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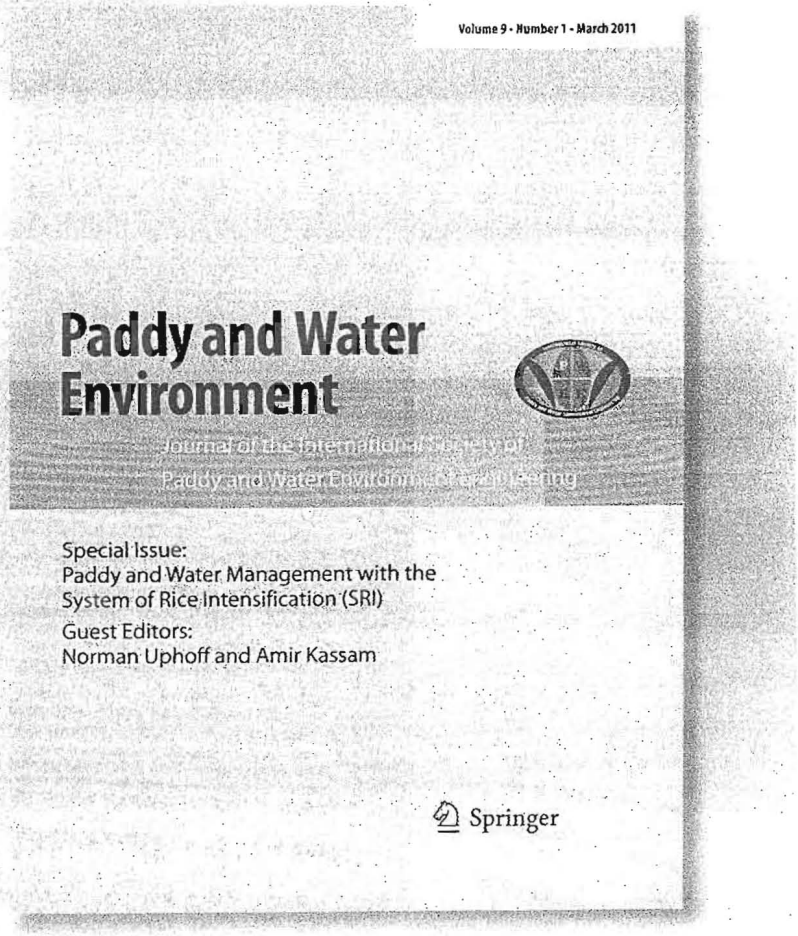

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REVIEW

A review of studies on SRI effects on beneficial organisms in rice soil rhizospheres

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Abstract This communication reports on separate research efforts in India and Indonesia to evaluate the effects that modifying methods of plant, soil, water and nutrient management could have on populations of soil organisms, particularly on those that can have beneficial consequences for crop growth and yield. Comparison of these parallel studies (Table 7) draws attention to the impacts that management can have on the soil biota, given that certain organisms are known to have positive implications for plants' nutrition, health, and productivity. Data from the three studies show SRI management associated with some significant differences in soil microbial populations; higher levels of enzyme activity in SRI plant rhizospheres, indicative of increased N and P availability; and more soil microbial C and N, which would enlarge the nutrient pool for both plants and microbes. The studies reported, although more exploratory than conclusive, show enough similarity to suggest that SRI practices, which make paddy soils more aerobic and enhance soil organic matter, are supportive of enhanced populations of beneficial soil organisms. If this relationship is confirmed by further assessments, it could help researchers and practitioners to improve paddy production in resource-conserving, cost-effective ways. This review was written to encourage more studies to assess these kinds of soil biotic relationships and dynamics.

Keywords Agriculturally beneficial soil microbes · Microbial biomass carbon · Microbial biomass nitrogen · Phosphate-solubilizing microorganisms · Rhizosphere · Root mass · Root length density · Soil biology · System of rice intensification (SRI)

Introduction

Farmers in a number of countries have been able to increase the yields from their current rice varieties with available resources by utilizing what is known as the system of rice intensification (SRI) (Kabir and Uphoff 2007; Namara et al. 2008; Sato and Uphoff 2007; Sinha and Talati 2007). Higher productivity is achieved by making certain changes in the management of rice plants and the resources upon which these depend—soil nutrients, air, water, soil biota, and solar energy (Ceesay et al. 2007; Lin et al. 2009; Thakur et al. 2010; Zhao et al. 2009).

The changes in practice that constitute this alternative cultural system for growing rice are reviewed in the "Discussion" section. There we consider how SRI management could affect two principal factors that apparently contribute to the productivity gains observed: enhanced size and functioning of *plant root systems*, and more

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abundant and diverse *soil biota* (Mishra and Salokhe 2008; Thakur et al. 2010; Zhao et al. 2010). That SRI agronomic concepts and practices are being extended successfully now also to rainfed rice cultivation and to other crops besides rice (http://sri.ciifad.cornell.edu/aboutsri/othercrops/index.html) suggests that there are positive impacts on the functioning of soil systems and/or on plant-soil interactions rather than just on the crop plants themselves.

It is relatively easy to observe and measure the above-ground effects of SRI practices, considering such parameters as plant height, tiller number, and grains per panicle. Increases in these parameters, however, must be supported by below-ground changes, which are more difficult to assess. Determining the length and volume of root systems is challenging enough, requiring their careful extraction from the soil. But such measurements are simpler than gaining details about the populations of soil organisms. Ascertaining their number, activity, and diversity is much more complex and ambiguous. For this reason, relatively little is known about the effects of SRI practices on the soil biota and about its effects, in turn, on SRI crop performance.

Several studies have documented the effects of SRI management practices on root development and functioning, e.g., Mishra and Salokhe (2008) and Thakur et al. (2010). Fewer have focused on the effects of these practices on the soil biota, exceptions being Sooksa-Nguan et al. (2009) and Zhao et al. (2010). In this issue of *PAWE*, Mishra and Salokhe (2011) and Lin et al. (2011) present some further evidence on this subject.

Here we report on the findings from research that looked at the effects of changes in water regime and associated SRI practices on microbial populations and activity in the rhizosphere soil of rice plants. The studies were done at three different locations, at Tamil Nadu Agricultural University (TNAU) and the International Crops Research Institute for the Semi-Arid Tropics (ICRISAT) in India, and at the national agricultural university in Indonesia, Institut Pertanian Bogor (IPB). Because this is a review of three research undertakings, the presentation reports on the materials and methods used in each, and then on the results from the respective studies.

The research question was: how might altering cultural practices for paddy rice production influence the populations of soil organisms in rice plant rhizospheres. We were interested first in *number*—whether there are significantly more of certain organisms—and second in *composition*—whether some species that are beneficial to crop performance are more abundant. The three studies are interesting to consider together. Their results have been reported in a previous paper with several other colleagues (Uphoff et al. 2009). Here, we want to consider their implications for better understanding the SRI, with the hope that comparative consideration of the findings may interest others to examine these effects further.

Since soil biological parameters vary so widely—and are so sensitive to even small changes in soil chemical, physical, thermal and hydrological conditions—these studies must be regarded as exploratory. Firmer conclusions will need to await many similar studies done in various locations, to get a better grasp of the range and distribution of relationships, and to see if any reliable central tendencies may emerge.

Materials and methods

The three studies reported below were done separately, so their materials and methods are somewhat different, although parallel. We report first, in chronological order, how the studies were conducted. They were not replications, having been designed without any communication among the researchers. However, their similarity of results makes them worth others' consideration. The observations will, we hope, encourage other researchers to assess the various magnitudes and contingent relationships more thoroughly, to get a better understanding of interactions among cultural practices, soil biota, and crop responses.

Studies in Tamil Nadu, India

Evaluations of the effects of SRI practices on rice plant roots, on associated soil biota, and on soil biochemical activity were initiated at Tamil Nadu Agricultural University in 2001, under the direction of the third author (Thiyagarajan) who was at the time director of TNAU's Centre for Crop and Soil Management Studies. He supervised two theses (Gyathry 2002; Nisha 2002) that assessed differences in roots' functioning and in soil biological activity in response to SRI versus conventional management at different stages of crop growth. These unpublished results are cited here with full credit to their authors.

Field experiments were conducted at the wetlands of the TNAU university farm in the wet season, September 2001–January 2002, with the hybrid variety CORH2 (125 days duration), and then in the dry season, February–June, 2002, with the hybrid variety ADTRH1 (115 days duration). The experimental site where the trials were conducted had clay-loam soil with pH of 8.3, electrical conductivity of 0.54 dSm^{-1}, organic carbon content of 8.2 g kg^{-1}, available N ($KMnO_4$-N) of 232 kg ha^{-1}, Olsen-P of 32 kg ha^{-1}, and available K (NH_4OAc-K) of 740 kg ha^{-1}.

In both experiments which evaluated the effects of conventional versus SRI practice, four management components were assessed, each with two levels. For all treatments, plant spacing was 20 × 20 cm with single

plants hill^{-1} (plant population of 25 plants m^{-2}), so plant density was not evaluated as a factor. Square planting permitted the crisscross use of a rotary weeder.

Treatment combinations were replicated four times, with gross plot size in both seasons of 26.4 m^2; the net plot size was 13.5 and 13 m^2 for the wet and dry season, respectively. Irrigation water use from transplanting time onwards was measured using a parshall flume, and rainfall during the growing season was monitored. Nurseries were started with 10-day staggering to permit varying of seedling age without introducing variations in the climate that could affect each set of treatments.

The respective management treatments were as follows

- Conventional practice: 24-day-old seedlings with the above plant density; plots were irrigated to a 5 cm depth 1 day after disappearance of ponded water; hand weeding was done three times; recommended fertilizers were applied: 150 kg ha^{-1} N, 60 kg ha^{-1} P_2O_5, 90 kg ha^{-1} K_2O, plus Zn. The P was applied basally, while N was applied in four splits: 40% basal and 20% each at active tillering, panicle initiation and first flowering stages. The K was applied in three splits: 50% basal and 25% each at tillering and panicle initiation stages.
- SRI practice: 14-day-old seedlings with the above plant density; 2 cm irrigation water was applied after hairline cracks appeared in the soil surface up to panicle initiation (PI); then after PI, irrigation was given 1 day after disappearance of ponded water. Inter-cultivation was done five times with a rotary weeder at 10-day intervals. The same recommended fertilizer was applied as with conventional practice, plus 6.25 t ha^{-1} green leaf manure.

We note that this was not a comparison of the full SRI methodology. Rather, the research was considering the combined effects of (a) transplanting younger seedlings, with (b) more active soil aeration and increased soil organic matter, provided by the green manure and by a weeding method that put weeds back into the soil rather than remove them, and (c) alternative water management regimes that introduced differences in passive soil aeration.

The measurements made from the experimental plots used standard methods: root length and volume (using root scanner CI203-RC and measuring cylinder); cation exchange capacity (CEC) of the roots (Crooke 1964); ATPase activity (Unbeit 1964); cytokinin content (Hansen et al. 1984); population density of bacteria (*Azotobacter*, phosphobacteria, and total diazotrophs in the rhizosphere) was enumerated by using serial dilution plate technique (Parkinson et al. 1971) and by most probable number (MPN) technique for *Azospirillum* (Okon et al., 1977); plus activity levels for dehydrogenase (Casida et al. 1965), urease and alkanine phosphate (Tabatabai and Bremner 1969), and nitrogenase in the rhizosphere soil (Rao and Venkateswarlu 1982).

The media used for enumeration of microorganisms were: nutrient agar medium (heterotrophic bacteria), N-free malate medium (*Azospirillum*), Waksman 77 medium (*Azotobacter*), and specific medium for diazotrophs, Sperber's hydroxyl apatite medium (phosphobacteria). The plates were incubated for appropriate period of time at 30°C, and populations were counted and expressed as gram per dry weight of the soil.

Studies in Andhra Pradesh, India

Starting in 2004, the World Wide Fund for Nature (WWF) began supporting SRI evaluation and then its dissemination in Andhra Pradesh state of India, working with the state agricultural university (ANGRAU), the Directorate of Rice Research of the Indian Council for Agricultural Research, and the International Crop Research Institute for the Semi-Arid Tropics (ICRISAT), through its Dialogue Project on Food, Water and the Environment with ICRISAT. The second author (Rupela) headed up the studies of soil biology under this collaborative research effort.

Some 200 farmers across 11 districts of the state participated in on-farm evaluations, using SRI practices and conventional practices on the same farms, which minimized effects from farmer or soil differences. ICRISAT undertook soil biology and root studies with 27 of these farmers in three of the districts in different parts of the state (Anantapur, Medak, and West Godavari). These were farmers willing to cooperate over four seasons, starting in the post-rainy (rabi) season 2004/05 and going through the rainy (kharif) season 2006. As it turned out, not all of these farmers were able to participate in the evaluation in all four of the seasons covered because of difficulties in water availability caused by no rain or by interruptions in irrigation.

The purpose of this research, a component of the larger study, was to assess what if any correlations would be found between soil biological parameters and observed differences in yield associated with SRI management (Rupela et al. 2006). As this was an on-farm study there were no replications; however, the study was carried out under realistic field conditions, and the soil analyses were done in ICRISAT facilities according to international standards.

Agronomic practices and inputs that could affect soil properties, such as fertilizers and compost, were recorded through a questionnaire the first two seasons, with additional comments or observations on crop and soil status

recorded during soil sampling at vegetative and harvest stages after discussions with the participating farmers.

Soil samples were collected from both SRI and control plots of the participating farmers at three times in each season: (a) at or before sowing, (b) at about 60 days after sowing, and (c) close to harvest. In the rainy season 2006, soil samples were collected from fields of four farmers each in two districts (West Godavari and Medak), first before sowing and then about 60 days later to make studies of various soil properties: chemical, microbiological, and biological.

Soil sampling was done using a 40 mm diameter soil corer. Each field sample was a pool of three or five subsamples from as many spots, with pooling done depth-wise. Part of the pooled samples were placed in fresh polythene bags soon after mixing and stored in an ice-cooled thermocol box and transported to the lab separately. These bags were transferred to a fridge at 4–10°C until used for microbiological analyses.

The rest of the pooled samples were air-dried under shade, pounded to break up large clods, sieved (<2 mm), and stored at 10°C until used for analyzing three soil chemical parameters (total N, total P, and % organic C), six soil microbiological measurements (population of total bacteria, total fungi, siderophore producers, N_2-fixing bacteria, P-solubilizing bacteria, *Pseudomonas fluorescens*), and three soil biological indicators (dehydrogenase, microbial biomass carbon [MBC], and microbial biomass nitrogen [MBN]).

Chemical properties

These were evaluated by the wet digestion method of Walkley and Black (1934, rapid titration method) for organic carbon (OC%); modified Kjeldahl method (Dalal et al. 1984) for total N (kg ha^{-1}), and colorimetric method (Olsen et al. 1954) for available P (kg ha^{-1}).

Microbiological properties

Enumeration of microorganisms present in soil samples was done on six different media using pour-plate method. Appropriate dilutions were plated on Luria agar for bacteria; on ¼ PDA + streptomycin (500 mg l^{-1}) for fungi; on Pseudomonas isolation agar (PIA) for *Pseudomonas fluorescens*; on sucrose medium for N_2-fixers; on chromazurol-S (CAS) agar medium for siderophore producers; Pikovskaya's medium with Benomyl (100 mg l^{-1}) for P-solubilizers was added to suppress fungi. The plates were incubated at 30 ± 1°C for 24 to 72 h.

Colonies with desired traits on the different media were counted and recorded. Data were log transformed and expressed as colony-forming units (CFU) $\log_{10} g^{-1}$ dry soil. Moisture in the different soil samples was determined, and the counts were converted to per gram dry soil. Isolates having at least one beneficial trait and/or representing all available diversity in colony morphology were isolated for further studies. Cultures having two to three beneficial traits were preserved in 20% glycerol at −13°C for further studies.

Biological properties

Moisture content of soil samples was adjusted to 55% water-holding capacity (WHC). Twenty gram (dry weight equivalent) of this soil was taken in 50-ml beakers in duplicates. One set was fumigated with chloroform ($CHCl_3$) and the other set was non-fumigated. Fumigation was done by placing the beakers in a large vacuum desiccator along with a beaker containing 50 ml of alcohol-free chloroform ($CHCl_3$) and anti-bumping granules. Evacuation was done with help of a vacuum pump until the chloroform started boiling, which was allowed for 1–2 min. The desiccator was then tape-sealed and placed in an incubator at 25°C for 24 h. Unfumigated samples were also kept in a desiccator and placed in the same incubator at 25°C to maintain similar conditions.

After incubation, the desiccator was made free of vacuum. After 24 h, the soil samples and a vial containing 20 ml of 1 N NaOH were placed in closed air-tight mason jars (one soil sample and one vial per jar) and were incubated at 25°C for a period of 10 days in order to determine biomass C. Amount of CO_2 evolved during the period of 10 days' incubation was absorbed in a known volume of 1 N NaOH. To know the amount of CO_2 absorbed by the aliquot of alkali, titration was carried out by using an automated titrator (Metohm, Germany). Microbial biomass carbon (MBC) was calculated by using Anderson and Domsch (1989) method. The microbial biomass nitrogen was measured following kjeldahl distillation method and was calculated as given by Jenkinson (1988). Soil dehydrogenase assay was determined by the method described by Casida (1977).

Root studies

Roots studies were done on plant samples collected during two of the four seasons, first in the post-rainy 2005/06 and then in the rainy season 2006. A total of 12 partner farmers, four in each of the three districts, were involved in the root studies. Root samples were collected close to harvest from both types of rice, from the top 30 cm soil profile in the post-rainy season 2005/2006 and from two depths (0–15 and 16–30 cm) in the rainy season 2006. For this purpose, soil from three spots (each of 0.5 m × 0.5 m) per treatment plot was excavated. The soil dug out was placed in

big buckets to become slurry in excess water and was passed through 2 mm sieve to collect roots and other debris and stored in plastic bags. The root samples were brought to the lab, washed and cleaned to remove debris. Root length density was measured using a root length scanner (EPSON expression 1640XL, Japan). The same roots were then used for measuring wet mass and were kept in an oven at 70°C for 3 days before taking dry mass.

Yield was assessed by multiple plot crop-cuttings of one meter square from each of the two treatment plots of a given farmer in all of the four seasons. In some cases, these data were checked against the data provided by participating farmers. The data values for a given parameter from all farmers' fields were averaged in each given season. The four seasons were treated as four replications of the two treatments being evaluated, i.e., SRI and conventional flooded rice, the latter considered as control. The data were analyzed using Genstat package 9.1.

Studies at the Institut Pertanian Bogor in Indonesia

More recently, from 2007, researchers at the Bogor Agricultural University (IPB) in Indonesia have started doing soil biology studies to evaluate SRI crop management with regard to greenhouse gas emissions from SRI vs. conventional plots. These studies under the direction of the first author (Iswandi) have assessed roots and soil biota in replicated treatments. These compared (a) conventional rice production methods using NPK fertilizer and organic fertilization, with (b) SRI practices using organic fertilization or a combination of NPK fertilizer and a bioorganic fertilizer (BF). The latter is a commercially available product known as FERTISMART, produced by PT Kujang Amanah Tani based in Purwakarta, West Java. This composite mixture is advertised as containing rock phosphate and dolomite (calcium magnesium carbonate), plus large numbers of beneficial microorganisms, N-fixing bacteria (*Azotobacter* spp., *Azospirillum* spp.) and phosphate-solubilizing fungi (*Aspergillus niger*).

The variety of rice planted in all of the trials was Ciherang, a national high-yielding variety with a crop cycle usually of 115 days. The set of practices considered as conventional rice cultivation involved continuous flooding of the plots, which had been planted with 30-day seedlings, six seedlings per hill, and 20 × 20 cm spacing between hills. Plot size was 4 m × 5 m. The plots had either inorganic or organic fertilization (T0 and T1 as discussed below). SRI cultivation, in contrast, used 10-day seedlings planted one hill^{-1} at 30 × 30 cm spacing, with intermittent irrigation at intervals of 5 days. These practices were assessed with organic fertilization (T2) or a combination of inorganic and organic fertilizers (T3).

The four different fertilization treatments were as follow:

- T0: Inorganic NPK (250 kg urea ha^{-1}, 200 kg single superphosphate (SP) ha^{-1}, 100 kg KCl ha^{-1}), used with the conventional cultivation practices described above;
- T1: Inorganic NPK as in T0, used with SRI crop, soil and water management practices;
- T2: Organic fertilization: 5t ha^{-1} of organic compost, used with SRI management practices;
- T3: Inorganic NPK as in T0 + 300 kg ha^{-1} bioorganic fertilizer (FERTISMART), used with SRI management practices.

Soil samples were collected at the time of planting (0 WAT, week after transplanting), 8 WAT, and 16 WAT from a depth of 0–10 cm at the center of four hills. From within each plot, five sub-soil samples were collected and then mixed together to have a single plot soil sample, air-dried to a water content around 30%, and stored in a refrigerator until used for microbial population studies using a ten-dilution series.

Total numbers of microbes, and specifically the numbers of *Azospirillum*, *Azotobacter*, and *Aspergillus niger*, were determined by plate-counting methods using appropriate growing media: nutrient agar for total number of propagules; N-free medium for *Azotobacter* and *Azospirillum*, and Pikovskaya medium for phosphate-solubilizing microbes (Subba Rao 1982; Okon et al. 1977). Dried soil samples were used for parameters other than population counts, with these counts converted to number of microbes g^{-1} of dry soil, adjusted for samples' water content.

Results

TNAU studies of root and soil biological responses to SRI management

The thesis research by Nisha (2002) confirmed that plants grown with SRI methods had greater root length and root volume, as well as about 40% higher cation exchange capacity (CEC) and about 27% more ATPase activity andcytokinin content of roots (Table 1). CEC reflects the capacity of roots to absorb cations and thus vital nutrients. ATPase is a key enzyme required for the absorption of nutrients, and cytokinin is a growth hormone involved in cytogenesis, being synthesized in the root tips and translocated to other parts of the plant. SRI root systems were thus not only larger, but could function more effectively in support of rice plants. The data were consistent with the reports that SRI methods affect the size and performance of

Table 1 Root characteristics and activity in the rice crop under different crop management conditions, Coimbatore, India, wet season, 2001–2002

Parameter	Treatment	Crop growth stages			
		Transplanting	Active tillering	Panicle initiation	Flowering
Total root length (m)	Conventional	1.02	6.1	17.4	55.7
	SRI	0.88	22.5	31.1	67.5
Root volume (cc hill^{-1})	Conventional	1.48	10.7	25.5	42.5
	SRI	0.83	15.5	26.3	57.5
CEC of dried and milled roots (me 100 g^{-1} of dry root)	Conventional	NA	7.2	9.8	10.6
	SRI	NA	10.6	14.6	13.4
ATPase activity of fresh root (μg of inorganic P g^{-1} h^{-1})	Conventional	NA	0.24	0.53	0.62
	SRI	NA	0.34	0.69	0.74
Cytokinin content of roots (pmol g^{-1})	Conventional	NA	46.2	73.6	50.5
	SRI	NA	58.9	86.0	72.5

CEC cation exchange capacity. Conventional practice and SRI practice: as described in "Materials and methods" above. *Source*: Nisha (2002)

Table 2 Microbial populations in rhizosphere soil under different crop management conditions, Coimbatore, India, wet season, 2001–2002

Parameter	Treatment	Populations at different crop growth stages: square-root transformed values per gram of dry soil			
		Active tillering	Panicle initiation	Flowering	Maturity
Total bacteria	Conventional	9.35	14.91	9.73	7.64
	SRI	14.66	21.64	10.99	7.51
Azospirillum	Conventional	4.69	7.39	3.13	1.42
	SRI	7.17	9.08	4.23	1.52
Azotobacter	Conventional	8.88	25.57	10.45	5.56
	SRI	20.15	31.17	10.92	6.45
All diazotrophs	Conventional	9.11	10.52	7.14	4.71
	SRI	14.62	22.91	7.68	5.43
Phosphobacteria	Conventional	9.15	17.65	7.76	2.28
	SRI	16.19	23.75	13.79	2.66

Conventional practice and SRI practice: same as in Table 1. *Source*: Gyathry (2002)

roots, which would reciprocally have positive effects on the soil biota through root exudation.

Concurrent studies of Gyathry (2002) investigated such effects directly. Her assessments of the impact of SRI management practices documented how changes in cultural techniques could alter the microbial profile as well as the abundance of beneficial soil microorganisms. The SRI practices assessed included: younger seedlings, soil-aerating weeding with a mechanical weeder, water management to avoid continuous soil saturation, and green manures to enhance soil organic matter.

These practices, in combination, had positive effects on soil biota (Table 2). Gyathry found that the numbers of all aerobic bacteria in the SRI rhizosphere were increased by more than 50% before and during panicle initiation, compared to those in the rhizosphere of conventionally grown rice of same variety. The populations of *Azospirillum* also increased similarly, while *Azotobacter*, another diazotroph (N-fixing bacterium) and phosphate-solubilizing bacteria increased by even more, by about 75%.

During panicle initiation, the numbers of diazotrophs were more than twice as high under SRI management as with conventional practice. Throughout the crop cycle, not only were more bacteria found in SRI rhizospheres overall, but there were even more of those species that enhance plants' nutrient availability. The levels of enzymes that reflect the processes of N and P mobilization and uptake in the soil were also measured. This showed enzyme levels significantly greater at almost all phases of crop growth when SRI practice altered the management of plants, soil, water and nutrients (Table 3).

ICRISAT study of SRI practices' impact on rhizosphere biology

This evaluation over four seasons found that farmers' average yields using SRI cultivation methods were about 25% higher, 7.68 t ha^{-1} compared to 6.15 t ha^{-1} with farmers' usual practices (Rupela et al., 2006). ICRISAT researchers were interested to know whether there were

Table 3 Microbial activity in rhizosphere soil under different crop management conditions, Coimbatore, India, dry season, 2002

Parameter	Treatment	Biochemical activity levels at different crop growth stage: square-root transformed values per gram of dry soil.				
		Active tillering	Panicle initiation	Flowering	Grain filling	Maturity
Dehydrogenase activity (μg TPF g^{-1} soil 24 h^{-1})	Conventional	81	263	78	24	16
	SRI	369	467	139	95	42
Urease activity (μg NH_4-N g^{-1} soil 24 h^{-1})	Conventional	189	1,794	457	134	87
	SRI	230	2,840	618	228	173
Acid phosphate activity (μg *p*-nitrophenol g^{-1} soil h^{-1})	Conventional	1,800	2,123	957	384	214
	SRI	1,984	2,762	2,653	995	686
Alkaline phosphate activity (μg *p*-nitrophenol g^{-1} soil h^{-1})	Conventional	261	372	332	124	120
	SRI	234	397	324	189	146
Nitrogenase activity (nano moles C_2H_4 g^{-1} soil 24 h^{-1})	Conventional	–	3.15	7.63	–	1.94
	SRI	–	3.70	11.13	–	1.87

Conventional practice and SRI practice: same as in Table 1. *Source*: Gyathry (2002)

Table 4 Rice root dry weight (in g m^{-3}), root length density (m m^{-3}), and root volume (cm^3 m^{-3} soil) in top 30 cm soil profile at harvesting stage from 10 farmers' fields, Andhra Pradesh, India, rainy season, 2006

Depth	Root oven dry weight (g m^{-3})		Root length density (m m^{-3})		Root volume (cm^3 m^{-3} soil)	
	SRI	Conv.	SRI	Conv.	SRI	Conv.
0–15 cm	392	19	19,820	2,386	3,391	252
15–30 cm	193	19	10,572	2,243	1,740	242
SE±	34.7* (38.9)		1,816.2* (2,122.7)		292.5* (331.6)	
Mean	293	19	15,196	2,315	2,566	247
SE±	21.2**		1,022.6**		174.8**	
CV (%)	79		77		79	

*, ** and *** statistically significant at 0.05, 0.01, and 0.001 level of significance, respectively

Values in parentheses are SEs for comparing means within the same treatment

Source: Rupela et al. (2006)

any correlations of yield increase with soil biological populations and activity. Significant differences in the growth of root systems under SRI management were clearly confirmed in this study (Table 4). Rice plants in the SRI plots had about 10 times more root mass, about 5 times more root length density, and about 7 times more root volume in the top 30 cm of soil profile, compared with roots in the plots of flooded rice. Root length in the top 15 cm of soil on SRI plots was 19.8 vs. 2.4 km m^3 with usual practice (Rupela et al., 2006).

Differences in total microbial numbers and activity were not of this magnitude, however, as shown in Table 5. The *composition* of the soil biota apparently has more bearing on crop performance than do aggregated measures. Total numbers of bacteria and fungi in the soils of SRI and control plots were not found to be much different, although mean microbial biomass carbon (MBC) was 2–41% higher in three of the four seasons. The differences were not statistically significant because of their wide variability. The numbers of certain microbial species known to be beneficial for crop growth—phosphate solubilizers, and siderophore producers, which help plants acquire Fe—were higher in SRI plots, although again the differences were not statistically significant.

Three differences between the two sets of plots were significant at the 0.01 confidence level: numbers of nitrogen (N_2) fixing bacteria, microbial biomass nitrogen (MBN), and levels of dehydrogenase (Table 5). This latter enzyme, which oxidizes a substrate by transferring one or more hydrogen ions [H^+] to an acceptor, usually NAD^+/$NADP^+$ or a flavin coenzyme such as FAD or FMN, is considered to be an indicator of the general level of life in the soil. Total N and total P as well as available P were also higher in SRI plots, but the differences were not statistically significant.

Table 5 Properties of soil samples from SRI and control rice plots at fields of selected farmers in Andhra Pradesh, India, during four seasons (post-rainy 2004/2005 to rainy 2006)

Parameter	SRI	Control*	SE±	CV (%)
Bacteria ($\log_{10}$ g^{-1} dry soil)	6.15	6.18	0.044^{NS}	1.4
Fungi ($\log_{10}$ g^{-1} dry soil)	4.35	4.35	0.029^{NS}	1.3
Siderophore producers ($\log_{10}$ g^{-1} dry soil)	4.48	4.33	0.117^{NS}	5.3
Phosphate solubilizers ($\log_{10}$ g^{-1} dry soil)	3.40	3.28	0.154^{NS}	9.2
Pseudomonas fluorescens ($\log_{10}$ g^{-1} dry soil)	4.20	4.20	0.035^{NS}	1.7
N_2-fixers ($\log_{10}$ g^{-1} dry soil)	4.47	4.20	0.020**	0.9
Microbial biomass carbon (mg kg^{-1} soil)	1242	1187	58.1^{NS}	9.6
Microbial biomass nitrogen (mg kg^{-1} soil)	30	25	0.7**	4.9
Dehydrogenase (μg TPF g^{-1} 24 h^{-1})	114	93	3.0 **	5.7
Total N (mg kg^{-1} soil)	1082	1050	15.0^{NS}	2.8
Total P (mg kg^{-1} soil)	589	545	5.7^{NS}	2.0
Available P (mg kg^{-1} soil)	20.2	17.8	0.60^{NS}	6.3
Organic carbon (%)	1.06	1.06	0.002^{NS}	0.3

NS not significant

* Mean from plots where same farmers used their usual practices

** Significant at 0.01 level of significance

Source: Rupela et al. (2006)

A confounding factor possibly reducing correlations in Table 5 is that this analysis combined measurements taken in both the kharif and rabi seasons. During the rainy season, many farmers participating in the study were not able, or did not try, to control and limit their water applications as recommended for SRI cultivation. Thus, their soil conditions in the rainy seasons were less aerobic than in the post-rainy seasons.

Yields in general were higher during kharif seasons, 5–16% more than in rabi. However, SRI yields were always higher irrespective of season, by 18–22%. We note that the yield increases with SRI management in Andhra Pradesh were less than those reported from many other SRI evaluations. Comparative cross-national studies that address soil biological as well as climatic differences have not been done to assess SRI management and understand it better.

IPB research on rhizosphere microbial populations and activity

The research conducted at Bogor Agricultural University (IPB) in Indonesia assessed the numbers of all microbes in the rhizospheres of plants grown with SRI methods vs. those grown conventionally, being particularly interested in beneficial soil organisms. The comparisons shown in Table 6 indicate that the total population of bacteria in treatment plots was doubled with the combined effect of inorganic and organic fertilization using SRI methods, while organic fertilization with SRI methods produced a total population two-thirds higher than SRI practices having inorganic fertilizer applied. Specifically, organic fertilization with SRI practices contributed to almost four times more *Azospirillum*, and almost doubled the numbers of *Azotobacter* and phosphate-solubilizing microbes.

Comparison of results

Numbers of organisms as well as the chemical indicators of their activity will usually vary considerably from one set of trials to another, because of soil, climatic and other factors. Many more evaluations like these three should be done to gain a better understanding of the factors that affect bacterial population dynamics in conjunction with crop, soil, water, and nutrient management variables. However, it

Table 6 Total microbes and numbers of beneficial soil microbes (CFU g^{-1}) in soil under conventional and SRI rice cultivation at Tanjung Sari, Bogor district, Indonesia, Feb–Aug 2009

Treatments	Total microbes ($\times 10^5$)	*Azotobacter* ($\times 10^3$)	*Azospirillum* ($\times 10^3$)	PSM ($\times 10^4$)
Conventional (T0)	2.3c	1.9b	0.9c	3.3b
Inorganic SRI (T1)	2.7c	2.2b	1.7bc	4.0b
Organic SRI (T2)	3.8b	3.7a	2.8ab	5.9a
Inorganic SRI + BF (T3)	4.8a	4.4a	3.3a	6.4a

CFU colony forming units, *PSM* phosphate-solubilizing microbes, *BF* Bio-organic fertilizer (FERTISMART), described in the "Materials and methods" section

Values with different letters in a column are significantly different by LSD at the 0.05 level

Source: Iswandi et al. (2009)

Table 7 Summary comparison of increases in number and activity of beneficial soil organisms in the rhizospheres of SRI rice plants compared to conventionally grown plants, from Indian and Indonesian evaluations

Increases in	TNAU study: Gyathry (2002)	ICRISAT study:[a] Rupela et al. (2006)	IPB study: Iswandi et al. (2010)
Total bacteria	312%	ND	65%
Total diazotrophs[b]	61%	6.4%**	NM
Azospirillum[b]	32%	NM	211%
Azotobacter[b]	36%	NM	94%
P-solubilizing microbes	53%	3.6%[ns]	78%
Dehydrogenase (μg TPF g^{-1} 24 h^{-1})	140%	22.5%**	125%
Microbial biomass N (mg kg^{-1} soil)	NM	20%**	NM

ND no difference, *NM* not measured

[a] These trials included wet-season results when water control was incomplete and therefore aerobic soil conditions were difficult to maintain

[b] N_2-fixing bacteria

** Significant at 0.05 level of confidence

appears that SRI practices contribute to positive crop results in part by creating conditions in which more beneficial soil microbes prosper. To the extent that these organisms thrive, so do the plants associated with them through plant root-microbial interactions and collaboration.

The findings of these different studies are summarized in Table 7, focusing on organisms that are known to perform beneficial functions for crops such as engaging in nitrogen fixation (*Azospirillum* and *Azotobacter*) and phosphate solubilization (*Aspergillus niger*). These processes provide plants with essential nutrients through microbial activity in the soil. The enzyme dehydrogenase is regarded as an indicator of total life in the soil, as is the level of microbial biomass nitrogen.

The capacity of SRI practices to affect soil bacterial populations, sometimes quite significantly, is evident from these data. The enzymatic 'footprint' of dehydrogenase is a strong indicator of biological activity within the rhizosphere, consistent with population estimates. We note that these evaluations did not assess changes in populations of mycorrhizal fungi, symbionts known to make important contributions to crop health and productivity by improving plant roots' access to both nutrients and water (Schreiner et al. 1997).

Discussion

Why certain beneficial soil organisms should be more numerous and more active in and around the roots of rice plants grown with SRI management practices remains to be studied more thoroughly before generalizable and actionable conclusions can be drawn. Maintaining paddy soils under mostly aerobic conditions—in contrast to the anaerobic conditions that prevail when rice is grown in conventionally flooded paddies—could explain a great deal of the differences observed. This effect could be augmented by the enhancement of soil organic matter with SRI management and when active soil aeration occurs through recommended SRI weeding practices. Rotating hoes and conoweeders put more organic matter (weeds) into the soil for decomposition and nutrient recycling at the same time that they churn up the surface soil.

Review of SRI practices and possible impacts on rhizosphere populations and activity

The changes in practice that are recommended according to the principles of SRI are just a few, and quite simple. Systematic research on why these changes are conducive to greater root growth and to changes in the soil biota is still limited. But from the SRI literature and from observations, some lines of explanation can be suggested for why these practices could be having an impact on rhizosphere populations as suggested by the studies reported above.

- Young seedlings: SRI starts with the transplanting of seedlings at the 2–3 leaf stage, and not beyond the start of their 4th phyllochron of growth (Nemoto et al. 1995). When older seedlings are transplanted, they have less potential for the production of tillers and roots. Also, young seedlings have less root system that can be traumatized during their removal from the nursery and transplantation into the field. Transplanting young seedlings and handling them carefully will reduce what is called 'transplant shock.' This trauma interrupts the growth of conventionally transplanted rice for 7–14 days, at a critical time for plants'

development. SRI transplants resume their growth quickly and readily. Plants that develop larger root systems as well as bigger canopies contribute more root exudates (carbohydrates, amino acids, etc.) to the rhizosphere, where they substrate for soil organisms.

- Upland nursery: SRI seedlings are grown in nurseries that are not kept flooded, as in conventional practice. Also, the seed rate is reduced to avoid crowding among the emerging plants. These changes have been shown to enhance rice plants' growth potential, and their root growth in particular (Mishra and Salokhe 2010). However, studies have not addressed the effect that SRI changes in nursery management could have on associated soil biota.
- Reduced plant population: SRI rice plants are transplanted singly and with wide spacing, in a square pattern, radically reducing plant populations. This practice, which differs from usual practice which assumes more plants will give more yield, gives the plants' root systems more room to grow. It has been seen that xylem exudation which benefits the shoot is increased with SRI management (Thakur et al. 2010, 2011). However, root exudation into the soil, which would nourish soil microorganisms, has not been studied very much. The first research on this (Tao et al. 2002) found SRI management inducing greater root growth and higher yield, but with some reduction in exudates. With larger root systems that do not senesce until late in the crop cycle when the soil is maintained aerobic (Kar et al. 1974), one can expect that more substrates will be exuded into the rhizosphere to support soil bacteria and fungi.
- Aerobic soil conditions: Under SRI management, paddy fields are not kept continuously flooded, which differs from common practice wherever there is sufficient water supply. SRI soil conditions are kept mostly aerobic throughout the vegetative growth period, and soils have less depth of flooding after panicle initiation. Soil that is not hypoxic is more conducive to healthy root growth and at the same time more supportive of diverse populations of beneficial (aerobic) soil biota.
- Active soil aeration: Weed control is more necessary when paddy fields are not kept inundated. With SRI, a simple mechanical hand weeder is used to eliminate weed growth. This implement, in addition to controlling weeds, has the effect of active soil aeration. This can be expected to promote both root growth and the biodiversity and abundance of soil organisms that can enhance plant health and performance.
- Enhanced soil organic matter: Although SRI methods can enhance crop yield when used together with chemical fertilizer, the best yields and greatest cost-saving for farmers have usually been attained with the application of compost or other organic fertilizers, as much as possible. This practice improves soil structure and fertility and is conducive to greater root growth and better functioning, as well as abundance and diversity of soil organisms.

Possible negative effects of SRI management

The effects of SRI management can be negative under some conditions, e.g., where there are inherent populations of root-feeding nematodes, which will be enhanced by more aerobic soil conditions. Research in Thailand documented significantly higher nitrification rates in the soil with SRI management, and also more diverse communities of ammonia-oxidizing bacteria (Sooksa-Nguan et al. 2009). However, SRI yields were not higher than on the control plots, and SRI plant roots were seen to have greater prevalence of nematode galls. Possibly some modification in water management could both limit nematode damage and yet support more extensive and active populations of aerobic organisms to derive their benefits. But such optimizing management has not been attempted.

Variability in soil biology effects of management practices

While increases in the numbers and activity of beneficial soil organisms were seen in the three studies, where SRI methods enhanced the yields obtained, there was also great variation in the results of our measurements. This could be due to intra-seasonal or intra-field variability. Variability in results is not necessarily a sign of measurement error as there is no single value that can be derived from sampling and subsequent analysis. Variability should be assigned as much as possible to factors like intra-field soil variations or inter-season effects like soil moisture versus aeration.

Most current concepts and methods for statistical evaluation implicitly assume that there is some single 'true' value that should be revealed through measurement. However, in soil biological analyses, many of the numbers to be determined remain highly variable, contingent not just on one or a few factors but on many. Further, these factors themselves interact, and not always in a linear, proportional manner. Measurements of soil biology will thus tend to be less stable and replicable than other measurements.

Need for further evaluations

Laulanié's recommendations (1993) for SRI practice were empirically derived, based on observations and experimentation without benefit of formal agronomic research.

Numerous studies in the peer-reviewed literature, referred to in the Introduction, have documented the merits of more aerobic soil–water management and of reduced plant populations. The beneficial effects of these alternative practices can be heightened, often dramatically, when they are combined with the transplanting of very young seedlings and increased applications of organic matter that improve soil structure and function (Chapagain and Yamaji 2009; Mishra and Salokhe 2008, 2010; Thakur et al. 2010, 2011; Zhao et al. 2009, 2010; Yang et al. 2004).

The insights and recommendations of this priest-agronomist have encouraged farmers, researchers, and policymakers to reconsider what had been thought to be settled agronomic questions. Producing more rice with less water, fewer seeds, and reductions in purchased inputs contrasts markedly with presently recommended 'best management practices.' Current understanding of how to produce rice crops with higher factor productivity, and in ways that are environmentally friendly and socially more beneficial, is undergoing revision. SRI methodology is assigning to plant roots and to the associated soil biota a focus for research and practice that they have long deserved.

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