 layer soil treated with 700 g/m^2 of urea to promote ammonia fungi was investigated in a Japanese red pine (*Pinus densiflora*) forest. Nine species of fungi were found during the study period of 18 months after the treatment. Of these, seven species belong to the ammonia fungi. WSC content of the treated soil was lower than that of the control. Methanol extract content increased initially after the treatment, then decreased to below the control level. There were no consistent differences in other components between the treated plot and the control. The abilities to decompose cellulose, lignin, chitin, protein and lipid in 18 strains in 10 species of the ammonia fungi were also screened. Cellulose was not lysed by *Pseudombrophila deerata, Hebeloma* spp. and *Laccaria bicolor*. Strong lignolytic activity was shown by *Lyophyllum tylicolor, Coprinus echinosporus* and *P. deerata*. Chitin was decomposed by *Amblyosporium botrytis, L. tylicolor, C. echinosporus* and *Hebeloma vinosophyllum*. All strains possessed proteolytic and lipolytic activities. Supply of glucose to the culture media resulted in weaker enzyme activities except for lignolytic ability.

Key Words——ammonia fungi; enzymatic activity; forest soil; glucose; organic matter composition.

Introduction

"Ammonia fungi" (Sagara, 1975), a chemoecological group of fungi, develop sporophores sequentially on forest soil that has been treated with such nitrogenous materials as urea and ammonia. Naturally, these fungi occur after the decomposition of the bodies or faeces of animals, or on other substrata (Sagara, 1992).

After such treatment, litter turns black, ammonia concentration increases, raising soil pH to 8–10, and water content also increases (Sagara, 1975; Suzuki, 1989). Under these conditions, bacteria and nematodes increase and influence nitrogen flow in the soil (Yamanaka, 1993). Through development of these organisms, soil organic matter is also expected to change quantitatively and qualitatively, and the ammonia fungi should adapt to such changes. Suzuki (1992) and Enokibara et al. (1993) reported some of the ammonia fungi possessed higher cellulolytic ability under alkaline condition. In this study, changes in organic matter composition of the forest soil on which the ammonia fungi occur were investigated, and the abilities of the ammonia fungi to decompose five types of organic matter were tested.

Materials and Methods

Site description The study was carried out in a 45-year-old Japanese red pine (*Pinus densiflora* Sieb. et Zucc.) forest at Kamigamo Experimental Station of Kyoto University Forest (35°04′N and 135°43′E; 120–140 m above sea level), about 12 km north of the Kyoto City, Japan. In 1990, air temperature at the station ranged from 0.8°C in January to 28.0°C in August, and annual rainfall was 1549 mm. The parent rock of this area consists of Paleozoic sandstone and slate. The soil is a dry brown forest soil, and the soil surface was covered with A_0 layer (5 to 11 cm in thick) in which three zones, the L, F and H layers, were recognized.

Treatment and sampling In the forest, seven plots, each 200 cm long and 50 cm wide, were established. To promote ammonia fungi, four of the plots were treated with urea by hand-scattering at the rate of 700 g/m^2 on 19 April 1990; the others were taken as the controls. After the treatment, fungal occurrence was observed. Soil samples were collected separately from the F and H layers in three of the urea-treated plots and the three control plots. Soil was also collected from the control plots on the day of treatment. The samples were transported to the laboratory and kept at 4°C in a refrigerator if not subjected immediately to analysis. Some soil properties (pH, water content, ammonium and nitrate concentrations, and the numbers of bacteria and nematodes) were surveyed as reported previously (Yamanaka, 1993).

Determination of organic matter composition The proce-

dures of Schlesinger and Hasey (1981) were adopted to analyze organic matter components of lignin, holocellulose, 50% (v/v) methanol extract, water-soluble carbohydrates (WSC) and phenolics (WSP), petroleum ether extract and ash in soil samples. After removal of plant roots, samples were oven-dried at 50°C for 24 h, then ground to pass a sieve (<0.3 mm). For each of the organic component analyses described below, approximately 0.5 g of the ground sample was employed. The samples were initially immersed in 10 ml of petroleum ether for 24 h at 40°C. After filtration and evaporation of solvent, the weight of the extract was measured. The ether-insoluble residue was then suspended in 10 ml of 50% methanol at room temperature for 24 h, and the methanol-insoluble residue was weighed. The filtrate was used for determining the concentration of WSC and WSP. WSC concentration was measured colorimetrically using the anthrone reaction and known concentrations of D-glucose as standard (Morris, 1948). WSP concentration was analyzed by the Folin-Denis method, using standards prepared with tannic acid (King and Heath 1967). The residue from 50% methanol extraction was employed for determining the amounts of holocellulose and lignin, after the method of King and Heath (1967). Ash content was also determined using approximately 0.5 g of the ground sample after heating at 550°C for 3 h. All data were measured separately for the F and H layers. Standard error of the mean was calculated for data on each sampling day. Differences between layers and between urea-treated and untreated samples were examined using Student's t-test after analysis of variance.

Screening of abilities to decompose organic matters

The following 18 strains from 10 species of the ammonia fungi were tested for their abilities to decompose organic matters: *Amblyosporium botrytis* Fres. (202 and 220), *Ascobolus denudatus* Fr. (152 and 222), *Peziza moravecii* (Svrček) Donadini (185), *Pseudombrophila deerata* (Karst.) Seaver (226 and 237), *Peziza* sp. (165 and 206), *Lyophyllum tylicolor* (Fr.: Fr.) Lange and Sivertsen (187 and 212), *Coprinus echinosporus* Buller (179 and 215), *Hebeloma vinosophyllum* Hongo (110 and 135), *Hebeloma* sp. (9 and 242) and *Laccaria bicolor* (Maire) Orton (252). The ammonia fungi are divided into two groups, early-stage and late-stage (Sagara, 1975). Of the species used, *A. botrytis, A. denudatus, P. moravecii, P. deerata, Peziza* sp., *L. tylicolor* and *C. echinosporus* are early-stage species; and *H. vinosophyllum, Hebeloma* sp. and *L. bicolor* are late-stage species. These strains were obtained from spores or tissues of fruit-bodies occurring on forest soil after urea treatment.

Substrates used for checking decomposition abilities were cellulose, lignin, chitin, protein and lipid. To examine the effect of readily assimilable carbon sources on the pattern of enzymatic activity, agar media with or without added glucose (1%, w/v) were prepared. Unless otherwise stated, all media were poured into 12 ml screw vials. Two layers of agar medium were made in the vial for the test of cellulolytic and chitinolytic activity. The upper layer contained basal medium with substrate for the test described below, and the lower one contained only basal medium.

The decomposition of cellulose was tested using cellulose azure (Sigma, Co.) after the method of Smith (1977). The basal medium consisted of 2 g of $NH_4H_2PO_4$, 0.6 g of KH_2PO_4, 0.4 g of K_2HPO_4, 0.5 g of $MgSO_4 \cdot 7H_2O$, 1 g of yeast extract (Difco), 55 mg of $CaCl_2$, 1 ml of trace metal stock solution (TMSS), 3 ml of vitamin stock solution (VSS), 12 g of agar and 996 ml of distilled water. TMSS contained 440 mg of $ZnSO_4 \cdot 7H_2O$, 500 mg of $MnSO_4 \cdot 4H_2O$, 100 mg of $CuSO_4 \cdot 5H_2O$, 500 mg of ferric citrate, 50 mg of $Na_2MoO_4 \cdot 2H_2O$ and 10 mg of $Na_2B_4O_7$ in 100 ml of distilled water. VSS contained 100 mg of thiamine, 10 mg of folic acid, 10 mg of nicotinic acid and 100 mg of inositol in 100 ml of distilled water. Cultures were inoculated on the surface of the medium and incubated at 20°C for 4 wk. Tubes were checked for dye release by examining the basal layer for blue coloration.

Lignin degradation was checked by the presence of laccase, which is associated with lignin decomposition (Ander and Eriksson, 1976). Culture medium containing 0.2% (w/v) yeast extract and 1.2% agar was prepared in 9-cm plastic petri dishes. Culture plates were inoculated and incubated at 20°C for 2 wk, after which a solution of 0.1% (w/v) syringaldazine in 95% (v/v) ethanol was dropped on the colony (Harkin and Obst, 1973). The presence of the enzyme was determined by the appearance of a violet coloration.

Chitin decomposition was assayed by the method of Hankin and Anagnostakis (1975). Culture medium contained 2 g of KH_2PO_4, 1 g of $(NH_4)_2SO_4$, 3 g of Na_2HPO_4, 0.1 g of $FeSO_4 \cdot 7H_2O$, 0.5 mg of $CaCl_2$, 0.2 g of yeast extract, 1 ml of TMSS, 3 ml of VSS, 12 g of agar and 996 ml of distilled water. Chitin was purified as follows before mixing with other constituents. Chitin powder (Sigma Co.) was dissolved in 50% (v/v) H_2SO_4, then quickly poured into 15 volumes of distilled water. The precipitated chitin was collected by centrifugation. Washing and centrifugation were continued until the acid was removed. After determination of the dry weight of the washed chitin, a 1% (w/v) solution of chitin in the basal medium was prepared and placed in vials. The vials were inoculated and incubated at 20°C for 4 wk, then the depth of the clear zone in the vials was measured.

Proteolytic activity was assayed using gelatin. Culture medium consisted of 3 g of beef extract (Difco), 5 g of Bacto-peptone (Difco) and 4 g of gelatin (Difco) in 1000 ml of distilled water. Cultures were incubated at 20°C for 2 wk, then the depth of liquefaction in the vials was measured.

Lipolytic activity was checked using the procedure of Carroll and Petrini (1983). The following medium was prepared: 2 g of KH_2PO_4, 1.4 g of $(NH_4)_2SO_4$, 0.3 g of $CaCl_2$, 0.3 g of $MgSO_4 \cdot 7H_2O$, 1 g of yeast extract (Difco), 10 g of Tween 40, 1 ml of TMSS, 3 ml of VSS, 12 g of agar and 996 ml of distilled water. After incubation of cultures at 20°C for 4 wk, the depth of clouding was measured in each vial. The clouding is due to the formation of crystals of the calcium salt of the lauric acid

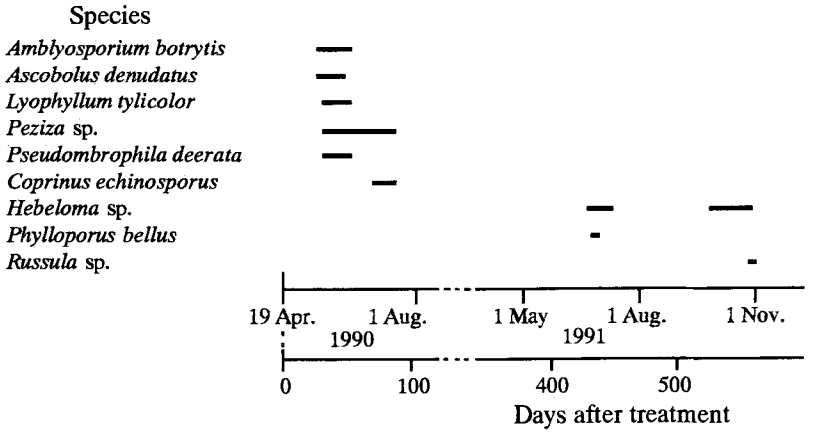

Fig. 1. Occurrence of the fungi on forest soil after treatment with 700 g/m^2 of urea on 19 April 1990.

liberated by the enzyme (Hankin and Anagnostakis, 1975). When the salts are completely degraded, a clear zone is observed around a colony.

The pH of the medium were adjusted to 8 before autoclaving at 121°C for 20 min. To avoid precipitation, potassium salts, calcium chloride, magnesium salts, TMSS and VSS were separately filter-sterilized with a 0.45 μm millipore filter, then mixed into the molten agar medium. Glucose solution was also filter-sterilized before addition. Duplicated cultures were prepared for each test and repeated twice.

Results of the test were scored subjectively on the basis of relative enzyme activity, as follows: (−)=no activity; (±)=suggestion of activity, but questionable; (+)=slight but definite activity; (++)=intermediate activity; (+++)=intense activity.

Results

Occurrence of fungi and some soil properties after urea treatment As Fig. 1 shows, nine species of fungi appeared successively after the urea treatment: *A. botrytis, A. denudatus, P. deerata, Peziza* sp., *L. tylicolor, C. echinosporus, Hebeloma* sp., *Phylloporus bellus* (Mass.) Corner and *Russula* sp. All were absent in the control plot, and all except *P. bellus* and *Russula* sp. have been reported to be ammonia fungi (Sagara, 1992).

Changes in soil properties (pH, water content, ammonia and nitrate concentrations, and the number of bacteria and nematodes) after treatment are shown in Table 1.

Changes in organic matter composition after urea treatment Differences between the F and H layers were found in the contents of ash and WSP (Figs. 2, 3b; Table 2). Ash content in the F layer was 5.9-8.9% in the control and 5.0-14.2% in the treated soil, while that in the H layer was 8.2-24.6% in the control and 9.6-29.7% in the treated soil. The difference in WSP content between the two layers was significant only in the control: contents were 0.246-0.471% in the F layer and 0.117-0.214% in the H layer.

Methanol extract, WSC and WSP contents differed significantly between urea-treated and control plots (Figs. 2, 3; Table 2). After treatment, the methanol extract increased to 11.5% in the F layer and to 11.2% in the H layer on 6 June 1990, while the control was 7.0% in the F layer and the 6.0% in the H layer. These subsequently decreased to 5.1% in the F layer and to 3.6% in the H layer on 20 February 1991, at which time the control was 8.1% in the F layer and 5.4% in the H layer. The concentrations of WSC and WSP in the treated soil were lower than those of the control. WSC of the treated soil was 0.19-0.38% in the F layer and 0.17-0.40%

Table 1. Changes in some soil properties after treatment with 700 g/m^2 urea on 19 April 1990.

Date	25 April 1990-17 July 1990[a]				15 Nov 1990-21 Nov 1991[b]			
	Treated		Control		Treated		Control	
	F layer	H layer	F layer	H layer	F layer	H layer	F layer	H layer
pH	8.9-7.8	8.6-7.8	4.1-3.7	3.5-3.8	4.6-4.1	4.5-4.0	4.1-3.6	4.0-3.6
Water content (%)	86.0-79.0	87.3-77.7	76.7-69.7	77.0-70.7	83.5-53.2	83.0-50.7	74.3-63.8	66.3-61.6
NH_4^+-N (μgN/g dry soil)	14000-7800	16000-6200	42-22	30-19	490-19	650-32	47-6.3	47-19
NO_3^--N (μgN/g dry soil)	11-5.9	12-5.8	5.5-3.3	5.3-3.6	170-1.6	300-3.0	2.6-1.5	1.7-1.1
Bacteria ($\times 10^8$ colonies/g dry soil)	1100-0.46	1500-1.5	7.8-2.5	4.4-3.2	130-4.6	370-4.9	3.8-2.4	2.0-1.6
Nematodes (numbers/g dry soil)	3300-0	3700-87	500-180	510-220	110-91	71-45	330-140	320-120

a, b; Results are the ranges of data determined in the study period, during which soil sampling was repeated (a) 4-5 times, or (b) 3 times.

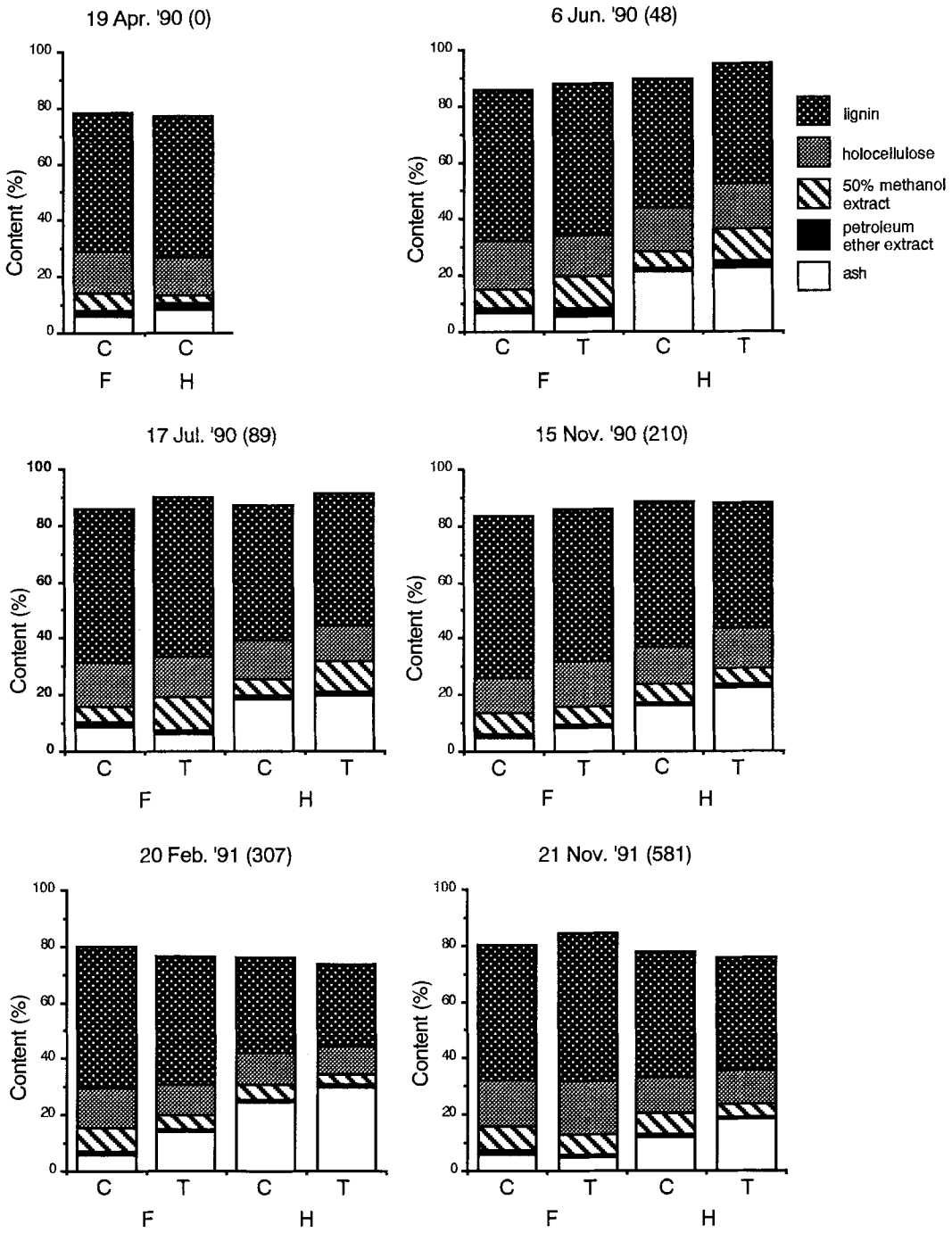

Fig. 2. Changes in organic matter composition of F and H layer soil after treatment with 700 g/m^2 of urea on 19 April 1990. F, F layer; H, H layer; C, control; T, treated. Values are percentages of the weight of soil oven-dried at 50°C for 24 h. Number in parentheses after sampling date shows days after treatment.

in the H layer, while those of the control were respectively was 0.60-0.92% and 0.38-0.68%. WSP content was decreased only in the F layer: that of the treated soil was 0.08-0.26% and that of the control was 0.25-0.47%. Contents of other types of the organic matter did not change consistently after the treatment.

Decomposition abilities of the ammonia fungi All strains showed good mycelial growth on the agar media used. Results of the test for decomposition abilities are shown in Table 3. Clear cellulolytic ability was observed in all species except for *P. deerata, Hebeloma* spp. and *L. bicolor*. Strong laccase activity was shown by *L. tylicolor, C. echinosporus* and *P. deerata*; while *Hebeloma* spp. and *Peziza* sp. showed slight laccase activity. Chitin decomposition was observed in *A. botrytis, L. tylicolor, C. echinosporus* and *H. vinosophyllum*. All species pos-

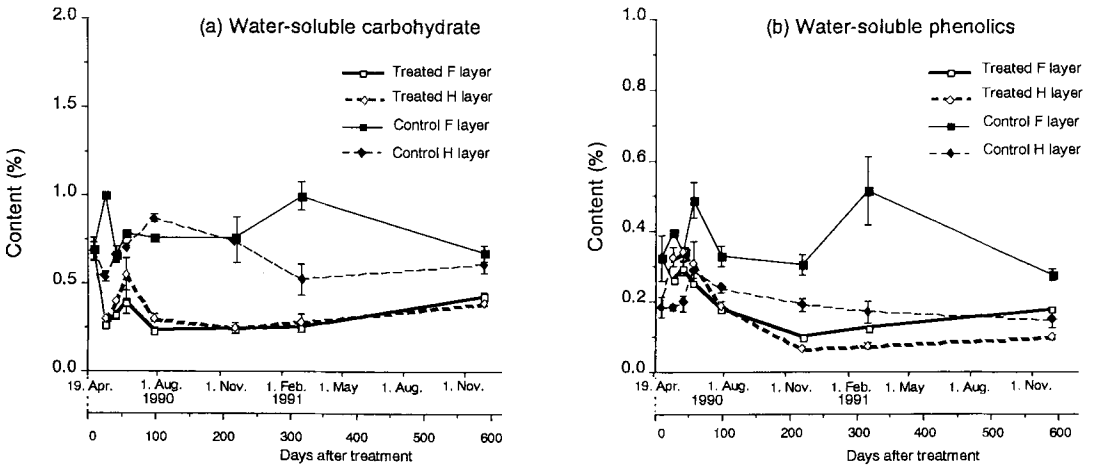

Fig. 3. Changes in the contents of water-soluble (a) carbohydrates and (b) phenolics in F layer and H layer soil treated with 700 g/m^2 of urea on 19 April 1990. Values are the means with standard errors.

sessed lipolytic and proteolytic activities.

When glucose was added to the culture media, laccase activity increased, but other enzyme activites decreased.

Discussion

Chitin, protein and lipid are components of animal and microbial materials. The abilities of the ammonia fungi to decompose these substances indicate that they can utilize animal and microbial remains, and therefore support the restricted occurrence of ammonia fungi under natural conditions, as summarized by Sagara (1992). In addition, bacteria and nematodes grow abundantly in the urea-treated soils (Table 1), and their waste materials and dead bodies could be decomposed and utilized by the ammonia fungi.

WSC concentration decreased after treatment (Fig. 3a). In this situation, enzyme activities allowing the fungi to acquire carbon sources from high-molecular substances are expected to be important. Early-stage species, which appear soon after treatment, show strong decomposition abilities (Table 3). These abilities, except for lignin decomposition, were strong without supply of glucose. Conversely, lignolytic ability was high in the presence of added glucose, an indication that this ability probably functioned for other reasons, e.g., acquisition of other sources, or removal of barrier for fungi to access cellulose (Cooke and Rayner, 1984). Late-stage species used in this experiment, which usually appear several months after treatment, have weak abilities to decompose high-molecular substances. These species are known as mycorrhizal species, and are likely to get carbon sources from associated roots. Differences in enzymatic activity between early-stage and late-stage species as found in this study were also reported by Enokibara et al. (1993).

Changes in WSC concentration also seem to influence the pattern of sporulation of the fungi. *Lyophyllum tylicolor*, an ammonia fungus, forms fruit-bodies at low concentrations of glucose in an agar medium (Yamanaka, 1994). At high concentrations of glucose,

Table 2. Statistical differences (n=3) in organic matter composition (a) between F and H layers, and (b) between urea-treated and untreated soil. The differences were examined using Student's t-test after analysis of variance. AS, ash; PE, petroleum ether extract; ME, 50% methanol extract; WC, water soluble carbohydrate; WP, water soluble phenolics; CE, holocellulose; LI, lignin. *$p<0.05$. **$p<0.01$. ns, not significant. —, not tested because of no data.

(a)

Days after treatment	Treated							Untreated						
	AS	PE	ME	WC	WP	CE	LI	AS	PE	ME	WC	WP	CE	LI
0[a]								ns	ns	ns	ns	ns	ns	ns
18	ns	ns	—	ns	ns	—	—	*	ns	—	**	**	—	—
33	**	ns	—	ns	ns	—	—	ns	**	—	ns	**	—	—
48	**	ns	ns	ns	ns	ns	ns	ns	**	ns	**	**	*	ns
89	*	*	ns	*	ns	**	ns	*	ns	ns	ns	**	*	**
210	**	ns	ns	ns	**	ns	**	*	ns	ns	ns	**	ns	*
307	*	ns	ns	ns	**	ns	*	*	**	*	**	*	ns	ns
581	*	ns	**	*	**	*	*	*	*	ns	ns	**	*	ns

a, soil was collected from the untreated plot only.

(b)

Days after treatment	F layer							H layer						
	AS	PE	ME	WC	WP	CE	LI	AS	PE	ME	WC	WP	CE	LI
0[a]														
18	ns	ns	—	**	**	—	—	ns	ns	—	**	*	—	—
33	ns	**	—	**	*	—	—	*	ns	—	**	**	—	—
48	ns	ns	**	*	**	ns	ns	ns	ns	**	ns	ns	ns	ns
89	ns	ns	**	**	**	*	ns	ns	ns	**	**	*	**	ns
210	ns	ns	ns	**	**	*	ns	*	**	ns	**	**	ns	*
307	ns	**	**	**	*	ns	ns	ns	ns	ns	ns	*	ns	ns
581	ns	*	**	**	**	ns	*	ns	*	**	**	ns	ns	ns

a, soil was collected from the untreated plot only.

Table 3. Abilities to decompose five organic substances with (+G) or without (−G) supply of glucose to culture media. Ratings were based on subjective estimates of relative abilities.

Species	Strain No.	Cellulose		Lignin		Chitin		Protein		Lipid	
		+G	−G	+G	−G	+G	−G	+G	−G	+G	−G
Amblyosporium botrytis	202	−/+++	++	−	−	−	+	++	++	±	++/+++
	221	−/+	++	−	−	−	+	+	+++	±	++
Ascobolus denudatus	152	++	++	−	−	−	−	++	+++	±	++
	222	+	++	−	−	−	−	±	++	++	++/+++
Peziza moravecii	185	+	++	±	−	−	−	++	+++	±	+
Pseudombrophila deerata	226	−	−	+++	+	−	−	++	++	±	±
	237	−	−	+++	−	−	−	++	++	+	++
Peziza sp.	165	+	++	+	±	−	−	++	+++	++	++
	206	+	++	−	−	−	−	+++	+++	++	++
Lyophyllum tylicolor	187	++	++	+++	+	−	++	++	+++	+	++/+++
	212	−	++	+++	+++	−	++	++	++	++	+++
Coprinus echinosporus	179	++	++	+++	−	−	++	+++	+++	++	++/+++
	215	+	++	+++	+	−	++	++	++	++	++/+++
Hebeloma vinosophyllum	135	−	−	+	−	−	+	±	±	+	+
	110	−	±	+	−	−	+	±	+	+	+
Hebeloma sp.	9	−	−	+	−	−	±	−	±	±	±
	248	−	±	+	−	−	−	±	+	±	+
Laccaria bicolor	252	−	−	−	−	−	−	±	±	±	+

it develops mycelial basidia and basidiospores rather than fruit-bodies.

The content of 50% methanol extract in soil rose above the control level for 3 months after treatment (Fig. 2). This means that decomposition of organic matter was enhanced by soil microorganisms. At this stage, the numbers of bacteria and nematodes increased (Table 1) and the saprophytic group of ammonia fungi appeared (Fig. 1). Subsequently, the content fell below the control level. At this stage, the numbers of bacteria and nematodes were lower than those in the earlier stage, and mycorrhizal species appeared.

In the control plots, WSP content was constantly higher in the F layer than the H layer (Fig. 3b). Limited vertical movement of water due to the low water content (Table 1) may have caused this difference. In the treated plots, water content was higher, and no such difference between the two layers was recognized.

In fungal succession on dung or freshly fallen leaves, the "nutritional hypothesis" (Webster, 1970) is taken into consideration to explain the order and period of each fungus (Lodha, 1974). In this hypothesis, species which show fast growth and weak enzyme activities should appear at the early stage, gradually being replaced by species which spread slowly and can lyse various high-molecular substances. During this succession, the organic matter composition of the substrates would also change, with readily assimilable components declining initially, followed by the more complex components, i.e., cellulose and lignin. In the case of the ammonia fungi, the earliest species, e.g., *A. botrytis* and *A. denudatus*, showed high levels of enzyme activities (Table 3). In additioin, the level of WSC in the treated soil was low from the beginning (Fig. 3a) and other components seemed not to change clearly in a way that would support this hypothesis (Table 2). Therefore, the nutritional hypothesis could not be employed to explain the succession of the ammonia fungi.

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Literature cited

Ander, P. and Eriksson, K.-E. 1976. The importance of phenol oxidase activity in lignin degradation by the white-rot fungus *Sporotrichum pulverulentum*. Arch. Microbiol. **109**: 1–8.

Carroll, G. and Petrini, O. 1983. Patterns of substrate utilization by some fungal endophytes from coniferous foliage. Mycologia **75**: 53–63.

Cooke, R. C. and Rayner, A. D. M. 1984. "Ecology of saprotrophic fungi," pp. 3–13. Longman, London.

Enokibara, S., Suzuki, A., Fujita, C., Kashiwagi, M., Mori, N. and Kitamoto, Y. 1993. Diversity of pH spectra of cellulolytic enzymes in Basidiomycetes. Trans. Mycol. Soc. Japan **34**: 221–228.

Hankin, L. and Anagnostakis, S. L. 1975. The use of solid media for detection of enzyme production by fungi. Mycologia **67**: 597–607.

Harkin, J. M. and Obst, J. R. 1973. Syringaldazine, an effective reagent for detecting laccase and peroxidase in fungi. Experientia **29**: 381–387.

King, H. G. C. and Heath, G. W. 1967. The chemical analysis of small samples of leaf material and the relationship between the disappearance and composition of leaves.

Pedobiologia **7**: 192–197.
Lodha, B. C. 1974. Decomposition of digested litter. In: "Biology of plant litter decomposition, vol. 1," (ed. by Dickinson, C. H. and Pugh, G. J. F.), pp. 213–241. Academic Press, London.
Morris, D. L. 1948. Quantitative determination of carbohydrates with Dreywood's anthrone reagent. Science **107**: 254–255.
Sagara, N. 1975. Ammonia fungi—a chemoecological grouping of terrestrial fungi. Contr. Biol. Lab. Kyoto Univ. **24**: 205–276.
Sagara, N. 1992. Experimental disturbances and epigeous fungi. In: "The fungal community, Its organization and role in the ecosystem, 2nd ed.," (ed. by Carroll, G. C. and Wicklow, D. T.), pp. 427–454. Marcel Dekker, New York.
Schlesinger, W. H. and Hasey, M. M. 1981. Decomposition of chaparral shrub foliage: losses of organic and inorganic constituents from deciduous and evergreen leaves. Ecology **62**: 762–774.
Smith, R. E. 1977. Rapid tube test for detecting fungal cellulase production. Appl. Environ. Microbiol. **33**: 980–981.
Suzuki, A. 1989. Analyses of factors affecting the occurrence and succession of the ammonia fungi. In: "Recent advances in microbial ecology," (ed. by Hattori, T., Ishida, Y., Maruyama, Y., Morita, R. Y. and Uchida, A.), pp. 275–279. Japan Scientific Society Press, Tokyo.
Suzuki, A. 1992. Physiological characteristics of the ammonia fungi and their role in bioscience. Proc. Asian Mycol. Symp., pp. 238–247.
Webster, J. 1970. Coprophilous fungi. Trans. Br. Mycol. Soc. **54**: 161–180.
Yamanaka, T. 1993. Microbial and chemical properties of soil under the successive occurrence of the ammonia fungi. Proc. 1st Intern. Symp. Mycol. Soc. Japan, pp. 42–45.
Yamanaka, T. 1994. Fruiting of *Lyophyllum tylicolor* in plate culture on Soytone-glucose agar and urea-treated soil extract agar. Mycoscience **35**: 187–189.