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Use of Green Manure Crops in Conrol of *Hirschmanniella mucronata* and *H. oryzae* in Irrigated Rice[1]

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Abstract: Four field experiments were conducted to study the effect of *Sesbania rostrata* and *Aeschynomene afraspera* as rotational and green manure crops on the population dynamics of *Hirschmanniella mucronata* and *H. oryzae,* and subsequent rice yields. The sequential cropping of the legumes with rice controlled both nematode species. In two experiments, yield of rice was related to the nematode population denisites at planting and harvesting of the second rice crop ($R^2 = 0.391$, $P < 0.001$, and $R^2 = 0.57$, $P < 0.001$), regardless of the treatments. Rice yield increases were attributed to nutritional effect of the green manure and the reduction of the nematode populations or the modification of a factor(s) linked to the nematode populations induced by their cropping. As the two leguminous crops do not generate direct return, using them to control the rice-root nematodes was not economical, despite the significant yield increase obtained.

Key words: Aeschynomene afraspera, control, green manure, *Hirschmanniella mucronata, Hirschmanniella oryzae,* nematode, *Oryza sativa,* rice, *Sesbania rostrata,* yield.

The rice-root nematodes *Hirschmanniella* spp. infest most of world's irrigated rice fields (7). They are pathogenic to rice and cause yield losses (1,3,4,11). Chemical control of nematodes in irrigated rice is seldom economical (8), and other management methods need to be developed.

Rice-root nematodes can be controlled with green manure legume crops. *Sphenoclea zeylanica* produces toxic plant exudates and can provide 99% control of *Hirschmanniella* spp. (10). *Aeschynomene afraspera* Leon. and *Sesbania rostrata* Brem. are potential green manure crops (2,14) that may be used in rotation with irrigated rice. They also have potential for control of rice-root nematodes (5,6). A microplot experiment (5) indicated that it was not necessary to incorporate *S. rostrata* as green manure to significantly increase the yield of the following rice crop. In this experiment, yield increase was attributed to the control of *Hirschmanniella oryzae* (Van Breda de Haan) Luc & Goodey, the leguminous crop acting as a trap crop for the nematode (5,12,13). The trapping action of *S. rostrata* starts 8 weeks after germination; the nematodes that have penetrated, developed, and reproduced inside the roots remain trapped and are destroyed within the roots (12). *Aeschynomene afraspera* seems to control the rice-root nematode in the same way: The nematodes penetrate the roots but are unable to exit them and are not recovered from them after 60 days (6). The mechanisms involved in nematode root exit inhibition and nematode destruction inside the root remain unknown.

The present investigation was undertaken to determine the effects of planting and (or) incorporating *S. rostrata* and *A. afraspera* on nematode populations and on rice (*Oryza sativa* L.) yield in irrigated fields infested with *Hirschmanniella mucronata* (Das) Luc & Goodey or *H. oryzae.*

MATERIALS AND METHODS

Four experiments were conducted on the experimental farm of the International Rice Research Institute (IRRI) in two fields with average initial population densities of 884 *H. mucronata*/1,000 cm^3 soil in field 1 and 539 *H. oryzae*/1,000 cm^3 soil in field 2. Experiments 1 and 3 were conducted in field 1 and experiments 2 and 4 in field 2. Treatments were arranged in a randomized complete block design with seven replications in experiment 1 and five replica-

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tions in experiments 2, 3, and 4. Individual plot area was 20 m^2. The rice cultivar IR 58 was used for all four experiments. Rice was grown for 90 days and *S. rostrata* and *A. afraspera* were grown for 120 days. The fields were permanently flooded throughout the growing seasons.

Treatments tested during first and second experiments were as follows: rice grown continuously; incorporation of shoots of *S. rostrata* or *A. afraspera* following the first rice crop (shoots were chopped and then mixed with the soil during a 25 cm deep plowing); removal of shoots of *S. rostrata* or *A. afraspera* following their growth as the first crop; and incorporation of shoots of *S. rostrata* or *A. afraspera* as green manure following their growth as the first crop. In the first experiment, 7 t/ha of *S. rostrata* and 14 t/ha of *A. afraspera* were incorporated. In the second experiment, 10 t/ha and 7 t/ha of *S. rostrata* and *A. afraspera* were used, respectively. No fertilizer was applied in experiments 1 and 2. In experiment 1, plants were sprayed once with a contact insecticide, 2-(1-methylpropyl)phenylmethylcarbamate, at the rate of 0.4 kg a.i./ha.

The cropping sequence and the fertilizer treatments applied during the third and fourth experiments were as follows: rice was grown continuously; sixty kg of nitrogen in the form of ammonium sulfate broadcasted at transplanting of the second rice crop; nitrogen (60 kg/ha) applied twice, at transplanting of the second crop and at maximum tillering; shoots of *S. rostrata* incorporated as green manure; removal of shoots and roots of *S. rostrata* after first growing season; removal of *S. rostrata* after first growing season; *S. rostrata* plants uprooted and then shoots but not roots incorporated into the soil; and roots and shoots of *S. rostrata* incorporated into the soil after the first growing season. Total nitrogen contained in *S. rostrata* used as green manure in experiment 3 and 4 was determined with the Kjeldahl method by the IRRI Analytical Laboratory. The nitrogen content of the green manure applied was equivalent to 296 and 271 kg of nitrogen per ha in experiments 3 and 4, respectively. Cypermethrin was sprayed three times at the rate of 0.05 kg a.i./ha to control *Nephotettix virescens* Distant, a vector of tungro virus.

Five samples of root and soil were collected at random from each plot to estimate the nematode population densities before planting and at harvest of each crop. At each sampling site, a hill was uprooted and 3 g of roots, together with 200 cm^3 of rhizosphere soil, were collected. In experiments 3 and 4, the initial nematode population densities were estimated at harvest of the preceding rice crop. Each soil and root sample was processed separately. Nematodes were extracted from 200 cm^3 soil by a combination of sieving and modified Baermann funnel techniques. Nematodes were extracted from 3 g of roots by macerating them with a blender and placing them for 48 hours in Baermann funnels.

In experiments 1, 2, and 4, yields of the second rice crop were estimated by harvesting the central 6 m^2 of each plot. Because of partial rat damage, yields in experiment 3 were estimated on a per hill basis (50 randomly selected undamaged hills per plot) rather than on a per plot basis.

Results and Discussion

Except for two treatments, one in experiment 2 (Table 2) and one in experiment 4 (Table 4), when *S. rostrata* and *A. afraspera* were incorporated in plots where rice was the previous crop, the yield of the second rice crop was higher ($P < 0.05$) than in the control plots (continuous rice cropping) (Tables 1–4). This indicated a fertilizer effect when the legumes were used as green manure.

With the exception of experiment 3, when all residues of *S. rostrata* were removed after the first crop (Table 3), the population densities of *H. mucronata* and *H. oryzae* were lower ($P < 0.05$) in plots

TABLE 1. Effects of growth of *Sesbania rostrata* and *Aeschynomene afraspera* and incorporation of crop residues on rice yields and *Hirschmanniella mucronata* population densities per 1,000 cm³ soil and per g fresh root weight in experiment 1.

Treatment†			*H. mucronata* population					
			Initial	Final				
				First crop		Second crop		Rice yield
1st crop	2nd crop	Incorporation of residues	Soil	Soil	Root	Soil	Root	(kg/ha)
R	R	−	860 a	766 a	6 a	783 a	12 a	2,198 e
R	R	+S	1,024 a	1,055 a	8 a	621 ab	21 a	3,202 bc
S	R	−S	775 a	9 b	0 b	446 ab	13 a	2,933 cd
S	R	+S	750 a	12 b	0 b	352 b	11 a	3,979 a
R	R	+A	881 a	692 a	11 a	598 ab	17 a	3,511 ac
A	R	−A	1,050 a	1 b	0 b	274 b	6 a	3,022 cd
A	R	+A	849 a	1 b	0 b	262 b	7 a	3,764 ab

Values are means of seven replicates. Means in each column followed by the same letter do not differ at $P < 0.05$ according to Duncan's multiple-range test.
† R = rice; S = *S. rostrata;* A = *A. afraspera;* − = no incorporation of crop residues; + = incorporation of crop residues.

where *S. rostrata* or *A. afraspera* were grown than in plots continuously cropped with rice. This confirmed previous results obtained in pots (12), microplots (5,12,13), and under field conditions (6) indicating that growing these two legumes efficiently reduced population densities of *H. mucronata* and *H. oryzae*. In experiments 3 and 4, the nematode population densities after the first crop in plots where *S. rostrata* had been uprooted were not different ($P < 0.05$) from those observed in plots where only the above-ground parts of the leguminous crop had been removed (Tables 3, 4). This result indicates that the nematodes present within the roots of the legumes were not able to exit even after the decay of these roots. In addition, in experiments 2, 3, and 4, the nematode population densities were reduced ($P < 0.05$) by the incorporation of shoots of *S. rostrata* and *A. afraspera* in plots where these plants had not been grown. The two legumes, therefore, appeared to control the rice-root nematodes during their growth and after their incorporation. During their growth, the plants may act as trap crops (5,12,13) or release metabolites detrimental to

TABLE 2. Effects of growth of *Sesbania rostrata* and *Aeschynomene afraspera* and incorporation of crop residues on rice yields and *Hirschmanniella oryzae* population densities per 1,000 cm³ soil and per g fresh root weight in experiment 2.

Treatment†			*H. oryzae* population					
			Initial	Final				
				First crop		Second crop		Rice yield
1st crop	2nd crop	Incorporation of residues	Soil	Soil	Root	Soil	Root	(kg/ha)
R	R	−	771 a	708 a	31 a	465 a	7 a	1,183 c
R	R	+S	500 a	705 a	28 a	40 b	1 b	3,036 a
S	R	−S	494 a	1 b	0 b	0 b	1 b	2,796 ab
S	R	+S	472 a	4 b	0 b	24 b	1 b	3,190 a
R	R	+A	427 a	606 a	14 a	24 b	7 ab	2,436 abc
A	R	−A	526 a	1 b	0 b	8 b	1 b	3,118 a
A	R	+A	586 a	1 b	0 b	2 b	0 b	2,914 ab

Values are means of five replicates. Means in each column followed by the same letter do not differ at $P < 0.05$ according to Duncan's multiple-range test.
† R = rice; S = *S. rostrata;* A = *A. afraspera;* − = no incorporation of crop residues; + = incorporation of crop residues.

TABLE 3. Effects of *Sesbania rostrata* growth and subsequent incorporation on rice yields and *Hirschmanniella mucronata* population densities per 1,000 cm³ soil and per g fresh root weight in experiment 3.

Treatment†			*H. mucronata* population						
			Initial		Final				
					First crop		Second crop		
1st crop	2nd crop	Fertilizer or crop residue	Soil	Root	Soil	Root	Soil	Root	Rice yield (g/hill)
R	R	−	513 a	22 a	394 bc	80 ab	390 a	16 a	15.4 d
R	R	F	192 a	13 a	705 a	59 b	168 b	19 a	16.7 cd
R	R	2F	606 a	7 a	689 a	55 b	212 b	12 abc	17 cd
R	R	+S	827 a	32 a	628 ab	105 a	123 bc	14 ab	18.3 bc
S	R	−S(total)	181 a	8 a	204 cd	0 c	64 c	4 c	19.6 ab
S	R	+Sr	363 a	12 a	46 d	0 c	25 c	4 c	19.8 ab
S	R	+Ss	485 a	10 a	95 d	0 c	37 c	3 c	21.2 a
S	R	+S(total)	474 a	16 a	38 d	0 c	26 c	5 c	21.4 a

Values are means of five replicates. Means in each column followed by the same letter do not differ at $P < 0.05$ according to Duncan's multiple-range test.

† R = rice; S = *S. rostrata;* − = no incorporation of crop residues or fertilizer; F = one application of fertilizer; 2F = two applications of fertilizer; +S = incorporation of shoots of *S. rostrata;* −S(total) = removal of roots and shoots of *S. rostrata* after first crop; +Sr = incorporation of *S. rostrata* roots after removal of shoots; +Ss = incorporation of *S. rostrata* shoots after roots were removed; +S(total) = incorporation of roots and shoots of *S. rostrata* after first crop.

nematodes. After incorporation, the decaying plant tissues may release nematicidal or nematostatic products or sustain the development of organisms antagonistic to nematodes.

The growing of both leguminous crops, regardless of incorporation of their aboveground parts in the soil, always resulted in an increase in yield ($P < 0.05$). The incorporation of these legumes in plots where they had been planted usually resulted in higher yields. However, except in experiment 1, these yields were not different ($P > 0.05$) from those obtained in plots where they were grown without subsequent incorporation.

These results indicate that these legumes may affect the yield of a following rice crop in two different ways. A nutritional effect was observed when these

TABLE 4. Effects of *Sesbania rostrata* growth and subsequent incorporation on rice yields and *Hirschmanniella oryzae* population densities per 1,000 cm³ soil and per g fresh root weight in experiment 4.

Treatment†			*H. Oryzae* population						
			Initial		Final				
					First crop		Second crop		
1st crop	2nd crop	Fertilizer or crop residue	Soil	Root	Soil	Root	Soil	Root	Rice yield (kg/ha)
R	R	−	17 a	1 a	258 a	29 a	160 a	70 a	3,368 e
R	R	F	104 a	4 a	196 a	24 a	67 b	45 b	4,046 cd
R	R	2F	88 a	2 a	267 a	39 a	67 b	26 c	4,210 bcd
R	R	+S	51 a	2 a	207 a	21 a	39 bc	17 cd	3,663 de
S	R	−S(total)	110 a	2 a	35 b	0 b	23 c	11 cd	4,415 bc
S	R	+Sr	13 a	1 a	35 b	0 b	9 c	4 d	5,125 a
S	R	+Ss	11 a	0 a	33 b	0 b	22 c	5 d	4,785 ab
S	R	+S(total)	147 a	2 a	24 b	0 b	15 c	5 d	4,623 abc

Values are means of five replicates. Means in each column followed by the same letter do not differ at $P < 0.05$ according to Duncan's multiple-range test.

† R = rice; S = *S. rostrata;* − = no incorporation of crop residues or fertilizer; F = one application of fertilizer; 2F = two applications of fertilizer; +S = incorporation of shoots of *S. rostrata;* −S(total) = removal of roots and shoots of *S. rostrata* after first crop; +Sr = incorporation of *S. rostrata* roots after removal of shoots; +Ss = incorporation of *S. rostrata* shoots after roots were removed; +S (total) = incorporation of roots and shoots of *S. rostrata* after first crop.

crops were incorporated as green manure. *Hirschmanniella* species are pathogens of rice (1,3,4), and their control may result in yield increase (8). Therefore, part of the enhancement of the productivity of the ecosystem induced by the legume crops could be attributable to the control of the rice-root nematodes or to other limiting factor(s) linked to the nematode population density. This was confirmed in experiments 3 and 4 by the relationships between rice yields obtained in individual plots and the nematode population densities at planting and harvesting of the second rice crop regardless of the treatments. Step-wise regression analyses were conducted using the relative yield (ratio of actual plot yield to the mean yield of the experiment) as the variable to be explained and a series of explanatory variables: the initial and final nematode populations, the amount of organic or inorganic nitrogen applied, and their interactions. The results indicate differences in the regression models obtained. In experiment 4, a simple equation was found that illustrates the yield-reducing effects of nematodes:

$$y = 1.114 - 10^{-4}\,(3.72\ \mathrm{Psi} + 26.4\ \mathrm{Prf}) \quad (R^2 = 0.391,\ P < 0.001),$$

where y = relative yield, Psi = initial nematode soil population, and Prf = final nematode root population.

Experiment 3 yielded a much more complex equation indicating significant interactions between nematode populations and nitrogen sources.

$$y = 1.095 - 10^{-6}\,(1.46\ \mathrm{No} \cdot \mathrm{Psi} + 2.19\ \mathrm{Ni} \cdot \mathrm{Psi}) - 0.0144\ \mathrm{Prf} + 10^{-5}\,(9.39\ \mathrm{No} \cdot \mathrm{Prf} + 13.5\ \mathrm{Ni} \cdot \mathrm{Prf}) \quad (R^2 = 0.57,\ P < 0.001),$$

where: y = relative yield, Psi = initial nematode soil population, Prf = final nematode root population, No = organic nitrogen, and Ni = inorganic nitrogen.

Fortuner (4) indicated that fertilizer application may partly compensate for yield loss from rice-root nematodes. In experiment 3, the last term of the equation obtained by step-wise regression analyses indicates a compensatory effect of nitrogen on the damage caused by nematodes.

Despite a significant increase in yield, growing *S. rostrata* for 120 days to control rice-root nematodes should not be considered economically feasible. The yield increase obtained during the second rice crop does not compensate the yield of a rice crop lost due to the growing of the legumes. The maximum duration for growing a leguminous green manure (LGM) in rotation with irrigated rice is 45–55 days; if a longer time is required, economics are in favor of a crop that has a market value (9). To achieve a high level of control of *Hirschmanniella* spp., it is necessary to grow *S. rostrata* for at least 60 days and preferably 90 days (6,12) when the legume is grown in rotation with rice. To save land, time, and water, LGM may be intercropped with rice at the beginning or the end of the rice crop cycle (15). Associating LGM at the beginning of the crop will not achieve nematode control because they will have invaded the rice roots. Seeding LGM into the standing rice 2 to 3 weeks before harvest will reduce time loss by a maximum of 21 days. However, 70 more days of LGM growth would still be necessary to achieve a high level of nematode control. Therefore, in areas where incorporation of organic matter is not essential to reclaim degraded soils, where cover crops are not necessary to protect the soil from erosion, or where LGM does not have a market value, its use to control rice-root nematodes would be difficult to justify.

Results obtained indicate that 1) the rice yield increase obtained after a rotation with *S. rostrata* and *A. afraspera* did not arise solely from a fertilizer effect of the LGM; 2) rice-root nematodes may cause yield losses of more than 25% in irrigated rice; and 3) *Hirschmanniella* spp. may be controlled by rotation with LGM. Because using LGM to control rice-root nematodes

does not appear to be economical, other technologies must be developed.

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