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Diversity of Root Fungal Floras: Its Implications for Soil-Borne Diseases and Crop Growth

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Abstract

Application of organic materials such as farmyard manure and crop residues to upland fields promoted root growth of crops and enhanced activities of nutrient uptake, resulting in an yield increase in five crops examined. Root fungal floras were diversified by the application of organic materials and the Brillouin's index of the diversity was positively correlated with the root biomass in all the crops. Continuous mono-cropping made the root fungal floras less diverse and the indices of diversity were smaller in the crops that were less tolerant against that type of cropping. The simplified floras of root fungi under continuous mono-cropping significantly corresponded to the fungal floras on the residual roots of the preceding crop. The simplification of root fungal floras resulted from less provisions of organic materials. Incidences of brown stem rot of adzuki beans caused by *Acremonium gregatum* were reduced by applying organic materials to the rhizosphere. This method is proposed for a practical use with the purpose of minimizing the rate of organic materials. The indices of diversity in root fungal floras were correlated negatively with the disease incidences. Diversification of root floras seems to enhance microbiostasis in the rhizosphere, whereby soil-borne diseases are suppressed and crop root growth is promoted. The critical role of organic materials applied is clearly revealed in maintaining biological fertility of soils, particularly in upland farming, where an incidence of soil-borne diseases in a major constraint to crop production.

Discipline: Soils, fertilizers and plant nutrition
Additional key words: continuous mono-cropping, organic materials, rhizosphere, root activity

Introduction

In intensive upland farming, it is not rare for farmers to adopt continuous mono-cropping and application of high doses of chemical fertilizers. Under such conditions rhizosphere microflora is easy to become disorderly in terms of normal plant growth. As a result, soil-borne pathogens often become injurious, hampering root growth of crops. In order to control the soil-borne diseases, application of agricultural chemicals including soil sterilants, deep tillage, crop rotation with grasses, and breeding of disease resistant cultivars have been put in practice. Despite of these practices, upland plants have been suffered from unidentified root diseases for many years.

Among the practices for controlling soil-borne diseases, provisions of organic materials with farmyard manure and composts have been widely employed by farmers. Many studies indicate that these materials increase available nutrients, soil biomass⁶⁾, cation exchange capacity²⁾, and improve soil reaction and soil structure. Organic materials also reduce the incidence of soil-borne diseases⁷⁾. In order to establish better soil management technologies to increase and stabilize a crop yield, it is an important subject to analyze the mechanisms how these organic materials improve the rhizosphere ecosystem, suppress the soil-borne phyto-pathogens and promote root growth.

In this connection, the present paper attempts to review microbiological characteristics of plant rhizosphere in relation to the development of root

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Table 1. Effects of farmyard manure and crop residues on the growth of upland crops in the middle stage of their growing period

Treatment	Sugarbeet		Maize		Soybean		Potato		Winter wheat	
	Shoot	Root	Shoot	Root	Shoot	Root	Shoot	Root	Shoot	Root
Control (CF)[a, b]	502	70	436	403	321	258	281	288	79	217
Rotation fields[c]										
Control (CF)	100	100	100	100	100	100	100	100	100	100
CF+FYM	115	149	111	111	102	100	95	136	111	118
CF+CR	88	100	110	116	105	108	109	136	110	134
CF+FYM+CR	116	119	143	126	124	98	103	131	119	107
Continuous cropping[c]										
CF	26	60	92	90	41	88	65	80	56	83
CF+FYM	37	69	89	102	52	85	89	119	71	93

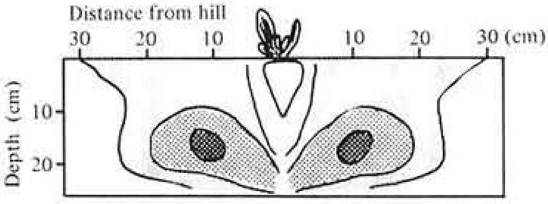
a): Chemical fertilizer applied.
b): Fresh weight (g/m^2).
c): Each figure shows a relative value to that of the corresponding control plots in rotation fields.

systems and the incidences of root diseases, with a special reference to the results of the studies conducted by the author regarding the biological control of soil-borne diseases through a provision of organic materials to the seedling rhizosphere.

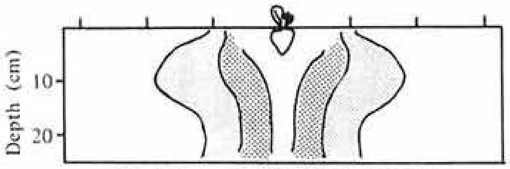
Diversification of root fungal flora and its implications for root growth

Seven-year investigations were made to compare the following two types of cropping system in regard to the effects of farmyard manure (FYM) and crop residues (CR) on root development, plant growth and yield as well as on some soil properties: one was a rotation of five crops; i.e. sugarbeet (*Beta vulgaris*), maize (*Zea mays*), soybean (*Glycine max*), potato (*Solanum tuberosum*) and winter wheat (*Triticum aestivum*); and the other was a continuous monocropping of seven crops each; i.e. kidney bean (*Phaseolus vulgaris*) and adzuki bean (*Vigna angularis*) in addition to the above five crops. The soils were of light-colored Andosol in Hokkaido, Japan[8].

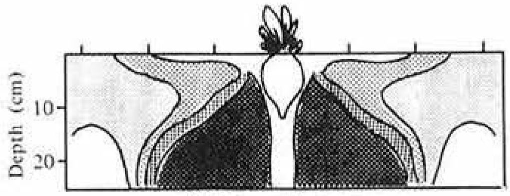
In all the plots with provisions of organic materials, the root development, including root elongation, branching and root-hair emergence, was promoted from the early growing stage. Table 1 shows significant increases in fresh root weight under applications of FYM, CR, or both. To evaluate the degree of improvement in root system, root activities in nutrient uptake were measured on the field with an Europium tracer technique[16]. Fig. 1 shows distributions of the active lateral roots of sugarbeet. It indicates that the root activities in the crop rotation plot with FYM were greater than those in the plot without FYM. The activities in continuous



Rotation without manure



Continuous cropping without manure



Rotation with manure

Fig. 1. Effects of farmyard manure and crop rotation on lateral roots activity of sugarbeet in field plots determined by an Europium tracer technique[16]
The denser color indicates higher root activities.

mono-cropping plots without FYM were greatly reduced, being restricted only around the tap root.

Among the various factors affecting the root growth, i.e. chemical and physical properties of soils rhizosphere microflora[1] and root growth regulators[17], behaviors of fungi in rhizosphere may be of particular importance, because of their high saprophytic/parasitic activities. Root fungal flora at the middle stage of the growing period measured by a serial root-washing technique[3,9] showed a significant difference between the plots with and without organic materials. It was also true among the crops studied and between the cropping systems with and without rotation. In the plots applied with organic materials the number of genera isolated was larger and the proportion of the predominant genera was lower than that in the plots without. In the continuous mono-cropping plots, however, the number of genera was smaller and the proportion of the predominant genera was higher than that in each of the corresponding rotation plots. It was generally observed that the application of FYM, CR, or the both diversified the root fungal flora and that the continuous mono-cropping made the fungal flora less diverse.

For comparative analyses of diversified root fungal flora, indices of diversity were calculated on the basis of the following Brillouin's equation[14]:

$$\text{Index of diversity} = 1/N \times \log(N!/N_1!\ N_2!\ \ldots\ N_n!),$$

where N is the number of total isolates of fungi from a plot, and $N_1 \ldots N_n$ are the numbers of isolates identified as genus "1" . . . genus "n". As shown in Fig. 2, high correlations were observed between the index and the root biomass in all the plants studied. The correlations were also observed in the field experiments with various organic materials. These results suggest that a greater diversity of fungal flora be obtained in further stimulated growth of roots. Fig. 2 also shows differences in the diversity among the crops. The indices for potato, winter wheat, soybean, sugarbeet, adzuki bean and kidney bean were decreasing in this order. The indices for the first two crops did not decrease even after a continuous mono-cropping for seven years, while those for sugarbeet and beans decreased significantly. This implies that potato and winter wheat are more tolerant against continuous mono-cropping than sugarbeet and beans. From these results, it is concluded that the diversity of root fungal flora could be a useful index of soundness or stability of rhizosphere ecosystem.

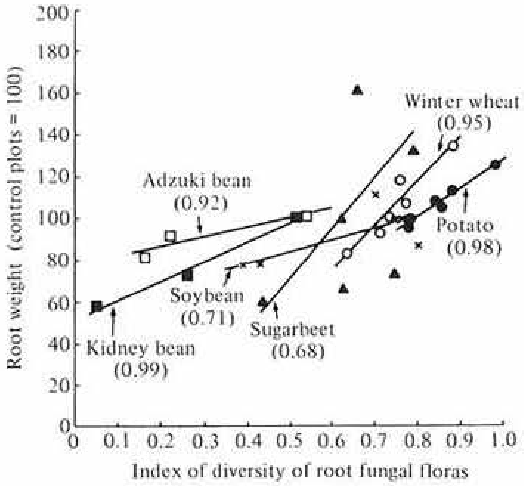


Fig. 2. Relationship between the index of diversity of root fungal floras and the root weight
Values in parentheses indicate correlation coefficients.

Root growth under continuous mono-cropping and roles of the root microflora

As mentioned above, a long-term continuous mono-cropping generally made the root fungal flora less diverse and hampered the root growth. To analyze the process involved in these changes, root fungal floras of seven crops under the continuous mono-cropping and the crop rotation plots were investigated[13]. As shown in Table 2, there are high similarities between the fungal flora of residual roots of the preceding crops and that of early growing roots of the succeeding crops in the two cropping systems under study. The data suggest that the residual roots of the preceding crops affect root fungal floras of the succeeding crops at their working sites in soils, while fungal floras in non-rhizosphere soils have a limited effect. Consequently, a cropping sequence affects plant growth through a formation of fungal floras and their diversity on roots which closely correlates with healthy root growth. Annual applications of organic materials to the continuous mono-cropping plots diversified root fungal

Table 2. Similarity of the fungal floras on the roots of succeeding crops at the early growing stage to those on the residual roots of preceding crops, and in the soils under continuous and rotation cropping systems[a)]

Succeeding crop	Similarity to fungal flora on residual root			Similarity to soil fungal flora	
	Continuous mono-cropping		Rotation[b)]	Continuous mono-cropping	Rotation[b)]
	Second	Third		Third	
Sugarbeet	0.49	0.67	0.62	0.15	0.14
Potato	0.66	0.31	0.47	0.51	0.58
Spring wheat	0.70	0.79	0.65	0.50	0.45
Maize	0.81	0.59	0.42	0.51	0.27
Soybean	0.98	0.67	0.99	0.70	0.58
Kidney bean	0.79	0.61	0.98	0.89	0.23
Adzuki bean	0.66	0.97	—	0.29	0.17

a): The indices of similarity are calculated after a Morishita's equation[11)].

b): Crop rotation plots are grown to adzuki bean as a preceding crop.

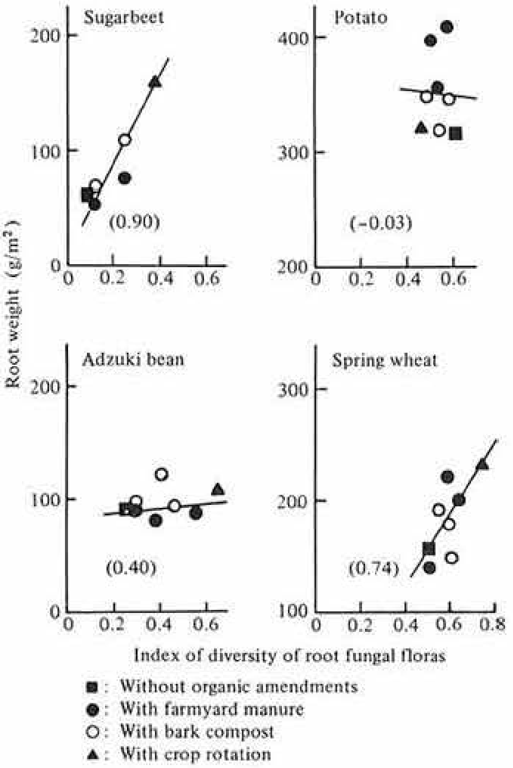


Fig. 3. Relationship between the index of diversity of root fungal floras and the root weight in continuous mono-cropping

Values in parentheses indicate correlation coefficients.

floras[10)] and improved the root growth. The indices of diversity were correlated with the root biomass as shown in Fig. 3, though the correlation coefficients were smaller than those obtained in the crop rotation plots as shown in Fig. 2. The application also reduced the incidence of soil-borne diseases, such as *Rhizoctonia* root rot of sugarbeet and brown stem rot of adzuki bean.

Microbial ecosystems in rhizosphere and non-rhizosphere which might be associated with the above status of root growth and disease incidence will be subjected to further analyses hereafter. Microbial proliferation depends primarily on the quality and quantity of some substrates. Organic materials applied to soils are decomposed by microbes and may release various intermediates, which may activate various kinds of microbes in the soils. In the rhizosphere, microbes may proliferate considerably due to an abundant supply of root exudates and senescent root tissues. Such an increase in various microbes may improve the microbial balance in non-rhizosphere and rhizosphere, which may suppress a drastic multiplication of such specific microbes as phyto-pathogens and stimulate crop root growth.

A great number of experiments have been conducted with an aim of establishing appropriate methods for biological control of root diseases by means of application of FYM, composts and crop residues[7)]. In the course of the implementation of those studies on biological control, it was demonstrated that a provision of organic materials produces antibiotics and activates microbial antagonism[4,5]. Though the details on dynamics of individual pathogens and antagonists are still subject to further studies, the results obtained by the author[9] indicate that it is very necessary to identify the dynamics of microbial ecosystem in the rhizosphere as a whole.

It is also recognized that a role of more diversified fungal floras in the rhizosphere microbiostasis will have to be identified, taking into account the behaviors of minor pathogens[15], since some data show that even non-pathogenic fungi occasionally provide high pathogenicity in case where they predominate in the rhizosphere. In addition, it is well known that some soils are disease suppressive and others are conducive. Therefore, further studies on soils will have to be implemented from the viewpoint of rhizosphere microbiostasis, which should be induced not only by actinomycetes and fluorescent pseudomonads[18] but also by diversified fungal floras on roots, as shown in the author's field experiment[12].

It is therefore concluded that the diversified fungal floras induced by organic materials applied and adequate crop rotation are closely associated with an increase in microbial buffering capacity of rhizosphere ecosystem. The critical role of organic materials applied is clearly revealed in maintaining soil biological fertility, particularly in intensive upland farming where an incidence of soil-borne diseases is a major constraint to crop production.

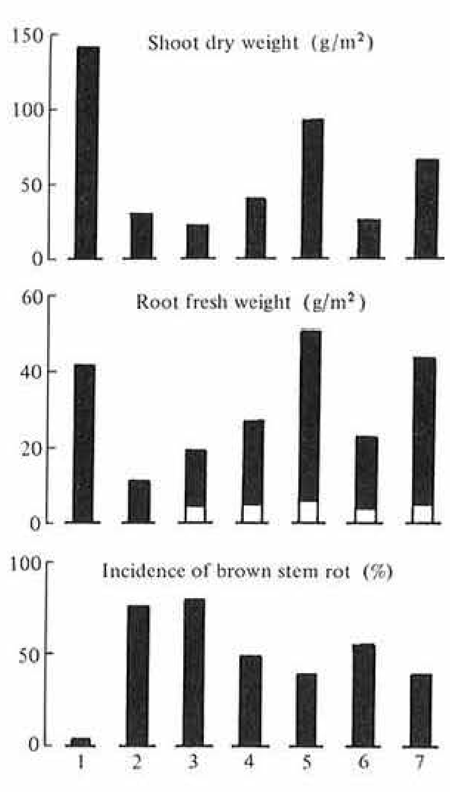


Fig. 4. Effects of the rhizosphere application of FYM, BC and CDS on the growth of adzuki bean and incidence of brown stem rot

1. Crop rotation without organic materials,
2. Continuous mono-cropping (CMC) without organic materials,
3. CMC and rhizosphere application (RA) to diseased soils,
4. CMC and RA to normal soils,
5. CMC and RA with FYM to normal soils,
6. CMC and RA with BC to normal soils,
7. CMC and RA with CDS to normal soils.

Disease control by applications of organic materials to seedling rhizosphere

In order to identify the most efficient use of organic materials in controlling soil-borne diseases, some experiments on biological control of brown stem rot (*Acremonium gregatum*) of adzuki bean were conducted. Under the study, the following organic materials were applied only to the seedling rhizosphere: FYM, BC (bark compost) and CDS (compost made of diseased stems of the beans)[12]. In a preliminary test under the severely diseased field condition, FYM, BC and CDS applications to a plow

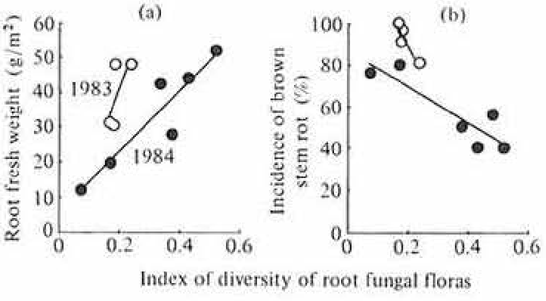


Fig. 5. Relationships of the index of diversity of root fungal floras with the root weight (a), and with the percentage of adzuki bean affected by brown stem rot (b)

Field experiments were carried out in 1983 and 1984 under the severely infected field condition by continuous mono-cropping.

layer at the rate of 50 t/ha, suppressed the incidence of the disease significantly and stimulated the plant growth.

In the rhizosphere application technique, paper pots of 3 cm in diameter and 5 cm in height, were packed with the mixture of normal soils and organic materials (1 : 1 w/w), in which adzuki beans were seeded. The pots with plants were transferred to the field applied with chemical fertilizers. Fig. 4 shows a result observed in the continuous mono-cropping plot. The growth of roots and shoots was greatly promoted with provisions of FYM and CDS. Fungal floras on the roots developed outside the paper pots with organic materials showed a greater diversity as compared with those without FYM and CDS; the same pattern of the fungal floras was also observed inside the paper pots. Fig. 5 shows that the indices of diversity are correlated negatively with the incidences of diseases infections, and positively with the root biomass.

The rhizosphere application technique was also examined in test fields with three types of soils. It was confirmed that the field test gave almost the same results as mentioned above. These results indicate that a great diversity of fungal floras is effective in controlling soil-borne diseases and promoting crop root growth. The amount of organic materials to be applied to the rhizosphere was estimated at a level of as small as 1.4 t/ha. Such a low level implies that the above technique is practicable for farmers to adopt under an intensive farming system, where organic materials are generally in short supply.

References

1) Bowen, G. D. & Rovira, A. D. (1961): The effects of micro-organisms on plant growth. I. Development of root and root hairs in sand and agar. *Plant Soil*, **15**, 166–188.

2) Harada, T. & Inoko, A. (1975): Cation-exchange properties of soil organic matter. I. Effects of conditions for the measurement on cation-exchange capacity values of humic acid preparations. *Soil Sci. Plant Nutr.*, **21**, 361–369.

3) Harley, W. B. & Waid, J. S. (1955): A method studying active mycelia on living root and other surface in the soil. *Trans. Br. Mycol. Soc.*, **38**, 104–118.

4) Homma, Y. et al. (1979): Mechanism of suppression of tomato *Fusarium*-wilt by amendment of chicken manure to soil. *Bull. Shikoku Agr. Exp. Sta.*, **34**, 103–121 [In Japanese with English summary].

5) Hori, K. et al. (1980): Relationship between soil productivity and microorganisms in the soil. IV. Control of tomato corky-root disease by the application of organic matter. *Bull. Shizuoka Agr. Exp. Sta.*, **25**, 26–35 [In Japanese with English summary].

6) Jenkinson, D. S. & Rayner, J. H. (1977): The turnover of soil organic matter in some of the Rothamsted classical experiment. *Soil Sci.*, **123**, 298–305.

7) Matsuda, A. (1981): Crop rotation and organic amendment for biological control of soil-borne disease. *Shokubutsu Boeki*, **35**, 108–114 [In Japanese].

8) Matsuguchi, T. & Nitta, T. (1987): Organic amendment to upland soil as a conditioner of the rhizosphere ecosystem. 1. Effects of organic amendment to upland soil with farmyard manure and crop residues on the root development and plant growth. *Jpn. J. Soil Sci. Plant Nutr.*, **58**, 653–660 [In Japanese].

9) Matsuguchi, T. & Nitta, T. (1987): Organic amendment to upland soil as a conditioner of the rhizosphere ecosystem. 2. Effects of organic amendment on rhizosphere microflora as affecting the root development of upland crops. *Jpn. J. Soil Sci. Plant Nutr.*, **58**, 661–670 [In Japanese].

10) Matsuguchi, T. & Nitta, T. (1988): Organic amendment to upland soil as a conditioner of the rhizosphere ecosystem. 3. Effects of organic amendment on root development and rhizosphere microflora of monocropped upland crops. *Jpn. J. Soil Sci. Plant Nutr.*, **59**, 1–11 [In Japanese with English summary].

11) Morishita, M. (1960): Measuring of interspecific association and similarity between community. *Mem. Fac. Kyushu Univ.*, **E3**, 65–80.

12) Nitta T. & Matsuguchi, T. (1988): Organic amendment to upland soil as a conditioner of the rhizosphere ecosystem. 5. Biological control of brown stem rot of adzuki beans through an organic amendment to the seedling rhizosphere. *Jpn. J. Soil Sci. Plant Nutr.*, **59**, 140–148 [In Japanese with English summary].

13) Nitta, T. et al. (1989): Effects of cropping systems and application of organic materials on root growth and rhizosphere microflora of upland crops. *Soil Microorganism*, **34**, 41–49 [In Japanese with English summary].

14) Pielou, E. C. (1977): Ecological diversity and its measurement. *In* Mathematical ecology, Wiley-Interscience Pub., New York, 299–307.

15) Salt, G. A. (1979): Increasing interest in minor pathogens. *In* Soil-borne plant pathogens, Academic Press, 289–312.

16) Shibuya, M., Futami, K. & Fujii, H. (1987): Diagnosis of root activity distribution of continued cropped cabbage in field. *Jpn. J. Soil Sci. Plant Nutr.*, **56**, 59–61 [In Japanese].

17) Torrey, J. G. (1976): Root hormones and plant growth. *Ann. Rev. Physiol.*, **27**, 435–459.

18) Weller D. M. & Cook, R. J. (1983): Pseudomonads from take-all conducive and suppressive soils. *Phytopathology*, **71**, 463–469.

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