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POSSIBLE ALLELOPATHIC INTERACTION BETWEEN *ORYZA PERENNIS* AND *LEERSIA HEXANDRA*[1]

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Abstract

An introduction experiment of *Oryza perennis* populations into different habitats indicated that *Leersia hexandra* was a key species determining the biotic environment of the former. To look into the interaction mechanisms of the two species, their allelopathic interrelations were examined by different methods, *i.e.*, assay of the effects of aqueous leachates and extracts from the two species on the radicle growth of rice and lettuce, on the growth of adventitious roots emerging from a node of cuttings of the two species, and the effects of powdered plant material added to soils on the root development of cuttings. Both grass species showed phytotoxic effects on the radicle growth of rice and lettuce, intra- and inter-specifically. *Leersia* showed in many cases higher phytotoxicity than *Oryza* although the pattern of variations was complex. The concentrations of many phytotoxins identified were higher in *Leersia* extract than in *Oryza*. An observation of plants occurring from buried seed pool in soils to which powdered plant material were added also showed higher phytotoxicity of *Leersia* than of *Oryza*. Probably, allelopathy plays an appreciable role in the successional replacement of the two species.

Introduction

The common wild rice, *Oryza perennis* Moench (=*O. rufipogon* Griff.), is distributed throughout the humid tropics, and its Asian race is considered to be the wild progenitor of cultivated *O. sativa* (Oka, 1964, 1974). The Asian race shows a perennial-annual continuum, varying greatly in various life-history traits among its varieties (Oka and Morishima, 1967; Oka, 1976). *Leersia hexandra* Sw. is a perennial grass with short rhizomes, commonly found in marshy habitats in Taiwan and other tropical Asian countries. It is a companion plant of *O. perennis* in about

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40 percent of the habitats observed in India and Thailand (Morishima *et al.*, 1980).

In Taiwan, three small populations of *O. perennis*, which were hybrid swarms with *O. sativa* (Oka and Chang, 1961), had existed in marshes along natural streams at Patu, Taoyuan Hsien, but they became extinct around 1975 as displaced by *L. hexandra* (cf. Kiang *et al.*, 1979). An introduction experiment of this wild rice into different lowland habitats after denudation also demonstrated that *L. hexandra* was a key species determining the regenerating success of the wild rice, suggesting that the niches of *O. perennis* and *L. hexandra* would largely overlap (Oka, 1984).

It is known that allelopathic interaction plays a significant role in regulating the process of secondary succession (Rice, 1974; Chou, 1980; Numata, 1982). Expectedly, the interaction between *O. perennis* and *L. hexandra* is at least partly ascribed to allelopathy. The purpose of this study was to evaluate the allelopathic potentials of *O. perennis* and *L. hexandra* in relation to the mechanisms of their interaction. Experiments were carried out in 1982 at both Nankang (by the first two authors) and Taichung (by the third author).

Materials and Methods

Plant Materials

The *O. perennis* plants used belong to a highly perennial strain of Indian origin (W120, from Cuttack). They were planted in July 1981 into abandoned marshy fields after denudation and tillage at Nankang (the experimental farm of Institute of Botany, Academia Sinica) and Hsisinpa, Taichung (a farm of National Chung Hsing University), and were observed with regard to persistence and other related characters (they were also planted at three other sites, but were not used for this study). The *L. hexandra* plants used were naturally occurring ones in the experimental fields at both Nankang and Hsisinpa. The plants of both species at the stage of flowering to seed maturity in November-December (1982) were sampled by cutting at the first elongated internode.

Preparation of Leachate and Extract

For leaching, fresh plants of each species weighing 500 g (excluding panicles) were put in 5,000 ml of distilled water by using a leaching apparatus described by Chou and Yang (1982; at Nankang only). Each leachate was obtained after continued cycling of water for 48 hours (Leachate I), 96 hours (II), and 144 hours (III). The leachates were filtered through Whatman 42 paper and stored below 0°C.

For extraction, air-dried leaves and stems of each species were ground, and 100 g powder was put in 2,000 ml of distilled water, shaken for 2 hours, and filtered through cheesecloth and Whatman 42 paper to obtain Extract I. To the residue was added 3,000 ml of distilled water to obtain Extract II by the same procedure, and again to obtain Extract III similarly (at Nankang). At Tachung, only Extract I prepared by the same method was used for experiment.

At Nankang, the osmotic concentrations of leachates and extracts were determined using a Fiske model G-66 osmometer, and were adjusted to about 25 milliosmols by diluting with distilled water before using for bioassay. Distilled water was used as the control. Their pH values were adjusted to about 7. At Taichung, the extract was directly used for bioassay in sand culture; its osmotic reading obtained by using a Wescor HR-33T dew-point microvoltmeter was about −2.8 bar (equivalent to 125 milliosmols). Therefore, two controls were used, one being tap water and the other being a 8.2% solution of polyethylene glycol which had the same osmotic concentration. Their pH values were about 6.5 when prepared, but were about 8 at the end of sand culture.

Bioassay Techniques

To evaluate the phytotoxicities of leachates and extracts, two methods of bioassay were employed at Nankang. One was to measure the radicle growth of a rice (*O. sativa* var. Taichung 65) and a lettuce (*Lactuca sativa* var. Great Lakes) cultivar (cf. Chou and Lin, 1976); 3 replications were made. The other method was to measure the development of adventitious roots from a node of excised stem segments (cuttings) of *O. perennis* and *L. hexandra* in respective leachates and extracts (water culture). Five fresh cuttings, 5 to 6 cm long and each having a node which was the third to fifth from the top, were put in about 80 ml of leachate or extract, or distilled water (control) in 100 ml beakers, with 4 replications. The beakers were kept at 25°C for 6 days in the dark, and the total length, number, and fresh weight of adventitious roots per cutting were recorded with cuttings initiating roots.

As to the rooting test of cuttings, it is reported in *O. perennis* that the development of adventitious roots from a node of excised stem segments, sampled at the stage of seed ripening and sand-cultured for 5 days, served as an index of ratooning ability or perenniality (Oka and Morishima, 1967); by this method, the values of perenniality index for the *Oryza* and *Leersia* plants used were 2.7 and 2.4 (3 being the highest), and the percentages of rooting were 98 and 72, respectively.

At Taichung, the root development from the node of cuttings was tested by sand culture with the Extract I of each species; water culture was also attempted, but was unsuccessful because of bacterial contamination. Two methods were used, each with two replications. By one method (A), 10 cuttings of each species were sand-cultured in a 100-ml beaker at about 25°C for 7 days in the dark, and the total root length of each cutting sprouting roots was recorded. By the other method (B), the cuttings were cultured in a tray with moist sand *a priori* for three days, and those initiating roots were selected and their total root lengths were recorded.

Then, they were planted in 100-ml beakers containing sand and plant extract, and were kept at about 25° for 5 days. Their total root length was measured again to determine root elongation during the 5-day culture by subtracting the first measurement of root length.

Furthermore, at Taichung, powdered leaves and stems of both species were added to pulverized dry soil (silty loam from a rice field) at 40 and 80 g/kg rates, mixed thoroughly, put in plastic trays (320 cm^2, 5 cm deep), watered, and kept moist for 30 days in a glass-house until the decay of plant powder was nearly complete. The same soil without plant powder was used as the control. Then, 10 cuttings per treatment of each species were planted with two replications. The trays were placed in a high humidity incubator (ca. 29°C) for 3 days to promote root initiation, and were were placed in a glass-house (20°–25°C) for 13 days. Total root length was recorded on each cutting which took root. After this experiment was over, the soils in trays were kept moist in the glass-house for 92 days to observe the plants occurring from buried seed pool.

Quantitative Analysis of Phytotoxins

Since most phytotoxins present in grasses are phenolics and water soluble, those in the aqueous extracts of *O. perennis* and *L. hexandra* were isolated using the extraction techniques described by Chou and Young (1975). The isolated compounds were identified and their concentrations were determined by using paper chromatography (Chou and Young, 1975) and a high pressure liquid chromatograph (Water Associates, model 6000A, with a Merck reverse-phase C-18 column); authentic compounds (Merck) were each run simultaneously for comparison. From the result, the concentrations of major phytotoxins in the two species were estimated.

Results

Phytotoxicity of Leachates and Extracts on Rice and Lettuce Radicles

The radicle growth of both rice and lettuce was inhibited by the leachates from *O. perennis* and *L. hexandra*, and more strongly by the extracts (Table 1). The ratio of root inhibition of leachate to extract was about 2/3 (66%) in those from *Oryza*, and about 1/3 (34%) in those from *Leersia*. Comparisons between treatments of mean root length in percent of the control showed that the rice roots were more strongly affected by leachates and less strongly by extracts than the lettuce roots (Table 1). The results indicate differential sensitivities of the plants.

The analysis of variance of the data indicated that in the effect on lettuce roots, *Leersia* extracts had significantly higher toxicity than *Oryza* extracts, while *Oryza* leachate showed higher toxicity than *Leersia* leachates. However, all *Oryza-Leersia* differences in the effect on rice roots were insignificant (Table 2).

Table 1. *Effects of aqueous leachates and extracts of* ***Oryza perennis*** *and* ***Leersia hexandra*** *on the radicle growth of rice and lettuce*

Data show the percent of radicle growth of the distilled water control.

Source	Sequence of leaching or extraction	Leachate		Extract		Leach./Extract[a] (%)
		Rice	Lettuce	Rice	Lettuce	
Control		100	100	100	100	
Oryza	I	81.5	90.8	69.3	66.3	
	II	81.5	75.2	69.7	60.2	
	III	61.2	85.2	73.7	73.0	
	Mean	74.7	83.7	70.9	66.5	66.5
Leersia	I	79.7	90.9	69.7	59.6	
	II	101.9	82.1	69.2	48.9	
	III	67.6	102.9	61.4	73.3	
	Mean	83.1	92.0	66.7	60.6	34.5

[a] Ratio of % reduction due to leachate and extract, mean for rice and lettuce.

Table 2. *F values obtained from analysis of variance for the effects of leachates and extracts on rice and lettuce root length*

Source of variation	*df*	Rice	Lettuce
Leachate:			
Treatment	6	3.9*	2.2
Control : Treated	1	6.5*	4.3
Oryza : *Leersia*	1	1.8	3.5
Oryza, I: II: III	2	2.3	2.1
Leersia, I: II: III	2	5.1**	2.5
Error	14		
Extract:			
Treatment	6	3.8	31.1**
Control : Treated	1	20.9**	135.0**
Oryza : *Leersia*	1	0.7	6.2*
Oryza, I: II: III	2	0.2	4.8
Leersia, I: II: III	2	0.5	17.9**
Error	14		

*, ** Significant at 5% and 1% levels, respectively.

*Intra- and Inter-specific Phytotoxicity of **O. perennis** and **L. hexandra***

The growth of adventitious roots from a node of excised stem segments of the two species were affected by the leachates and extracts from both species considerably. The data for leachates and for extracts, recorded at Nankang, are given in Tables 3 and 4, respectively. The root-inhibiting effect of extracts showed a decreasing trend following the sequence of extraction, I>II>III, which was not found in the experiment with rice and lettuce radicles. No such trend was observed in the effect of leachates.

The data were subjected to analysis of variance (Table 5). Against the total root length of *Oryza* cuttings, *Leersia* leachates exhibited significantly lower inhibition, but *Leersia* extracts gave significantly higher inhibition than did *Oryza*

Table 3. *Effects of aqueous leachates on the growth of adventitious roots of **Oryza perennis** and **Leersia hexaudra** cuttings*

Root growth from a node of cuttings of *Oryza perennis* and *Leersia hexandra* cultured six days in aqueous leachates of the two species, as compared with the control (in distilled water). Data in the parenthesis indicate the percent of distilled water control.

Leachate source	Leaching sequence	Total root length (mm) per cutting	Root number per cutting	Root fresh wt (mg) per 5 cuttings	Mean[a] %
		Oryza perennis cuttings:			
Control		177	3.63	61	
Oryzr	I	95 (54)	2.69 (74)	43 (71)	
	II	23 (13)	2.00 (55)	6 (10)	
	III	72 (41)	3.02 (83)	35 (57)	
	Mean	63 (36)	2.57 (71)	28 (46)	51
Leersia	I	83 (47)	2.47 (68)	28 (46)	
	II	41 (23)	1.88 (52)	16 (26)	
	III	134 (76)	3.65 (101)	42 (69)	
	Mean	86 (49)	2.66 (73)	29 (47)	56
		Leersia hexandra cuttings:			
Control		171	4.28	79	
Oryza	I	82 (48)	4.34 (101)	35 (44)	
	II	99 (58)	4.98 (116)	51 (65)	
	III	89 (52)	2.51 (59)	44 (56)	
	Mean	90 (53)	3.94 (92)	43 (55)	67
Leersia	I	61 (36)	2.80 (65)	16 (20)	
	II	69 (40)	4.24 (99)	24 (30)	
	III	101 (59)	3.74 (87)	52 (66)	
	Mean	77 (45)	3.59 (84)	31 (39)	56

[a] Mean of percentages for 3 traits.

Table 4. *Effects of aqueous extracts on the growth of adventitious roots of* ***Oryza perennis*** *and* ***Leersia hexandra*** *cuttings*

Root growth from a node of cuttings of *Oryza perennis* and *Leersia hexandra* cultured in aqueous extracts of the two species, as compared with the control (in distilled water). Data in the parenthesis indicate the percent of distilled water control.

Extract source	Extracting sequence	Total root length (mm) per cutting	Root number per cutting	Root fresh wt (mg) per 5 currings	Mean[a] %
		Oryza perennis cuttings:			
Control		292	6.55	203	
Oryza	I	77 (26)	3.61 (55)	72 (35)	
	II	143 (49)	6.52 (99)	104 (51)	
	III	306 (105)	5.56 (85)	304 (150)	
	Mean	177 (60)	5.23 (80)	160 (79)	73
Leersia	I	51 (17)	3.40 (52)	69 (34)	
	II	105 (36)	6.43 (98)	105 (52)	
	III	175 (60)	9.05 (138)	182 (90)	
	Mean	110 (38)	6.29 (96)	119 (58)	64
		Leersia hexandra cuttings:			
Control		116	4.58	97	
Oryza	I	23 (20)	2.50 (55)	22 (23)	
	II	28 (24)	2.65 (58)	30 (31)	
	III	68 (59)	3.46 (75)	72 (74)	
	Mean	26 (34)	2.87 (63)	41 (43)	43
Leersia	I	23 (19)	2.74 (60)	28 (28)	
	II	23 (20)	3.74 (82)	39 (40)	
	III	56 (48)	4.64 (101)	65 (67)	
	Mean	34 (29)	3.71 (81)	44 (45)	52

[a] Mean of percentages for 3 traits.

leachates and extracts, respectively (Tables 3, 4 and 5). However, on *Leersia* cuttings, no significant *Oryza-Leersia* differences were found in the root-inhibiting effects of leachates and extracts. The same pattern of variations was also found in root fresh weight (Tables 3 and 4) although the results of analysis of variance for this trait are not presented to save room, since root fresh weight and total root length were highly correlated ($r=0.943$ for leachate; $r=0.924$ for extract; $P<0.01$ in both). The two traits may be considered as similarly representing the overall root development.

On the other hand, root number per cutting would represent the activity of root initiation from primordia in the stem node. In this trait, *Oryza* extracts showed higher inhibitory effect than *Leersia* extracts on the cuttings of both species although the *Oryza-Leersia* differences in the effect of leachates were nearly insignificant. Root number per cutting was correlated with root fresh weight (r= 0.713 for leachate; r=0.666 for extract; P<0.01 in both), but the correlation coefficients were lower than those found between total root length and root weight. In addition, mean root length (=total length/number) was examined, but it did not necessarily represent root development since root initiation would continue during the culture and the initiation of many short roots would lower the value. It was weakly correlated with root number per cutting (r=0.525, P<0.05 for leachate) or uncorrelated (r=0.342 for extract) although it was significantly correlated with root fresh weight. Therefore, total root length and root weight were considered most appropriate for evaluating phytotoxicity in the bioassay.

The pattern of variations observed in the effect of leachates indicated that autotoxic effects were greater than interspecific allelotoxic effects in both *Oryza* and *Leersia* (Table 3). In contrast, in the effect of extracts, allelotoxic effects were greater than autotoxic effects. This trend was clearly recognized in the mean of percentages of control for the three traits (total root length, root number and root weight) although it was endorsed by significant differences only partly. The

Table 5. *Analysis of variance for the effects of leachates and extracts of **Oryza perennis** and **Leersia hexandra** on the root growth from a node of cuttings of the two species*

Mean squares are shown.

Source of variation	df	*Oryzr perennis*		*Leersia hexandra*	
		Leachate	Extract	Leachate	Extract
Total root length (mm) / cutting:					
Treatment	6	11167**	40599**	5209*	4931**
Control : Treated	1	35555**	76288**	26051**	21534**
Oryza : ***Leersia***	1	3174	25611*	1014	288
Oryza, I:II:III	2	5321*	55447**	312	2450*
Leersia, I:II:III	2	8815**	15402**	1784	1463**
Error	21	949	3656	1724	437
Root number / cutting:					
Treatment	6	2.05	15.09**	3.18**	3.22*
Control : Treated	1	3.54	2.13	0.90	5.74*
Oryzr : *Leersia*	1	0.06	6.79	0.72	4.21*
Oryza, I:II:III	2	1.08	8.83	6.59**	1.06
Leersia, I:II:III	2	3.27	31.98**	2.13*	3.62*
Error	21	1.05	3.22	0.61	0.98

*, ** Significant at 5% and 1% levels, respectively.

root-inhibiting effect of leachates and that of extracts were weakly intercorrelated when corresponding values were compared ($r=0.353$, $P<0.05$ in total root length; $r=0.500$, $P<0.1$ in root weight). The low values of correlation coefficients reflect the differential phytotoxicities of leachate and extract on the two species.

In this context, it is worth to note that the inhibitory effect of leachates on *Oryza* was stronger than that of extracts, while the inhibitory effect of extracts on *Leersia* was stronger than that of leachates (Tables 3 and 4). In other words, *Oryza* was more sensitive to leachate, and *Leersia* was sensitive to extract, relatively. The tendency of leachate/extract ratio to be greater in those from *Oryza* than in those from *Leersia*, as found in the effects on rice and lettuce radicles, was not disproved although insignificant differences were found in the bioassay with cuttings.

The sand-culture test of extracts at Taichung also revealed considerable root-inhibiting effects (Table 6). In experiment A, in which the effect on root initiation

Table 6. *Effects of aqueous extracts on total root length (A) and root elongation (B) of* ***Oryza perennis*** *and* ***Leersia hexandra*** *cuttings*

Total root length (mm) per cutting of *Oryza perennis* and *Leersia hexandra* sand-cultured for 7 days (A), and root elongation per cutting when rooted cuttings were sand-cultured for 5 days (B), with aqueous extracts of the two species (at Taichung).

Treatment	*Oryza perennis*		*Leersia hexandra*	
	Mean±σ	%	Mean±σ	%
A: Before rooting:				
Control (water)	322±72	100^a	182±127	100^a
Extract from *Oryza*	218±58	68^b	104±24	57^b
Extract from *Leersia*	187±39	58^b	111±17	61^b
t(*Oryza* : *Leersia*)	1.99(*)		1.07	
B: After rooting:				
Control (water)	201±52	100^a	128±21	100^a
Control (PEG)[1)]	215±64	107^a	114±24	89^a
Extract from *Oryza*	134±53	67^b	105±23	82^b
Extract from *Leersia*	87±30	43^b	75±19	59^c
t(*Oryza* : *Leersia*)	3.47**		4.52**	
LSD (5%)	67.8		23.3	

*, ** Significant at 5% and 1% levels, respectively.
(*): Close to 5% level.
a, *b*, *c*—Classes differing at 5% level of significance.
σ—Standard deviation on single determination.
1) Polyethylene glycol, 8.2% solution, having the same osmotic concentration as of extracts.

Table 7. *Analysis of variance for the suppressing effects of extracts in sand culture and of powdered plant tissues in soil of* ***Oryza perennis*** *and* ***Leersia hexandra*** *on root development from cuttings of the two species*

Source of variation	*df*	MS, root length (mm)	
		Oryza	*Leersia*
Sand culture:			
Treatment, A : B	1	30,907**	2,670**
Control: *Oryza* : *Leersia*	2	16,346**	4,351**
Control : Extract	1	29,610**	8,437**
Oryza : *Leersia*	1	3,081	265
Interaction[a]	2	353	780*
Replication	1	7,752*	1,430*
Error	5	899	106
Soil experiment:			
Treatment	4	4,890**	1,076*
Control : Treated	1	16,241**	2,544**
Oryza : *Leersia*	1	2,048*	1,326*
Concentration	1	1,201	435
Interaction[b]	1	71	1
Replication	1	1,300	1,588*
Error	4	221	136

*, ** Significant at 5% and 1% levels, respectively.

[a] Between (A : B) and (Control : *Oryza* : *Leersia*).

[b] Between (*Oryza* : *Leersia*) and concentration (40:80 g/kg).

was involved, the autotoxic effects were slightly less powerful than interspecific allelopathic effects. The *t*-test based on single measurements showed that *Leersia* extract had stronger inhibiting effect on *Oryza* roots than *Oryza* extract (close to the 5% level of significance), but their effects on *Leersia* roots did not differ significantly. In experiment B, in which the effect on root initiation was excluded, the *t*-test showed that the inhibiting effect of *Leersia* extract was significantly stronger than that of *Oryza* extract on both *Oryza* and *Leersia* roots. As there were only two replications, the analysis of variance of treatment means failed to detect *Oryza*-*Leersia* differences (Table 7). The data also showed that there was no significant difference between cultures with tap water and polyethylene glycol solution (PEG). Shoot length, although measured, showed no meaningful differences. In general, the phytotoxicity of extracts on *Oryza* cuttings, assayed by sand culture at Taichung, was comparable to that assayed by water culture at Nankang, but that on *Leersia* cuttings was lower than the results obtained at Nankang.

The experiment in soil also indicated the phytotoxic effects of powdered plants added (Table 8). By this method, the root-inhibiting effect of *Oryza* powder was stronger than that of *Leersia* powder, particularly on *Leersia* cuttings (Tables 7 and 8). However, the decay of *Oryza* powder in soil was less complete than that of *Leersia* powder, as the soils with *Oryza* powder had a bad smell after a three-day incubation. Therefore, the stronger phytotoxicity of *Oryza* than *Leersia* as observed in this experiment was considered incomparable to others.

Phytotoxicity of Plant Powder in Soil on Plants Emerging from Buried Seeds

After the above soil experiment was over, the soils in trays were kept moist in the glass-house for 92 days (December 1982 to March 1983) to observe the number and biomass of plants occurring from the buried seed pool. The control and treated soils are expected to have had the same composition of weed seeds while germination of the seeds and seedling growth could be affected by the plant powder added; this observation would represent the overall effect of plant residues in the soil on various weedy plants. After the three months of culture (138 days after the addition of plant powder to soil), 10 species were identified, which were

Table 8. *Effects of decomposing plant powders of* ***Oryza perennis*** *and* ***Leersia hexandra*** *in soil on the root growth of* ***O. perennis*** *and* ***L. hexandra*** *cuttings*

Total root length (mm) per cutting of *Oryza perennis* and *Leersia hexandra* cultured in soil with powdered tissues of the two species for 16 days (at Taichung).

Treatment	*Oryza perennis*		*Leersia hexandra*	
	Mean ± σ	%	Mean ± σ	%
Field soil (Control)	188±47.0	100^a	65±25.5	100^a
Plus *Oryza*,				
40 g/kg	80±20.6	43^c	20± 8.4	31^c
80 g/kg	62±15.0	33^c	6± 4.2	9^c
Mean	71±18.0	38^c	13± 6.6	20^c
Plus *Leersia*,				
40 g/kg	118±35.5	63^b	46±12.4	71^b
80 g/kg	88±20.5	47^{bc}	31±21.2	48^{bc}
Mean	103±29.2	55^b	39±17.4	59^b
t(*Oryza* : *Leersia*)	5.89**		8.67**	
LSD (5%)	35.2		27.6	

** Significant at 1% level.
a, b, c—Classes differing at 5% level of significance.
σ—Standard deviation on single determination.

Table 9. *Plants emerging from buried seed pool in soils to which powdered plant tissues were added*

(per 320 cm^2, in trays)

Treatment	Total no. of plants	*Cyperus rotundus*	Total dry wt (mg)	No. of species[a]	H'[b]
Control (field soil)	37	15	27	9	1.62
Plus *Oryza*,					
40 g/kg	38	18	46	7	1.44
80 g/kg	18.5	12.5	15	6	1.07
Plus *Leersia*,					
40 g/kg	27.5	16	25	7	1.26
80 g/kg	13	8.5	6	4	0.94
F value (error df=4),					
Control:Treated	6.2	1.2	0.4	27.5**	10.2*
40:80 g/kg	13.8*	39.3**	18.0*	15.3*	7.0(*)
Oryza : *Leersia*	3.1	8.4*	6.5(*)	0.3	0.7

[a] Total number of species found in 2 replications.
[b] $H' = -\Sigma p_i \ln p_i$, showing species diversity.
*, ** Significant at 5% and 1% levels, respectively;
(*): Close to the 5% level.

in the order of abundance: *Cyperus rotundus* L. (140 plants in total) > *Polygonum plebeium* R. Br. > *Soliva anthemifolia* R. Br. > *Vandellia cordifolia* G. Don. > *Cardamine parvifolia* L. > *Gnaphalium luteo-album* L. > *Eleusine indica* Gaertn. > *Eclipta prostrata* L. > *Cynodon dactylon* Pers. = *Centipeda minima* A. Braun (the last two being only one plant each).

The soils used as the control and those with 40 g/kg of plant powder produced a nearly equal number of plants, but the soils with 80 g/kg of plant powder produced much less plannts, particularly in those with *Leersia* powder (Table 9). The soils with 40 g/kg of *Oryza* powder gave greater plant biomass (dry weight) than the control soils, possibly reflecting the effect of increased nutrients due to the decay of plant powder. Yet, the addition of 80 g/kg powder, particularly of *Leersia*, sharply reduced plant biomass, suggesting the phytotoxic effect of powdered plants on the germination of buried seeds and seedling growth. The differences in these measurements between 40 and 80 g/kg were significant (Table 9). On the number of *Cyperus rotundus* plants which were most dominant, *Leersia* powder showed significantly higher phytotoxicity than *Oryza* powder.

The control soils had the largest number of species and greatest species diversity as shown by information content, $H' = -\Sigma p_i \ln p_i$ (1.62; p_i=proportion of the i-th species). The H' values were greater in the order: Control > 40 g/kg *Oryza* >

40 g/kg *Leersia* > 80 g/kg *Oryza* > 80 g/kg *Leersia* (Table 9). This pattern is comparable to that observed in weed communities on copper-polluted fields as reported by Morishima and Oka (1977, 1980), and suggests that plant residues in the soil can affect plant communities in a similar manner as did polluting agents like copper.

Identification and Quantitative Assay of Phytotoxins

The phytotoxins present in the leaves of *Leersia* and *Oryza* were confirmed by paper and high-pressure liquid chromatographic techniques (Table 10). Eight kinds of phytotoxins were found, which were largely comparably to those found in *Miscanthus floridulus* (Chou and Chung, 1974) and other subtropical grasses (Chou and Yang, 1975). The concentrations of gallic, *p*-hydroxybenzoic, *o*-hydroxyphenylacetic, and *p*-coumaric acids were higher in *Leersia* than in *Oryza*, while those of vanilic, syringic, and ferulic acids were higher in *Oryza* than in *Leersia*. The total concentration of these organic acids in *Leersia* extract was nearly twice as much as that in *Oryza* extract. In view of the reliability of liquid chromatographic analysis so far experienced, the differences in the content of phytotoxins found between the two species could be considered significant. On the whole, it may be asserted that *Leersia* contains more phytotoxins in the leaves than *Oryza*.

An Overall Comparison of Phytotoxicity between ***Oryza*** *and* ***Leersia***

The results of the above-mentioned experiments are not fully consistent with one another in respect of the relative phytotoxicity of the two species. To have

Table 10. *Quantitative comparison of phytotoxins present in the aqueous extracts of* ***Oryza perennis*** *and* ***Leersia hexandra***

Compound	Concentration, $\mu g/g$[a]		*Oryza* / *Leersia* (%)
	Oryza	*Leersia*	
Gallic acid	0.16	0.23	70
Protocatechuic acid	trace	trace	
p-Hydroxybenzoic acid	0.15	0.25	60
o-Hydroxyphenylacetic acid	0.16	0.25	64
Vanillic acid	0.18	0.17	106
Syringic acid	0.015	0.007	214
p-Coumaric acid	0.29	0.99	29
Ferulic acid	0.063	0.020	315
Total	1.018	1.917	53

[a] Based on comparison with the standard curve of each authentic compound run simultaneously with the extract, using a high pressure liquid chromatograph.

Table 11. *Summary of comparisons of phytotoxicity between* ***Oryza*** *and* ***Leersia***

(in % reduction due to treatment)

Method of experiment	Assayed with	% reduction, treated with		Difference *Leersia-Oryza* ($\sin^{-1}\sqrt{\%}$)
		Oryza	*Leersia*	
(Table 1)				
Leachate	Rice seed	25.3	16.9	− 5.9
	Lettuce seed	16.3	8.0	− 7.4
Extract	Rice seed	29.1	33.3	2.6
	Lettuce seed	33.5	39.4	3.5
(Table 3)				
Leachate	*Oryza*, root length	64.2	51.2	− 7.6
	Oryza, root weight	54.2	53.0	− 0.7
	Leersia, root length	47.3	54.9	4.4
	Leersia, root weight	45.1	61.0	9.2
(Table 4)				
Extract	*Oryza*, root length	39.9	62.3	12.9
	Oryza, root weight	21.2	41.5	12.7
	Leersia, root length	65.7	70.0	3.1
	Leersia, root weight	57.3	54.6	− 1.6
(Table 6)				
Extract (A)	*Oryza*, root length	32.3	41.9	5.7
	Leersia, root length	42.8	39.0	− 2.2
Extract (B)	*Oryza*, root length	33.3	56.7	13.7
	Leersia, root length	18.0	41.4	15.0
(Table 9)				
Plant powder in soil (80 g/kg)	Weed, plant number	50.7	64.9	8.7
	Cyperus rotundus number	16.0	43.3	17.1
	Weed dry weight	44.4	77.8	20.1
	Species number	33.3	55.5	13.0
	Sp. diversity (H′)	34.0	42.0	4.7
	Hean	38.3	48.1	5.76
	$\sigma/\sqrt{n}$ (n=21)			1.78
	t			3.42 (P<0.01)

an overall judgment, the percentage reduction of measurements due to treatment observed in the various experiments were compared between treatments with *Oryza* and *Leersia*, excluding the data from the soil experiment which were considered incomparable to others. The *Leersia-Oryza* differences were subjected to *t*-test after transformation of the percentages into arc-sine values (Table 11). Although the assembly of results from different experiments may not be a random sample from a mother population which is a complex of responses, each pair of comparison has a common variance. The *t*-test of null hypothesis showed that *Leersia* had significantly higher allelopathic potential as the whole than *Oryza*. The *Oryza*/*Leersia* ratio was 80% (38.3/48.1 in Table 11), which was higher than that for total concentration of phytotoxins, 53% (Table 10).

Discussion

An experiment on the regenerating success of *O. perennis* populations introduced into different lowland habitats indicated that in many of the habitats, *L. hexandra* came into competition with *O. perennis* and displaced the latter in the second or third year of secondary succession (Oka, 1984). Another experiment by Oka (in preparation) suggested that a deep water condition helped *Oryza* to persist while an application of fertilizers increased the aggressiveness of *Leersia*.

In general, it is known that the neighbor effect between plants results from competition for some limited resource, which may be light (above-ground), water or nutrient (below-ground), or for space and time if integrated (Harper, 1977, Chap. 6). Under a uniform environment, two species (or genotypes of the same species) having the same niche dimension can hardly coexist, while niches become differentiated with succession as the result of natural selection for resource-allocation pattern and other life-history traits (cf. Parrish and Bazzar, 1979). The growth rate at a critical stage at which neighbors begin to interfer in the canopy was found to be an important determinant of the result of interactional competition, while the interaction of roots would start at an earlier stage and affect the growth rate (Donald, 1958; Kawano and Tanaka, 1967; Snaydon, 1971; Assémat *et al.*, 1981).

Allelopathy was considered in the early stage of study as a means of self-defense of particular plants, but it has now become known to play an appreciable role in structuring plant communities in a wide range of environments (Chou, 1980; Numata, 1982). It was detected in plants representing different seral stages of succession: *Erigeron* and *Solidago* species (Numata, 1982), *Miscanthus floridulus* (Chou and Chung, 1974), 12 other grasses (Chou and Young, 1975), bamboos (Chou and Young, 1982), and trees (cf. Grime, 1979, p. 136). However, the evaluation of importance of allelopathy relative to other means of plant interaction remains largely untouched.

A phytotoxin may produce both auto- and allelo-toxic effects, and its metabolism will consume energy. Generally, a particular ability like stress tolerance and performance in favorable environment are inversely associated among plant genotypes, resulting in a trade-off relationship (metal tolerance: Cook *et al.*, 1972;

Hickey and McNeilly, 1975; Morishima and Oka, 1977, 1980; Cyanogenesis: Whiteman, 1973; Dritschilo *et al.*, 1979). Most probably, even though not yet evidenced, there could be a trade-off relationship between allelopathic potential and general performance in non-competitive environments or competitive success due to rapid growth when ecologically similar plants with and without phytotoxic metabolites are compared. From this viewpoint, it may be presumed that allelopathy develops in environments in which competitive strategy or stress-tolerant strategy is selected (cf. Grime, 1979, pp. 51–53).

Autointoxication does not imply a harmful effect on the cells and tissues producing and containing toxic metabolites, but it will suppress sprouting of juveniles from the mother plant, seed germination and seedling growth. The self-regulation of population density by self-thinning and phenotypic plasticity is an important adaptive mechanism of plants. There may be conditions in which natural selection works in the direction of reducing autointoxication and increasing allelopathic potential. Yet, the selective advantage of allelopathy would change according to coexisting plants and community structure, rendering the mode of selection diffuse and disruptive. This might have resulted in the complexity of response patterns as observed in the present study.

Both *O. perennis* and *L. hexandra* grow in marshy habitats in the humid tropics and subtropics which are necessarily crowded by various plants if not disturbed. Such habitats would impose a pressure of density-dependent mortality on the plants resulting in selection for higher carrying capacity (*K*-selection). Presumably, in such a condition, allelopathy helps the plants to survive the struggle for existence to some extent. As mentioned, *O. perennis* shows a perennial-annual continuum, and annual types are found which have life-history characteristics of *r*-selected plants (Oka and Morishima, 1967; Oka, 1976; Sano and Morishima, 1982). They are found in temporary swamps which are parched in the dry season (Oka, 1964; Morishima *et al.*, 1980). Expectedly, the annual types would have a lower allelopathic potential than the perennial type presently studied. If this was evident, the above hypothesis of selection for allelopathy would be partly proven.

Furthermore, in cultivated rice, autointoxication was found to be a factor in lowering the summer (second) crop yield in Taiwan (Chou *et al.*, 1977, 1981; Chou and Chiou, 1979). Since *O. perennis* could be the wild ancestor of the cultivars, an experiment to compare between wild and cultivated rices with regard to allelopathic potential is envisaged.

The present experiments clearly demonstrated that both *O. perennis* and *L. hexandra* contained and exuded phytotoxic metabolites. However, as to their relative allelopathic potentials and interaction, the data were complex. The two species were differentiated to a measurable extent not only in the root-inhibiting effects of, but also in the sensitivities to their leachates and extracts. As an overall judgment, *Leersia* was considered to have a higher allelopathic potential than *Oryza*. Probably, although not investigated presently, the rhizomes of *L. hexandra* release phytotoxic substance into water-logged soils as was observed in *Agropyron repens* (Welbank, 1963). It may be concluded that their allelopathic interaction plays an appreciable role in their successional replacement, but various problems are left for investigation in the future.

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Literature Cited

Assémat, L., H. Morishima, and H. I. Oka. 1981. Neighbor effects between rice (*Oryza sativa* L.) and barnyard grass (*Echinochloa crus-galli* Beauv.) strains II. Some experiments on the mechanisms of interaction between plants. Acta OEcologica **2**: 63-78.

Chou, C. H. 1980. Allelopathic researches in the subtropical vegetation in Taiwan. Comp. Physiol. Ecol. **5**: 222-234.

Chou, C. H., Y. C. Chiang, and H. H. Cheng. 1981. Autointoxication mechanisms of *Oryza sativa* III. Effect of temperature on phytotoxin production during rice straw decomposition in soil. J. Chem. Ecol. **7**: 741-752.

Chou, C. H. and S. J. Chiou. 1979. Autointoxication mechanisms of *Oryza sativa* II. Effects of culture treatments on the chemical nature of paddy soil and on rice productivity. J. Chem. Ecol. **5**: 839-859.

Chou, C. H. and Y. T. Chung. 1974. The allelopathic potential of *Miscanthus floridulus*. Bot. Bull. Academia Sinica **15**: 14-27.

Chou, C. H. and H. J. Lin. 1976. Autointoxication mechanisms of *Oryza sativa* I. Phytotoxic effects of decomposing rice residues in soil. J. Chem. Ecol. **2**: 353-367.

Chou, C. H.., T. J. Lin, and C. I. Kao. 1977. Phytotoxins produced during the decomposition of rice stubbles in paddy soil and their effect on soil leachable nitrogen. Bot. Bull. Academia Sinica **18**: 45-60.

Chou, C. H. and C. M. Yang. 1982. Allelopathic research of subtropical vegatation in Taiwan II. Comparative exclusion of understory by *Phyllostachys edulis* and *Cryptomeria japonica*. J. Chem. Ecol. **8**: 1489-1507.

Chou, C. H. and C. C. Young. 1975. Phytotoxic substances in twelve subtropical grasses. J. Chem. Ecol. **1**: 183-193.

Cook, S. C. H., C. Lefèbvre, and T. McNeilly. 1972. Competition between metal tolerant and normal plant populations on normal soil. Evolution **26**: 366-372.

Donald, C. M. 1958. Competition among pasture plants I. Intraspecific competition among annual pasture plants. Aust. J. Agr. Res. **2**: 355-376.

Dritschilo, W., J. Krummel, D. Nafus, and D. Pimentel. 1979. Herbivorous insects colonizing cyanogenic and acyanogenic *Trifolium repens*. Heredity **42**: 49-56.

Grime, J. P. 1979. Plant Strategies and Vegetation Processes. John Willey & Sons, Chichester (UK), pp. 51-53.

Harper, J. L. 1977. Population Biology of Plants. Academic Press, London, 892 pp.

Hickey, D. A. and T. McNeilly. 1975. Competition between metal tolerant and normal plant populations: A field experiment on normal soil. Evolution **29**: 458-464.

Kawano, K. and A. Tanaka. 1967. Studies on the competitive ability of rice plants in competition. J. Fac. Agr., Hokkaido Univ., Sapporo, Japan **55**(3): 339-362.

Kiang, Y. T., J. Antonovics, and L. Wu. 1979. The extinction of wild rice (*Oryza perennis formosana*) in Taiwan. J. Asian Ecol. **1**: 1-9 (Biol. Dept., Tunghai Univ., Taichung).

Morishima, H. and H. I. Oka. 1977. The impact of copper pollution on weed communities in Japanese rice fields. Agro-Ecosystems **3**: 131-145.

Morishima, H. and H. I. Oka. 1980. The impact of copper pollution on water foxtail (*Alopecurus aequalis* Sobol.) populations and winter weed communities in rice fields. Agro-Ecosystems **6**: 33-49.

Morishima, H., Y. Sano, and H. I. Oka. 1980. Observations on wild and cultivated rices and companion weeds in the hilly areas of Nepal, India and Thailand. Rep. Natl. Inst. Genetics, Misima, Japan, 97 pp.

Numata, M. 1982. Weed-ecological approaches to allelopathy. *In* W. Holzner and M. Numata (eds.), Biology and Ecology of Weeds. Dr W. Junk Publ., The Hague, pp. 169-173.

Oka, H. I. 1964. Pattern of interspecific relationships and evolutionary dynamics in *Oryza*. *In* IRRI (ed.), Rice Genetics and Cytogenetics. Elsevier, Amesterdam, pp. 71-90.

Oka, H. I. 1974. Experimental studies on the origin of cultivated rice. Genetics **78**: 475-486.

Oka, H. I. 1976. Mortality and adaptive mechanisms of *Oryza perennis* strains. Evolution **30**: 380-392.

Oka, H. I. 1984. Secondary succession of weed communities in lowland habitats of Taiwan in relation to the fate of introduced wild-rice (*Oryza perennis*) populations. Vegetatio (accepted)

Oka, H. I. and W. T. Chang. 1961. Hybrid swarms between wild and cultivated rice species, *Oryza perennis* and *O. sativa*. Evolution **15**: 418-430.

Oka, H. I. and H. Morishima. 1967. Variations in the breeding systems of a wild rice, *Oryza perennis*. Evolution **21**: 249-258.

Parrish, J. A. D. and F. A. Bazzaz. 1979. Difference in pollination niche relationship in early and late successional plant communities. Ecology **60**: 597-610.

Rice, E. L. 1974. Allelopathy. Academic Press, New York, 353 pp.

Sano, Y. and H. Morishima. 1982. Variation in resource allocation and adaptive strategy of a wild rice, *Oryza perennis* Moench. Bot. Gaz. **143**: 518-523.

Snaydon, R. M. 1971. An analysis of competition between plants of *Trifolium repens* L. J. Appl. Ecol. **8**: 687-697.

Welbank, P. J. 1963. Toxin production during decay of *Agropyron repens* (Couch grass) and other species. Weed Res. **3**: 205-214.

Whiteman, R. J. 1973. Herbivore feeding and cyanogeneis in *Trifolium repens* L. Heredity **30**: 241-245.

野生稻與李氏禾間潛在的植物相尅作用

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野生稻（*Oryza perennis*）與李氏禾（*Leersia hexandra*）是亞洲熱帶低濕地之重要禾草。在自然族羣中，李氏禾的存在常決定野生稻的消長。兩者間之交互作用機構至爲複雜。本文探討兩者之種內及種間之植物相尅作用潛能。吾人取兩植物之水溶淋洗液（Leachate）及水溶萃取液（Extract）分別對該兩植物幼莖側根生長及對野生稻及萵苣之胚根生長之影響，結果顯示李氏禾之水溶淋洗液及水萃取液中所含植物毒性程度顯著地高於野生稻者，且前者萃取液中所含的植物毒物質之濃度亦較高於後者。該兩者植物之磨粉物混以土壤讓其分解，再觀查所埋入之其他雜草種子之發芽及生長，其結果顯示李氏禾之磨粉物之植物毒性亦較野生稻者高。綜合上述結果顯示，植物相尅作用扮演一個明顯的角色以決定兩者在自然族羣消長中的地位。