Rice-Wheat Consortium Paper Series 16

Soil Health and Sustainability of the Rice-Wheat Systems of the Indo-Gangetic Plains

Proceedings of the End of Project Workshop

7 – 10 May 2002
Kathmandu, Nepal

Rice-Wheat Consortium for the Indo-Gangetic Plains
CIMMYT and CABI Bioscience

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CIMMYT is an internationally funded, not-for-profit organization that conducts research and training related to maize and wheat throughout the developing world. Drawing on strong science and effective partnerships, CIMMYT works to create, share, and use knowledge and technology to increase food security, improve the productivity and profitability of farming systems, and sustain natural resources.

Financial support for CIMMYT's work comes from many sources, including the members of the Consultative Group on International Agricultural research (CGIAR), national governments, foundations, development banks, and other public and private agencies.

CABI Bioscience is a division of CAB International – a not for profit enterprise contributing to global development, the environment and human welfare through knowledge generation and delivery in the applied life sciences. CABI Bioscience integrates four former international biological institutes, the International Institute of Biological Control (**IIBC**), the International Institute of Entomology (**IIE**), the International Institute of Parasitology (**IIP**) and the International Mycological Institute (**IMI**). CABI Bioscience has mutually productive partnerships with many global organisations, agencies, foundations and corporations.

Rice-Wheat Consortium for the Indo-Gangetic Plains is an Ecoregional program of the Consultative Group on International Agricultural Research (CGIAR), managed by CIMMYT, involving the National Agricultural Research Systems, the International Agricultural Research Centers, and the Advanced research Institutions. Its main objective is to promote research on issues that are fundamental to enhance the productivity and sustainability of rice-wheat cropping systems in South Asia.

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Disclaimer: The names of the micro-organisms in the eight following papers have generally been kept as indicated by the first author of the paper and may not reflect the current accepted nomenclature following international standard rules in taxonomy. It does not constitute an endorsement by CIMMYT and CABI of the nomenclature used in these proceedings.

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Editors:
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Abbreviations

CABI	CAB International
CCS HAU	Chaudhary Charan Singh Haryana Agricultural University, India
CDRI	Crop Diseases Research Institute, Pakistan
CGIAR	Consultative Group for International Agricultural Research
CIMMYT	International Maize and Wheat Improvement Centre
CSD	Chinese Seed Drill
CT	Conventional tillage
DFID	UK Department for International Development
IAAS	Institute of Agriculture and Animal Science, Nepal
IARI	Indian Agricultural Research Institute
MEA	Malt Extract Agar
MT	Minimum tillage
NARC	Nepal Agricultural Research Council
NARS	National Agricultural Research Systems
PDA	Potato Dextrose Agar medium
PPN	Plant Parasitic Nematodes
RCT	Resource Conserving Technologies
RLI	Root Lesion Indexing
RKI	Root Knot Indexing
RWC	Rice-Wheat Consortium for the Indo-Gangetic Plains
ZT	Zero Tillage

Introduction to the Soil Health Project

Resource conserving technologies in rice-wheat systems

The consecutive cropping of wheat after rice is one of the world's principal agricultural production systems. This system is practised across an area of about 13 million hectares in South Asia and provides food, income and employment to over 200 million rural and urban producer and consumers. About 50 million farm families rely on this agricultural system for their subsistence. Typically in the region wheat is grown from November to April followed by rice during the monsoon from June-July to October-November.

However, concerns have emerged regarding the sustainability of the rice-wheat systems. They are showing signs of stress and their sustainability is at risk, especially where there is a continuous rice-wheat rotation and system diversity is low. Problems include low water and nutrient use efficiency, groundwater depletion, water logging, poor water control, salinity and the build-up of weeds, pests and diseases. Yield stagnation and in some cases decline are reducing rural incomes and threatening food security in many areas.

Many agricultural research institutions have worked to address these serious concerns. Much of this work has focused on developing resource conserving technologies (RCT) helping to produce cereals at a lower cost while attempting to improve soil health through reduced tillage and stubble retention. Pioneering work by the Govind Ballabh Pant University of Agriculture and Technology (Pantnagar), Haryana Agricultural University, the Rice-Wheat Consortium for the Indo-Gangetic Plains (RWC) and other research organisations have identified new tillage and crop establishment options for rice and wheat that address system ecology problems. These technologies mostly involve reducing tillage and are being widely and rapidly adopted by farmers across the Indo-Gangetic Plains in what has been described as a new green revolution.

The Rice-Wheat Consortium has worked in close collaboration with South Asian National Agricultural Research Systems (NARS). These joint initiatives and participatory research involving scientists and farmers through several special projects have accelerated the adoption of the new and improved tillage methods. They include DFID CRF project (R6406) "Crop Establishment and Reduced Tillage Activities in the Rice-Wheat Systems in the Indo-Gangetic Plains" and DFID CRF funded project (R7259) "Harnessing Tillage by Nutrient Management Interactions using Participatory Approaches to Improve Rice-Wheat Systems Productivity and Sustainability".

Several kinds of these resource conserving technologies are being adopted according farmers' needs and situations. In zero tillage, wheat seeds are drilled into unploughed fields which retain the residues from the rice crop. In reduced tillage, the seeds are surface sown onto rota-tilled soil using a two wheel power tiller or Chinese seed drill (CSD) a technique that is particularly suitable in smallholdings. The surface seeding of wheat into standing rice or after rice harvest has been long used by farmers in parts of South Asia where soil moisture is too high and hampers conventional tillage. These practices are overall referred to as resource conservation technologies (RCT) and form a basket of options to farmers.

These practises offer a number of benefits. Farmers save on tillage costs and can sow their wheat, on average, two weeks earlier than under conventional tillage to give a longer growing season and thus more yield. The savings on production costs and increased yields are making an important contribution to livelihood improvement amongst small farmers. The technologies are also more sustainable as they use less energy and are more environmentally friendly.

However, concern is being felt that the reduction in tillage and changes in soil physical conditions or crop residue management may be lead to a build up of soil borne pathogens or pests in the rice stubble. Tillage is traditionally seen as an important way of keeping crop pathogens in check. This could become particularly harmful to the many small and resource poor farmers who have adopted the new technology.

The soil health project

Recent research in South Asia, including work by Cornell University using soil solarization trials, suggests that reduced soil fertility alone does not sufficiently explain reductions in yield productivity. Soil borne pathogens, including fungi and nematodes, could also be an important cause of the reduced yields in the Indo-Gangetic continuous rice-wheat systems. Bimb and Dubin (1994) identified a serious lack of knowledge about soil borne fungi in rice- wheat production systems. In particular little is known about the effects of zero tillage on soil health and its long term effects on soil biota.

To begin to fill these gaps in knowledge, the International Maize and Wheat Improvement Centre (CIMMYT), along with CABI Bioscience (UK) and NARS carried out a series of studies under the project ‘Soil Health and Sustainability of the Rice-Wheat Systems of the Indo-Gangetic Plains’. This project was financially supported by the UK Department for International Development [DFID Project (R7263)]. The project was carried out on behalf of the Rice-Wheat Consortium (RWC) of the Consultative Group on International Agricultural Research’s (CGIAR) based in New Delhi, India. It ran from April 1999 to March 2002 in India, Pakistan, Nepal and Bangladesh.

The main aim of the study was to assess whether the adoption of zero and minimal tillage for wheat following rice in the Indo-Gangetic plains had led to a build up or increase in plant pathogenic organisms compared to conventional tillage systems. An important part of the study was to increase overall knowledge about soil health, including mapping soil microbiota profiles under resource conservation tillage in South Asia.

The project’s research was carried out in an integrated regional network. The field work and much of the laboratory work was carried out by scientists from the national agricultural research programs, with backstopping from CIMMYT and CABI. Studies were carried out by the G.B. Pant University of Agriculture and Technology Pantnagar, India; Haryana Agriculture University, India; the Crop Diseases Research Institute, Islamabad, Pakistan; and the Nepal Agricultural Research Council in association with the Institute of Agriculture and Animal Science, Nepal.

Research plans were developed at planning workshops in New Delhi and Islamabad in 1999. The NARS, CABI, and CIMMYT staff selected the study sites and farmers’ fields and ensured that rice-wheat cropping and zero tillage were maintained on an area of at least one acre at each site for the project duration. The farmers at Mona and Muridke sites in Pakistan, at Pantnagar in

Uttaranchal and at Kaul in Haryana had previously been involved in similar research activities. These sites were selected to build on previous tillage research conducted in collaboration with CIMMYT (DFID project R6406) and to link into on-going work by the RWC.

The farmers agreed to adopt zero tillage on part of their land while continuing to use conventional tillage on adjacent fields. A few fields that had a break crop before the project were also selected. To increase synergies and research efficiency it was decided to study the same fields in this project as studied by DFID CRF Project (R7259) (C) "Harnessing Tillage by Nutrient Management Interactions using Participatory Approaches to Improve Rice-wheat Systems Productivity and Sustainability". This was done to obtain more detailed information on soils and on agronomic and biological soil constraints under different tillage practices. It was only possible to monitor a limited number of the fields at each site as collecting soil health data and identifying microbiota is very time consuming.

The project also linked with rice-wheat research undertaken by the World Bank funded National Agricultural Technology Project and its long-term trials such as those at Pantnagar. The project worked with four farmers in Pantnagar and four in Kaul. In Pakistan, four farmers participated in the research at Mona and Muridke. In Bangladesh and Nepal the project worked with NARS plant pathology staff alongside their existing commitments to Cornell University research activities. In addition, from the 2000 monsoon season, six farmers fields in the three communities of Santapur, Belwa and Benauli in the Nepal Tarai were studied, covering a complete rice-wheat sequence (2001-2002).

The project developed standard research protocols for field and laboratory-based disease assessments, soil and plant sampling and isolation of fungi and nematodes (Bridge *et al*., 1999). These were strictly followed during the research across most sites to allow results to be compared. However, at Kaul the protocols could not be followed and these farmers were studied using the Kaul station's standard criteria for rice and wheat research.

The project provided training manuals and reference materials to help the scientists carrying out the studies to identify organisms and understand soil health concepts. CABI scientists provided training on fungal and nematode isolation methods and identification during the 2000 wheat season in Islamabad, Pakistan and during the 2000 rice season in Pantnagar, India. Pakistani, Nepali and Bangladeshi scientists participated in a March-April 2000 training workshop in Pakistan while senior scientists from Pakistan and Nepal were given specialized training on fungal identification at CABI Bioscience (UK). The Indian Agricultural Research Institute assisted by training an Indian nematologist.

CABI fungal taxonomists Paul Kirk and Graham Kinsey gave several in-country courses and David Brayford helped to identify *Fusarium* species. The CIMMYT scientist was mainly responsible for managing the project, coordinating contact between project stakeholders, and preparing technical reports. The final compilation of the data was done at CABI UK including the final identification of microorganisms. Excellent coordination and communication was maintained between CIMMYT and CABI throughout the project.

In all selected farmers' fields, the studies (1) determined fungal and nematode profiles in the soil, (2) assessed root health of the two crops, and (3) evaluated the main disease problems in the crop canopy that may have resulted from soil biotic constraints. The technical information was collected for each rice and wheat crop by first discussing the state of the crop with the farmer. A rapid visual assessment was carried out of the disease situation in each field, the cereal variety and other factors influencing crop status and soil health. Ten soil and whole plant samples were then collected according to a defined pattern to adequately represent the entire field. Leaf diseases were recorded on ten randomly selected tillers using a 0-5 disease severity assessment scale and root health was evaluated using a 0-3 scale score for root necrosis. The root samples were evaluated for galls indicative of root knot nematodes.

Composite soil samples were used to extract and assess the different nematode species and trophic groups using an illustrated identification key prepared for the project (Hunt 2000). Root colonization and infection by pathogenic nematodes was recorded. Root samples showing necrosis or discoloration were assayed to isolate fungal pathogens. Dilution plating method with two replicates and five specialized semi-selective culture media were used to isolate and quantify specific fungal groups. Total fungal colony counts were also recorded to calculate the colony forming units per gram of soil. Fungi emerging on these culture media were sub-cultured and identified where possible to species level to obtain fungal profiles. Some identifications were carried out in the UK due to a lack of black light facilities to induce sporulation and due to difficulties experienced by NARS in identifying some species. The accurate identification of all pathogenic, and particularly non-pathogenic, nematodes and fungi proved difficult without the support of specialists from CABI or CIMMYT.

Overall project findings

The project findings strongly suggest that the adoption of RCT is not adversely affecting soil biology. Nematodes, which have been thought to effect grain yield production, were not found in significant numbers possibly as a result of the regular flooding that takes place in the rice crops. Numerous fungal microorganisms, including several indicators of biocontrol activity, were observed in soil profiles. In Nepal, a clearer understanding of the taxonomy of the *Fusarium* group was gained, including confirmation of the occurrence of *F. nygamai.* The findings over the short period of this study suggest that there is little risk of inoculum build-up of rice or wheat pathogens or significant carry-over between the two crops leading to new biotic constraints. These results endorse the further promotion of zero tillage and other resource conserving technologies. Two other important outcomes of the project were (i) to assist the Rice-Wheat Consortium to build its links with CABI and other international partners and (ii) the collection of a large amount of information on soil microbiota under resource conservation tillage in South Asia.

The eight papers in this publication are the proceedings of the end of project workshop held jointly with the CIMMYT tillage project group in Kathmandu on 9 May 2002. This workshop facilitated cross-site comparisons and exchanges between scientists from different disciplines and regions. It confirmed that a multi-disciplinary approach was needed to address the problems of the South Asian rice-wheat cropping systems.

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Effect of Tillage Practice on Rhizosphere Fungi and Diseases of Rice and Wheat in Haryana State, North India

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ABSTRACT

Soil health studies were carried out at CCS HAU Rice Research Station, Kaul and in farmers' fields in Teek (Kaithal) and Uchana (Karnal) to compare zero and conventional tillage practices in rice-wheat sequence cropping in Haryana, north India. Considerable differences were observed in the population dynamics of rhizosphere fungi. Counts of soil fungi were higher in conventional than zero tillage fields at crown root initiation (CRI) and dough stage of the wheat crop while no uniform trend was observed for paddy. The predominant fungal species found in the rhizosphere of wheat and rice were *Fusarium* species, *Drechslera rostrata*, and *Penicillium* species. The population of *Fusarium pallidoroseum* was more in conventional plots. Amongst the fungal genera identified in the rice and wheat rhizospheres, *F. moniliforme*, *F. pallidoroseum*, *D. oryzae* and *D. rostrata* are well known to be pathogenic on paddy, and *Alternaria triticina* and *Bipolaris sorokiniana* are pathogenic on wheat. The study found little difference between the two cropping practices in the incidence and severity of disease. The only noticeable differences were the higher incidence of foot rot, bakanae, and grain discolouration in zero tillage and stem rot in conventionally sown fields.

INTRODUCTION

Interest in no-tillage and conservation tillage systems is increasing due to scarcity and increasing cost of fossil fuels, periodic food shortages, inclement weather conditions, and concerns over soil erosion. Changing the tillage practice can lead to changes in the physical and chemical properties of soil which in turn is likely to influence the occurrence of plant diseases. Key factors in the occurrence of plant diseases include the survival and activity levels of pathogens, host susceptibility, and the population of other soil microorganisms. Reduced tillage can favour pathogens by lowering soil temperatures, increasing soil moisture, changing root growth, changing nutrient uptake, and changing the population of plant pathogen vectors. The decomposition of rice plant residues may release phytotoxins and stimulate toxin producing microorganisms, thereby predisposing plants more to pathogen attack (Sturz *et al.,* 1997). However, relatively high soil microbial activity can lead to competition effects that may affect pathogen activity and survival and thus reduce harmful pathogen inoculum pressures. This microbial antagonism in the root zone can be beneficial for farmers by leading to the formation of disease suppressive soils. Thus, on the one hand leaving plant debris on the surface or partially buried may allow pathogens to survive to the next crop, but it may also make conditions more favourable for the biological control of plant pathogens (Sumner *et al.,* 1981).

Reduced tillage has long been used by rice farmers in Arkansas, USA, for economic and soil conservation reasons. Since the mid-1990s a similar practice, the zero tillage sowing of wheat after rice, has become popular particularly with wheat farmers in Haryana, northwest India. The growing importance of this technique has led to the need for research into its characteristics. DFID and CIMMYT therefore supported research into the soil health of rice-wheat systems. The aim of this November 1999 to March 2002 project was to study the long term impact of zero tillage on the population dynamics of soil fungi and the incidence and severity of diseases in rice-wheat sequences.

The study selected two sites from Karnal district and one from Kaithal district, Haryana (Table 1). The farmers of these sites had been practising both zero and conventional tillage for sowing wheat after rice for the previous four or five years.

The study collected soil samples from these farms at the crown root initiation (CRI) and dough stages of wheat, and the tillering and flowering stages of paddy. The population of soil fungi was recorded on potato dextrose agar amended with 300 ppm strepto-penicillin for general isolation. It was collected on modified Czapek (Dox) agar for selective isolation of *Fusarium moniliforme* using the dilution plate method.

Table 1. Details of soil health study sites in Kaithal and Karnal, Haryana, India

Description of farmers	Location	Tillage practice	Wheat		Paddy	
			Sowing time	Sampling time	Planting time	Sampling time
Nakli Ram S/o Singh Ram	Teek (Kaithal)	Zero	04-Nov-1999 11-Nov-2000 29-Oct-2001	27-Nov-1999 30-Mar-2000 03-Dec-2000 25-Mar-2001 17-Dec-2001	01-Jul-2000 21-Jun-2001	04-Sep-2000 15-Sep-2001
		Conventional	07-Nov-1999 11-Nov-2000 05-Nov-2001	27-Nov-1999 30-Mar-2000 03-Dec-2000 25-Mar-2001 17-Dec-2001	24-Jun-2000 21-Jun-2001	04-Sep-2000 15-Sep-2001
Naresh S/o Malkhan Singh	Uchana (Karnal)	Zero	04-Nov-1999 17-Nov-2000 05-Nov-2001	28-Nov-1999 03-Apr-2000 04-Dec-2000 22-Mar-2001 20-Nov-2001	10-May-2000 22-Jun-2001	16-Jun-2000 06-Sep-2000 02-Jul-2001 22-Sep-2001
		Conventional	04-Nov-1999 15-Nov-2000 06-Nov-2001	28-Nov-1999 03-Apr-2000 04-Dec-2000 22-Mar-2001 20-Nov-2001	10-May-2000 22-Jun-2001	16-Jun-2000 06-Sep-2000 02-Jul-2001 22-Sep-2001
Dharamvir S/o Dal Singh	Uchana (Karnal)	Zero	04-Nov-1999 03-Nov-2000 04-Nov-2001	28-Nov-1999 03-Apr-2000 04-Dec-2000 22-Mar-2001 20-Nov-2001	30-Jun-2000 15-Jun-2001	16-Jun-2000 06-Sep-2000 02-Jul-2001 22-Sep-2001
		Conventional	04-Nov-1999 06-Nov-2000 03-Nov-2001	28-Nov-1999 03-Apr-2000 04-Dec-2000 22-Mar-2001 20-Nov-2001	30-Jun-2000 15-Jun-2001	16-Jun-2000 06-Sep-2000 02-Jul-2001 22-Sep-2001

The disease data was recorded according to percentage leaf area affected for foliar blight of wheat, and the IRRI evaluation system for false smut, sheath rot, brown spot, grain discolouration, foot rot, bakanae, neck blast, and narrow brown leaf spot of paddy (Anonymous, 1988).

RESULTS AND DISCUSSION

In both tillage practices, from amongst the three media (potato dextrose agar supplemented with 300 ppm strepto-penicillin [PDA+S], malt extract agar [MEA], and Martin rose bengal agar), most soil fungi were recorded on the PDA+S media. Analysis of the soil samples showed that the population of soil fungi varied significantly with growth stage, tillage practice, crop season, and between farmers' fields. The study found that the fungal counts were almost twice as much in the conventionally sown ploughed fields compared to the zero tillage fields at both the CRI and dough stages of the wheat (Table 2). In paddy the fungal population was found to be more in conventional tillage at the tillering stage while it was less under conventional tillage site at flowering.

In the rhizosphere of wheat sown by both tillage methods, the predominant fungi were *Fusarium* species (*F. moniliforme*, *F. pallidoroseum* and *F. oxysporum*), *Drechslera* (*D. rostrata*, *D. oryzae*) and *Penicillium* species. The next most common fungi were *Alternaria triticina*, *Bipolaris sorokiniana*, *Cladosporium cladosporioides* (a common black and sooty mold), *Curvularia tetramera* and *Mucor* species.

Table 2. Counts of soil fungi in wheat during 1999-2000, 2000-2001, and 2001-2002 and statistical analysis; Medium used: PDA amended with 300 ppm strepto-penicillin

Farmer	Sampling	Number of fungal counts (x 10^3)							
		Zero				Conventional			
		99-00	00-01	01-02	Mean	99-00	00-01	01-02	Mean
Nakli Ram	November	2.7	22.3	6.0	10.3	18.8	98.3	6.3	41.1
	March	5.1	2.7	3.3	3.7	5.4	1.3	3.0	2.9
Naresh	November	15.2	13.7	7.3	12.1	6.0	14.3	3.7	8.0
	March	3.0	2.3	2.7	2.7	0.8	5.0	5.0	3.6
Dharamvir	November	3.8	18.0	5.0	8.9	14.0	8.3	7.7	10.0
	March	3.0	3.3	3.7	3.3	6.3	3.3	4.0	4.5
Mean	November	7.2	18.0	6.1	10.4	12.9	40.3	5.9	19.7
	March	3.7	2.8	3.2	3.2	4.2	3.2	4.0	3.8

	1999-2000	2000-01	2001-02
CD 5% for Tillage Practice:	0.8	4.2	N.S.
Farmers:	1.0	5.2	N.S.
Crop Stage:	0.8	4.2	0.5
TxF:	1.4	7.3	0.9
TxCS:	1.1	6.0	N.S.
F x CS:	1.4	7.3	N.S.
TxFxCS:	1.9	10.4	1.4

The main fungi isolated from the paddy rhizosphere were also *Fusarium* species, followed by *Penicillium* species, *Mucor* species, *Curvularia tetramera*, and *Drechslera rostrata.* The other fungi recorded in wheat were *Rhizopus oryzae*, *Aspergillus flavus*, *A. nidulans* var. *dentatus*, and a few sterile species. Among these, *Fusarium moniliforme* has been reported to cause foot rot, bakanae, withering of shoots, and sheath rot in paddy; and head scab of wheat. *F. moniliforme*, *Drechslera rostrata*, *Aspergillus flavus* and *Penicillium* species have been associated with wheat grains in the field and in storage. *Drechslera oryzae* and *Alternaria triticina* and *Bipolaris sorokiniana* are well known pathogens that cause brown spot of paddy and foliar blight of wheat.

This study found the pathogens of rice *F. moniliforme*, *F. pallidoroseum*, *D. oryzae* and *D. rostrata*, and the wheat pathogens *Alternaria triticina* and *Bipolaris sorokiniana* (Table 3). *F. moniliforme* caused bakanae of paddy and doubled the usual seedling height from 75 mm to 150 mm, besides causing seedling mortality. *F. pallidoroseum* caused only seedling mortality (Table 4). The population of *F. moniliforme* was found to be significantly higher in conventionally tilled ($1.6x10^2$ fungal colonies) than in zero tilled plots ($0.6x10^2$ fungal colonies) in the wheat rhizosphere. The population was more in zero drill sown wheat fields at tillering than in the following paddy (Table 5). However, this fungus could not be recovered at flowering stage in either type of practice using Sharma and Singh's selective medium (1973). The incidence of foot rot and bakanae in paddy cultivar Taraori Basmati was more in rice fields that followed zero till wheat (9.6%) than in the conventionally tilled plots (3.2%). Typical withering of the plants due to foot rot and bakanae was also evident in zero till fields at the time of maturity. In a similar study by Cartwright *et al.* (1996) in Arkansas, USA a significantly higher level of *Rhizoctonia solani* was observed in no-till fields than in conventional fields but there was no significant difference in incidence and severity of paddy sheath blight.

Fusarium pallidoroseum has been reported as a potential bioherbicide for managing *Parthenium* weed species (Jeyalakshmi *et al.*, 2001). The population of this fungus was higher in soil from the conventionally sown fields with both crops (Table 6).

Table 3. Fungi found pathogenic on wheat and paddy in Haryana, India

Fungus	Pathogenic on	Symptoms
Fusarium moniliforme	Paddy	Bakanae (elongated seedlings)
Fusarium pallidoroseum	Paddy	Seedling mortality
Cochliobolus miyabeanus	Paddy	Brown spot
Drechslera rostrata	Paddy	Small, oval spots with grey centre and darker border on lower most leaf sheath
Alternaria triticina	Wheat	Foliar blight
Bipolaris sorokiniana	Wheat	Foliar blight

Table 4. Pathogenicity of *Fusarium* spp. in test tubes on paddy cultivar Taraori Basmati

Treatment	Elongated seedling* (%)	Seedling mortality (%)	Seedling height (mm)
F. moniliforme	100.0	47.5	149.7
F. pallidoroseum	0.0	32.5	71.8
Uninoculated	0.0	0.0	76.5

Mean of 4 replicates each comprising 10 seeds per test tube;Inoculum dose: 5 drops of mycelial suspension having 2 x 10^7 conidia/ml per test tube

Table 5. Counts of *Fusarium moniliforme* in wheat and paddy rhizosphere during 1999-2000

Farmer	No. of fungal colonies (x10^2)							
	Wheat				Paddy			
	Zero		Conventional		Zero		Conventional	
	Nov-99	Mar-00	Nov-99	Mar-00	Jul-00	Sep-00	Jul-00	Sep-00
Nakli Ram	0.5	0.6	4.7	2.3	0.0	0.0	0.3	0.0
Naresh	0.2	1.0	0.5	1.3	0.6	0.0	0.3	0.0
Dharamvir	1.1	0.0	0.9	0.0	0.6	0.0	0.0	0.0
Mean	0.6	0.5	1.6	1.8	0.4	0.0	0.2	0.0

Table 6. Counts of *Fusarium pallidoroseum* in wheat and paddy rhizosphere during 1999 - 2000

Farmer	No. of fungal colonies (x10^2)							
	Wheat				Paddy			
	Zero		Conventional		Zero		Conventional	
	Nov-99	Mar-00	Nov-99	Mar-00	Jul-00	Sep-00	Jul-00	Sep-00
Nakli Ram	1.4	3.7	0.4	0.7	0.3	0.0	8.3	0.0
Naresh	1.4	7.0	1.0	7.7	3.7	0.0	20.0	0.0
Dharamvir	1.3	3.0	5.0	6.3	2.7	0.0	8.3	0.0
Mean	1.4	4.6	2.1	4.9	2.2	0.0	12.2	0.0

High incidences and severity of a number of foliar and root pathogens has been reported from conservation tillage fields. These include *Septoria tritici* causing speckled leaf and septoria blotch, *Septoria nodorum* causing leaf and glume blotch, *Rhizoctonia solani* causing *Rhizoctonia* bare patch and root rot, *Pythium* species causing seed and root rot, *Erysiphe graminis* causing powdery mildew, and *Fusarium* species causing crown rot of wheat. In contrast the fungi *Bipolaris sorokiniana* (foliar blight), *F. culmorum,* and *F. avenaceum,* which are causal agents of common root rot in wheat, are partially or completely controlled by reduced tillage.

Other studies on no till regimes have shown reduced pathogen activity. Rothrock (1992) reported that the inoculum of *Gaeumannomyces graminis* var. *tritici* was found to spread only 10 cm in undisturbed soil compared to 2.5m in cultivated soil. Wiese and Ravenscroft (1975 found that ploughing under wheat residue reduced the sporulation of *Cephalosporium gramineum* in the top 7.6 cm of soil. However, the current study found no significant difference in the incidence and severity of foliar blight of wheat, false smut, sheath rot, brown spot, neck blast and narrow brown leaf spot of paddy between the two tillage regimes. It did however find a higher incidence of foot rot, bakanae, and grain discolouration in zero tillage and a higher incidence of paddy stem rot in the conventional tillage plots (Tables 7 and 8). These observations on stem rot, foot rot and bakanae in paddy are for a single cropping season as they did not appear in the other years.

FUTURE RESEARCH

This study is only an initial attempt to look at the effect of zero tillage on rhizospere fungi and diseases of rice and wheat. More interdisciplinary research needs to be carried out to better understand the effect of different tillage practices on plant growth and plant diseases. Further research is needed to better understand the ecological factors responsible for changes in disease incidence and the severity of pathogen attacks. In particular, the effect of not ploughing-in crop residues needs continuous monitoring. This study's observations need to be confirmed by large scale testing in farmers' fields across diverse agro-climatic regions.

Table 7. Effect of zero tillage on severity of foliar blight (*B. sorokiniana* and *A. triticina)* of wheat

Farmer	Leaf area infected (%)*	
	Zero	Conventional
Naresh	4.5	3.8
Dharamvir	7.6	6.3
Mean*	6.0	5.1

* Mean of *rabi* 1999-2000 and 2000-01 for wheat varieties HD 2687 and PBW 343 during 1999-2000 and 2000-01; CD at 5% : tillage practice, not significant; Farmers, 1.5; Tillage x farmer effect, not significant

Table 8. Incidence and severity of various diseases in rice paddy variety HKR 126

Disease	Incidence/Severity*	
	Zero	Conventional
False smutted tillers (%)	2.3	0.8
Sheath rot (%)	24.9	22.4
Stem rot (%)	29.1	38.3
Brown spot (0-9)	3.0	2.3
Grain discolouration (%)	15.2	9.8

* Mean of kharif 2000 and 2001 of 3 replicates of 10 hills from each plot; Source: Nakli Ram, Teek (Kharif 2000); Naresh, Uchana (*Kharif* 2001)

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Soil Health Research in Parwanipur Out-reach Command Area, Nepal

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ABSTRACT

This study compared different tillage practices for wheat within Parwanipur Regional Agricultural Research Station (Nepal) out-reach command area. The objective was to determine the effect on soil health and the microbiota profile of using minimum tillage and surface seeding to grow wheat compared to using conventional tillage. Two farmers growing successive rice-wheat crops were selected from each of Bara, Parsa, and Rautahat districts. In each district one farmer was growing wheat under conventional tillage and the other was using minimum tillage. Four more fields were sampled at Ranighat research station, Birganj.

The study recorded the population and identity of soil fungi. Soil and plant samples were collected for the rice in September 2001 and for the wheat in March 2002. At the same time the study recorded the incidence and severity of diseases on the leaves, stems and roots. Soil dilution plating was used to determine fungal population isolation on five different agar.

Sheath blight (*Rhizoctonia solani*) and brown leaf spot (*Cochliobolus miyabeanus*) were the main pathogens found on rice, while *Bipolaris sorokiniana* and *Pyrenophora tritici-repentis* were the major pathogens causing wheat leaf blight. Disease severity varied depending on location, date of planting and variety. There was less leaf spot in the rice than in the wheat crop. Root necrosis was significantly higher at two of the sites under minimum tillage in both the rice and wheat crops. Total fungal colony counts were higher to slightly higher in the conventionally tilled rice and wheat. *Rhizoctonia oryzae-sativae* and *Nakataea irregulare* were isolated from both crops. Other fungi isolated from both rice and wheat were *Aspergillus niger, Gliocladium roseum* and *Trichoderma harzianum.* No indications were found of any adverse effect on soil health of using minimum and surface seeding techniques for growing wheat in the study area.

INTRODUCTION

Rice and wheat are Nepal's most important crops. The rotation of rice and wheat is the predominant cropping practice in Nepal's Terai (lowlands) and midhills. This practice produces more than five million tons of food for the country each year. It is practiced on about 0.5 million hectares with more than 85% of all wheat in Nepal grown in this way, following a rice crop (Hobbs and Adhikari, 1997).

Much of the wheat grown in the Terai is improved varieties. However, wheat yields are not increasing and there is insufficient rice production to feed the growing population. The stagnant yields are due to the low use of fertilizer, declining soil fertility, zinc deficiencies and insect and disease problems (Gami *et al*., 1997). Several resistant varieties of rice have been developed that are less susceptible to disease, and several disease management approaches are being encouraged. The Rautahat survey found that farmers are mainly growing improved varieties of rice including Sabitri, Mansuli, Radha 11 and Makwanpur. However the effects of these changes have been little studied (Gami *et al*., 1997).

Research on crop root health in rice-wheat systems has been carried out at Naldung, Kabhre (hills) during the 1999-2000 wheat season. In this study four farmers were selected from each of a low hill (Sipaghat), a midhill (Bageswari) and a high hill (Naldung) location. The main pathogens isolated from the wheat crop were *Bipolaris sorokiniana* from the low hill location and *Fusarium graminearum* from the midhill and high hill sites (Anonymous, 2001). The important root inhibiting organisms found in the rice were *Curvularia* spp. at the low hill site and *Fusarium proliferatum* from the midhill site. *Fusarium proliferatum* was isolated from bakanae and root rot infested fields. Root knot nematode (*Meloidogyne* spp.) was found both in the soil and in the crop in the midhill rice fields.

Organisms found in the soil have both beneficial and harmful effects on crop growth. Soil plays an important role for sheltering pathogenic fungi, bacteria, viruses and nematodes. Soil contains a profile of different microorganisms in its ecology including *Fusarium* spp., *Penicillium* spp. and *Aspergillus* spp. Several types of fungi, bacteria, and nematodes are beneficial to the rhizosphere as they are antagonistic to harmful organisms. Some cereal diseases are solely caused by soil borne pathogens such as *Rhizoctonia* spp., *Pythium* spp. and *Sclerotium* spp. Several other diseases are caused by both soil and seed-borne pathogens including leaf blight of wheat and foot rot of rice. Nematodes often occur in large numbers in rice and wheat plots and some species are parasitic causing mechanical injury, necrosis and root knot galls.

The size and composition of microbial populations have been used to look at changes in the soil biota that occur in response to land management changes. They are often a good indicator of the biological status of soil. High levels of soil borne pathogens often indicate poor soil health because of the increased threat of root disease. Different types of cropping systems and tillage practices can also influence soil health for crop growth as they will influence the number of detrimental and beneficial organisms in the rhizosphere. There are many examples from Australia where minimum tillage favours the growth of sheath blight (*Rhizoctonia solani*) (Pankhurst *et al*., 1997).

The resource conservation tillage practices of minimum tillage, zero tillage and surface seeding are being rapidly adopted across the Indo-Gangetic Plain of north India and in parts of southern Nepal. In Nepal, farmers in Bhairahawa and Parwanipur in central southern Nepal are using these techniques. The results have been promising. The increasing popularity of the techniques has led to the need for research into their effects on soil health. This study was initiated to investigate any differences in soil microbiota populations (pathogenic and non-pathogenic) of rice and wheat in minimal tillage and conventional tillage. The study also investigated whether or not there is any carry over effect of pathogens from one crop to another.

MATERIALS AND METHODS

In mid-August 2001, one farmer practising conventional tillage and one practising zero or minimum tillage were selected from each of Bara, Parsa and Rautahat districts. Plant and soil sampling was carried out during the rice season in September 2001 and during the wheat season in March 2002. The study sites were at Belwa in Parsa district, Santapur in Rautahat district, and Benauli in Bara district. The Ranighat research sites were included in the study to provide more information. In these areas rice is the main summer crop with lentils and wheat as the winter crops.

Plants with soil samples were taken from each of ten points along a diagonal transect at the six farm sites. Observations were made of crop health and any disease or other crop health problems. Each sample was then assessed for disease in the laboratory and scored on a 0-5 scale with 0 representing no disease (see Bridge *et al*., 1999). Five leaves were assessed for each plant. The presence of root diseases was also assessed and scored on a 0-3 scale with 3 denoting severely diseased roots. Soil dilution methods were used to isolate fungi from the soil and to estimate fungal population. Media used for this were Malt extract agar (MEA), Malt extract-benomyl-malic acid agar, dichloran-rose bengal-chloramphenicol agar, actidione-keratin bait-tap water, and tap water agar (TWA) with hemp seed bait.

After combining and mixing the ten soil samples, one gram of soil was taken from each of the three zero or minimal tillage fields and the three conventionally tilled fields and diluted to 10^{-1}, 10^{-2} and 10^{-3} with sterile water. Each of the five agar media were inoculated with 0.1 ml of dilution suspension in two replicates. The growth of the fungal colonies was measured after two days and then a further two or three times. Total fungal counts were then counted for each dilution and each medium. Distinct fungal colonies developing on each media were transferred to Potato Dextrose agar (PDA) and MEA. Identification to genus level were made at the Plant Pathology Division, Kumalthar and to species level at CABI Bioscience, UK. Fungi isolations from roots, stems and leaves was made on PDA and malt agar and identified at species level.

RESULTS AND DISCUSSION

The survey showed that sheath blight (*Rhizoctonia solani*) was present on rice at all locations with high incidence but low severity at all sites except for the conventionally tilled Benauli fields where severity was high (Table 1). The farmer at this site grew Sona Masuli variety of rice which is known to be particularly susceptible to sheath blight. This is the third most damaging disease of rice in Nepal after blast and brown leaf spot. It was previously noted that this disease was increasing in farmers' field in the Terai (Anonymous, 1984). The current study identified the disease on leaves in the Belwa minimum tillage fields and Benauli surface seeded fields, with more in the Belwa fields.

The second most common rice disease was brown leaf disease (*C. miyabeanus*), although the severity of attack was not serious. This disease is the second most serious fungal disease of rice in Nepal (Manandhar, 1987). The most common disease observed in wheat was foliar blight -- a major disease of wheat in the Indo-Gangetic Plain that often occurrs as a complex of *Cochliobolus sativus* and *Pyrenophora tritici-repentis* in Nepal (Saari, 1998). Seeds and soils are the important source of inoculum and causing seedling and root rot.

Leaves of rice were found to be much less diseased than those of wheat (Table 1). Leaf damage was slightly higher in two of the minimum tillage rice fields and one of the minimum tillage wheat fields. Levels of root necrosis were higher in minimum tillage than conventional tillage at Belwa and Benauli, but lower at Santapur, in both the rice and wheat crops (Figure 1).

Table 1. Incidence and severity of rice and wheat diseases at six Nepal Terai sites

Village	Tillage practice	Plot	Crop	Variety	Disease						
					Sheath Blight		Bacterial Leaf Streak		Leaf blight		Leaf Rust
					Incidence	Severity*	Incidence	Severity*	Incidence	Severity*	
Santapur	Surface seeding	A 1	Rice	Sabitri	75-100	1	75-100	1	-		-
Santapur	Conventional	A 2	Rice	Sabitri	75-100	1	75-100	1	-		-
Belwa	Minimum tillage (CSD)†	B 1	Rice	Radha 12			75-100	1	-		-
Belwa	Conventional	B 2	Rice	Radha12	50-100	1-3	75-100	1	-		-
Benauli	Surface seeding	C 1	Rice	Sona Masuli	75-100	2	100	1	-		-
Benauli	Conventional	C 2	Rice	Sona Masuli	10-25	1-3	100	1	-		-
Santapur	Surface seeding	A1	Wheat	NL297	-		-		100	60	TR
Santapur	Conventional	A2	Wheat	NL297	-		-		100	50	TR
Belwa	Minimum tillage (CSD)†	B1	Wheat	Rohini	-		-		100	30	0
Belwa	Conventional	B2	Wheat	NL297	-		-		100	20	TR
Benauli	Surface seeding	C1	Wheat	NL297	-		-		100	100	
Benauli	Conventional	C2	Wheat	NL297	-		-		100	70	20S
Ranighat	Flat	R2††	Wheat	Bhrikuti	-		-		100	20	-
Ranighat	Ridge bed	R4	Wheat	Bhrikuti	-		-		90	10	-

* Bridge et. al., 1999
† Chinese Seed Drill
†† R2 = furrow irrigated raised beds (FIRB) with transplanted rice; R4= flat direct seeded in rice
TR = traces
A, B, C plots = Santapur, Belwa and Benauli; 1= resource conserving tillage practice, 2= conventional tillage

Figure 1

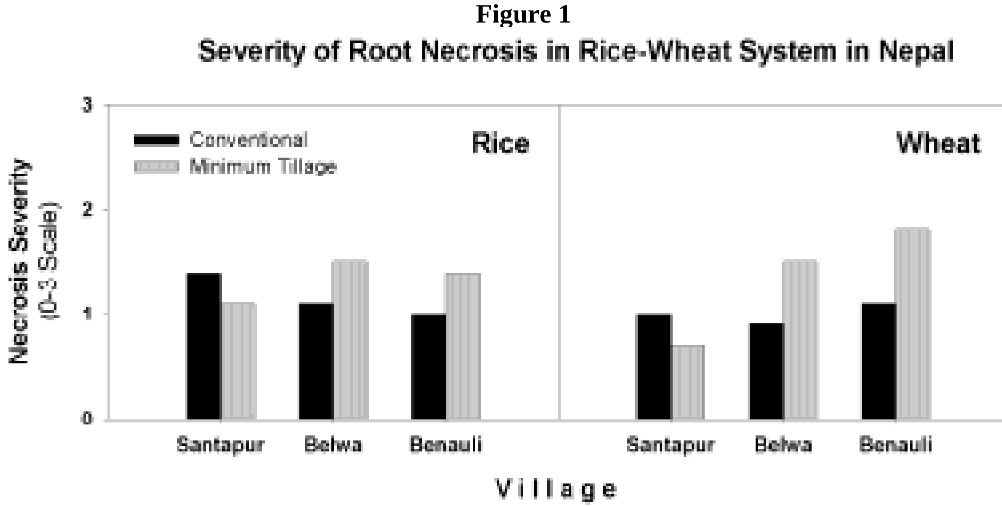
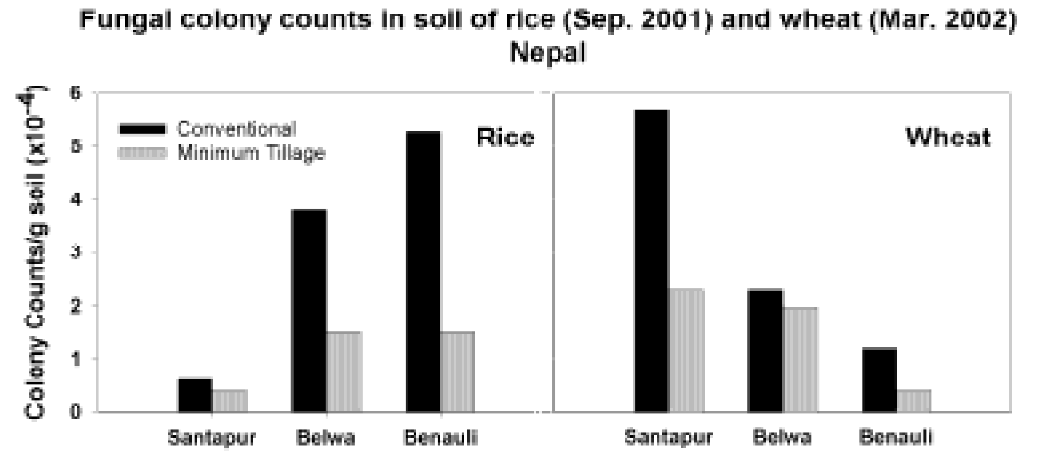


Figure 2



At all six sites the total fungal counts were higher in conventional than zero tillage fields for both crops (Figure 2). The total fungi colony counts on Rose Bengal medium were highly variable.

Eighty four types of fungi were isolated from all samples. Initial identification was done at the plant pathology laboratory, Kumalthar, and identification to species was done for the rice crop samples at CABI Bioscience, UK. Table 3 shows the seven fungi isolated from the stems of sampled rice plants. Laboratory isolation on TWA revealed the presence of *Rhizoctonia oryzae-sativae* in the rice stems from Belwa. Hashiyoka and Makino (1969) reported occurrence of this pathogen in Japan from sheath spots on rice. Another pathogen isolated from the stem of Santapur rice samples was *Nakataea irregulare*. This pathogen causes stem rot and has historically caused heavy damage to rice crops (Ou, 1985). Severe attack causes plants to weaken and fall over and incomplete rice grain formation. It was reported from Nepal in 1968 by Khadka *et al*. The fungus *Khuskia oryzae* was also isolated from the rice stems and leaves at four sites. It is mainly saprophytic but does sometimes cause leaf and ear blight (Ou, 1985). *Curvularia pallescens* was only isolated from stems.

Pyrenophora tritici-repentis was the fungus occurring most commonly on the wheat plants (Figure 3). Many occurrences were recorded from both Santapur and both Ranighat sites. At Benauli the most commonly recorded wheat fungus was *C. sativus*. Other fungi occurring were *Cercospora* sp. and *Alternaria* sp. They were also isolated from wheat stems and the occurrence was high (Figure 4). *C. sativus* occurred frequently in the conventionally tilled Santapur and Benauli sites.

Laboratory analysis of the wheat roots showed only a low occurrence of *C. sativus* at the Santapur surface seeded site, the Belwa minimum tillage site and the Benauli conventionally tilled site (Figure 5). The current study isolated *Cochliobolus lunata*, *Chaetomium sulphureum* and *Penicillium spinulosum* from the rice samples.

Aspergillus niger, *Aspergillus niveus* and *Aspergillus terreus* were commonly isolated from both rice and wheat soil samples (Table 2). *Fusarium oxysporum, Fusarium solani* and *Fusarium nygamai* were identified from rice crop soil. The study found two distinct *Fusarium graminearum*, both pathogenic on wheat in Nepal. The occurrence of *Fusarium nygamai* has been reported earlier by Bimb and Dubin (1994) at the National Wheat Experiment Research Station, Bhairahawa, and its occurrence was confirmed in this research and appears related to an African isolate. The current study isolated the bio-control fungi *Trichoderma harzianum* and *Gliocladium roseum* from both rice and wheat fields, and *Trichoderma koningii* from a rice field.

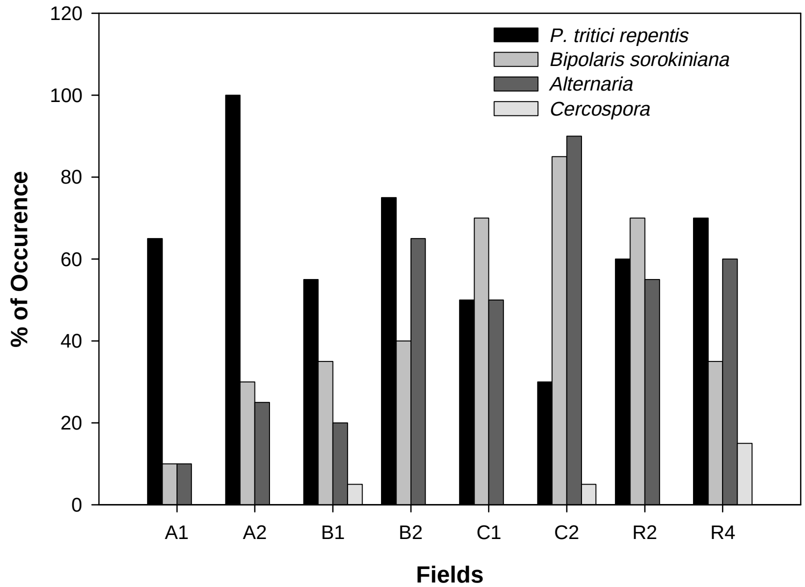


Figure 3. Occurrence of fungi in wheat leaves under different tillage practices; the field numbers refer to village details given in Table 1

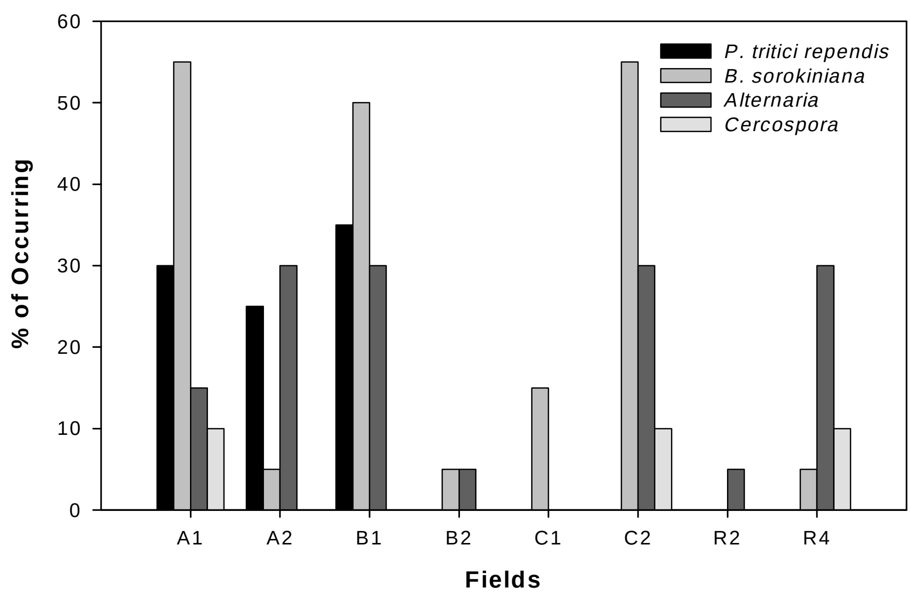


Figure 4. Occurrence of fungi in wheat stems under different tillage practices; the field numbers refer to village details given in Table 1

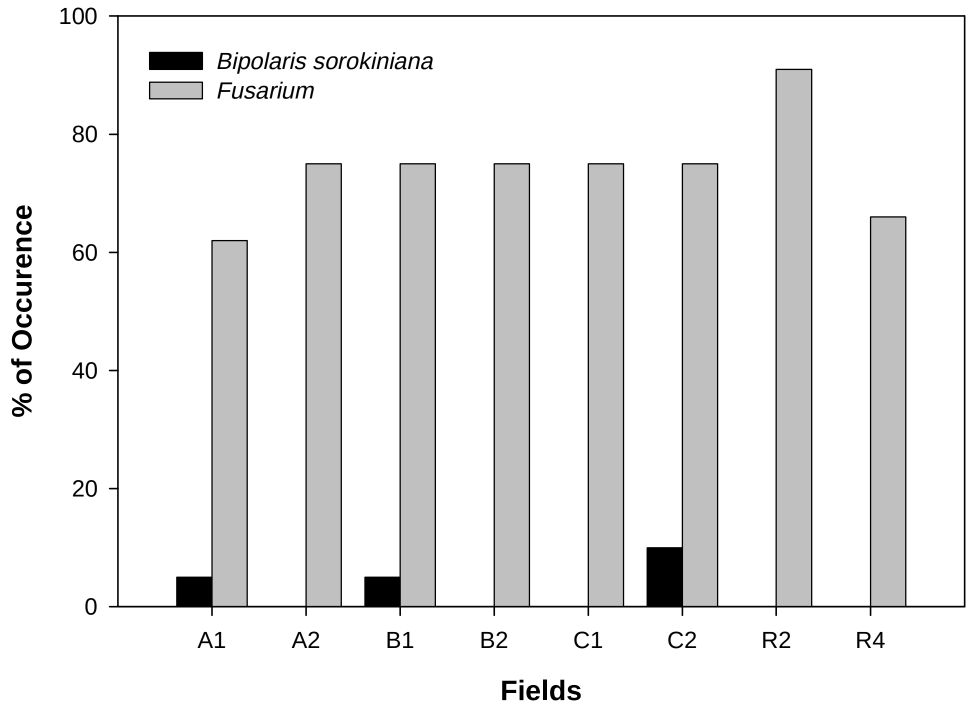


Figure 5. Occurrence of fungi in wheat roots under different tillage practices; the field numbers refer to village details given in Table 1

Table 2. Fungi isolated from rice and wheat crop soil at six sites in the Nepal Terai in 2001

Fungi	Santapur		Belwa		Benauli		Ranighat	
	Rice	Wheat	Rice	Wheat	Rice	Wheat	Rice	Wheat
Aspergillus niveus	+	+			+	+	+	+
Aspergillus terreus		+			+	+		+
Aspergillus flavus	+	+	+	+	+	+		+
Aspergillus niger	+		+		+	+	+	+
Trichoderma harzianum			+	+	+	+		+
Fusarium solani						+	+	+
Alternaria longissima	+	+	+		+		+	+
Acrophylophora fusispora	+						+	+
Gliocladium roseum			+					+

Table 3. Pathogenic fungi isolated from rice plants at six sites in the Nepal Terai

Fungi	Santapur		Belwa		Benauli		Ranighat	
	A1	A2	B1	B2	C1	C2	R2	
Rhizoctonia solani				+	+	+		
Rhizoctonia oryzae			+		+	+		
Nakatea irregulari					+			
Khuskia oryzae	+		+	+		+		
Cochliobolus oryzae	+	+	+	+	+	+	+	+
Cochliobolus lunata							+	
Curvularia pallescens	+							+

CONCLUSION

The study findings showed no significant variation in the occurrence and types of microbial flora between the resource conservation and conventional tillage sites. The adoption of minimum tillage and surface seeding for growing wheat does not appear to have increased the occurrence of crop pathogens. No distinctly different pathogens were isolated from the soil of the rice and wheat conservation tillage sites. The pathogens were only isolated from plant parts. Both *Rhizoctonia* were not isolated from soil. *Trichoderma* spp. were found in sheath blight infested fields and could be having an antagonistic effect to reduce pathogens. These findings may suggest that rice-wheat systems are sustainable. The occurrence of the beneficial organism *Trichoderma* spp. in sheath blight infested fields was also observed earlier at Sipaghat. Foliar disease was slightly higher in the minimum tillage and surface seeding sites. This could however be due to varietal differences and dates of planting. Less wheat blight was observed in the ridge beds than in the flat beds at Ranighat. Initial assessment during this study indicates that using resource conservation technologies to grow wheat does not have an adverse impact on soil health but more research is needed to evaluate the possible long term effect.

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Effect of Zero Tillage on Diseases of Rice and Wheat in Uttaranchal State, India

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ABSTRACT

This paper outlines the work conducted during the soil health project in Pantnagar University area from 1999 to 2002. The work focused on the monitoring of eight farmers' fields located in four villages (four each under zero and conventional tillage). Technical protocols developed by CABI Bioscience were followed. Results indicated that the average number of colony forming units on Rose Bengal (DRBC) medium was very similar in both tillage systems. Overall, there was no difference between the two tillage systems regarding the incidence and overall severity of rice or wheat diseases. Foliar blight severity, although slightly higher in zero tillage fields in 2000-2001 was not significantly influenced by the tillage practice used. In rice, sheath blight caused by *Rhizoctonia solani* was a major problem.

INTRODUCTION

The rice-wheat cropping systems of the Indo-Gangetic plains face several productivity and sustainability problems. These include late wheat sowing, low water and nutrient use efficiency, groundwater depletion, water logging, poor water control, salinity and the build-up of weeds, pests and diseases. The adoption of reduced and zero tillage cultivation of rice and wheat can address some of these problems. It can improve the timeliness of sowing and help farmers cope with many productivity and sustainability constraints (Harrington, 2000). However, any major shift in farming practices, such as reducing or eliminating tillage, will inevitably impact the microenvironment of crops. This, in turn, is likely to affect the type and quantity of plant pathogens.

Many plant pathogens have been reported to increase to damaging levels under no-till conditions and become constraints to efficient, profitable farming. Wheat pathogens have been reported to be either favoured or controlled by zero tillage. The fungal pathogens *Fusarium graminearum*, *Pyrenophora tritici-repentis*, *Septoria tritici* and *Pythium spp.* are reportedly favoured by zero tillage whereas *Bipolaris sorokiniana*, *Fusarium culmorum* are reduced under no till conditions (Bockus and Shroyer, 1998). In the rice-wheat systems of the Indo-Gangetic plains no information is available on the effect of zero tillage on soil health, particularly as regards the effect on potential soil borne pathogens. This study looks at the effect of zero tillage on plant and soil health in rice-wheat cropping systems in the Terai of Uttaranchal state, India.

MATERIALS AND METHODS

Four sites in Narayanpur were selected for the study. The experimental area of each site was divided into two equal parts, one for zero tillage and the other for conventional tillage. Sampling and assessments, as described below, were initiated in November 1999. Observations were recorded during three wheat seasons (1999-00, 2000-01, 2001-02) and two rice seasons (2000, 2001). An extra six study sites in Bareilly were added for the second and third seasons (Table 1).

Field based assessments -- Samples were taken from 10 points along a diagonal transect in each field. Overall crop health was scored for the presence and absence of disease symptoms. For wheat, field and laboratory assessments were done at the pre-sowing, tillering and flowering stages. The rice crop was assessed at the pre-transplanting, tillering and flowering stages.

Field assessment of aerial disease symptoms -- In each field an overall assessment of the crop was done and general observations on disease and other crop health symptoms were noted. The nearest plant to each of the 10 sampling points was scored for key foliar diseases using the 0-5 scale where 0 = no spots; 1 =1-5% spots on <50% leaves; 2 = 5-20% spots on < 50% leaves; 3 = 5-20% spots on >50% leaves; 4 =20-50% spots on < 50% leaves and 5 = >50% spots on >50% leaves.

Table 1. Details of survey fields: Fertilizer dose rice = 120:60:60 and wheat = 120:60:0; Herbicides = Butachlor @ 1-1.5 ai/ha in rice; Isoproturon and 2,4-D @ 0.75-1.25 ai/ha in wheat

Field code	Farmer	Village and district	Area (acre)	Tillage system
A-1	Anil Singh	Narayanpur, U.S.Nagar	1	Zero tillage
A-2	Anil Singh	Narayanpur, U.S.Nagar	1	Conventional
B-1	Markandey Mishra	Narayanpur, U.S.Nagar	1	Zero tillage
B-2	Markandey Mishra	Narayanpur, U.S.Nagar	1	Conventional
C-1	G.P. Mishra	Narayanpur, U.S.Nagar	1	Zero tillage
C-2	G.P. Mishra	Narayanpur, U.S.Nagar	1	Conventional
D-1	U.N.Shahi	Narayanpur, U.S.Nagar	1	Zero tillage
D-2	U.N.Shahi	Narayanpur, U.S.Nagar	1	Conventional
E-1	Azad Singh	Maksoodanpur, Bareilly	1	Zero tillage
E-2	Azad Singh	Maksoodanpur, Bareilly	1	Conventional
F-1	Madan Lal	Mitipur, Bareilly	1	Zero tillage
F-2	Madan Lal	Mitipur, Bareilly	1	Conventional
G-1	Sunder Singh	Maksoodanpur, Bareilly	1	Zero tillage
G-2	Sunder Singh	Maksoodanpur, Bareilly	1	Conventional
H-1	G-Block; univ. farm-121	Pantnagar, Bareilly	1	Zero tillage
H-2	G-Block; univ. farm-121	Pantnagar, Bareilly	1	Conventional
I-1	M-Block; univ. farm 210	Pantnagar, Bareilly	1	Zero tillage
I-2	M-Block; univ farm 210	Pantnagar, Bareilly	1	Conventional
J-1	S-Block; univ. farm-346	Pantnagar, Bareilly	1	Zero tillage
J-2	S-Block; univ. farm-346	Pantnagar, Bareilly	1	Conventional

Field based sampling of soil and plants -- After each plant had been assessed for aerial disease symptoms,, the whole wheat plant and a portion of the rice plant hill (approximately 25%) including tillers, roots and soil was dug out to a depth of 20 cm. Soil adhering to the roots was left intact. The plant and soil were placed in polythene bags for laboratory analysis.

Laboratory assessment -- Plant samples were visually scored for severity of foliar diseases using the 0-5 scale and for root lesion severity using a 0-3 scale (0 = no disease, 1 = low , 2 = medium, 3 = high level of root lesions). For soil analysis, a portion of soil adhering to the roots of each of the 10 samples was removed and the 10 portions mixed to give a composite sample. One gram of this was then used to determine fungal populations by using the serial dilution technique. The population was enumerated at three dilutions (10^{-1},10^{-2},10^{-3}) and on five different media of malt extract agar (for total fungi), malic acid agar + benomyl (for isolation of basidiomycete fungi), dichloran-rose bengal-chloramphenicol agar (DRBC) (for slow growing fungi associated with food spoilage), tap water agar (TWA) with hemp seed bait (for zoosporic fungi) and actidione-keratin bait-tap water agar (for keratinophilic fungi) (Waller *et al.*, 1998). Fungal colonies that appeared after incubation were counted at regular intervals and afinal count taken after seven days. The plant parts (leaves, roots, sheaths/stems) were cut into small pieces and plated on TWA, both before and after surface sterilization. Emergent colonies were sub-cultured onto fresh plates.

Fungal identification and statistical analysis -- The colonies purified from soil and plant parts were observed under the microscope and were identified locally to genus and species level where possible. Unidentified cultures were sent to CABI Bioscience, UK for identification. The entire data were subjected to statistical analysis using the paired t-test.

RESULTS AND DISCUSSION

The wheat field observations in the first two seasons showed no difference in the foliar blight index (Severity %) between zero tillage and conventional tillage sites. However, by the third year, all zero tillage fields showed a higher disease index than conventionally tilled fields (Figure 1).

The disease index in rice was variable with no significant difference between zero tillage and conventional tillage fields. This was the case during both rice seasons.

Laboratory assessment of the sampled plants revealed:

- no significant difference in severity of foliar blight (spot blotch caused by *C. sativus*) or average disease severity between zero tillage and conventional tillage fields in both rice and wheat seasons;
- no significant difference in severity of sheath blight and bacterial leaf blight between zero tillage and conventional tillage fields during the 2001 rice season;
- a slightly higher average disease index (severity) in zero tillage fields, indicating that there was more bacterial leaf blight in zero tillage fields;
- no difference in disease index (severity) for sheath blight of rice between zero tillage and conventional tillage fields;
- no difference in severity of root lesioning of wheat and rice plant;
- higher mean lesion severity in wheat in zero tillage fields than conventional tillage fields in the 2001-02 seasons; and
- a higher mean root lesion severity on rice in 2001 in zero tillage fields.

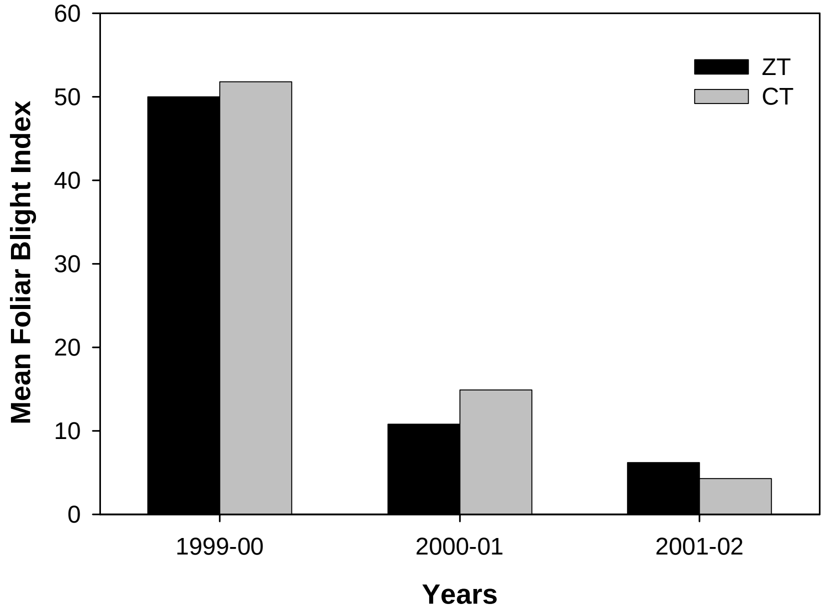

Figure 1. Foliar blight severity (%) under zero tillage (ZT) or conventional tillage (CT) in wheat over three cropping seasons

Population dynamics of fungi in soil and diversity

The total fungal population counts on DRBC agar medium were found to be very similar for both tillage systems (Figure 2).

Eleven fungal genera were isolated from wheat plants during both the 2000-01 and 2001-02 seasons. These included two pathogenic or suspected pathogenic species, two mycoparasitic and seven saprophytic genera. The frequency of isolation was similar in zero tillage and conventional tillage fields, but *Penicillium purpureogenum* was only found in conventional tillage fields. No species was specific to zero tillage wheat fields. During the second season a similar pattern was observed with the frequency of fungal isolations being similar in conventional tillage and zero tillage fields. The plant pathogenic genera identified from soil during the two wheat seasons belonged to the *Verticillium, Cochliobolus* and *Fusarium* genera.

A total of 68 fungal species were identified from the rice field soil during the 2000 crop season. These were 11 pathogenic or suspected pathogenic species, 5 mycoparasitic species and 52 saprophytic species. Amongst these were the important pathogenic fungi *Alternaria*, *Cochliobolus lunatus*, *Colletotrichum* spp. and *Curvularia* sp. Overall isolation was more frequent in the rice field soil in the zero tilled fields. Twenty fungal species, six of which are pathogenic, were specific to zero tillage fields while 15 species (4 pathogenic) specific to conventional tillage rice fields. The important mycoparasitic fungal species *Trichoderma hamatum*, *T. harzianum*, and *T. longibrachiatum* were identified.

Twenty-nine species were identified in the 2001 rice season with a higher frequency obtained for the zero tillage fields. These were 6 pathogenic, 3 mycoparasitic, and 20 saprophytic species. Amongst these, ten fungi were specific to zero tillage fields while seven were specific to conventional tillage fields. Important pathogenic species isolated from zero tillage fields included *Cochliobolus lunatus* and *Corynascus verrucosus*. In the conventional tillage fields, *Curvularia verruculosa* and *Sporotrichum pruinosum* were identified as the important pathogens.

Isolations from rice plant parts during the 2000 season yielded 23 fungal species, which included 7 pathogens and 14 suspected pathogens. Five species were specific to zero tillage fields and 4 to conventional tillage fields. During 2001, 8 fungal species were identified with 4 pathogens and 4 suspected pathogens. No fungal species were specific to zero tillage fields while 3 were specific to conventionally tilled fields. *Alternaria padwickii, Cochliobolus miyabeanus and Colletotrichum* and *Rhizoctonia genera* were the important pathogenic fungi isolated during 2000 from plant parts, while during 2001 the genera *Sclerotinia, Rhizoctonia* and *Glomerella* were identified.

CONCLUSION

Overall the study suggested a higher microbial diversity in zero tillage fields than in the conventional tillage fields in the soil during both rice seasons. In contrast, the diversity was higher in conventional tillage fields when the plant parts of the samples were analyzed.

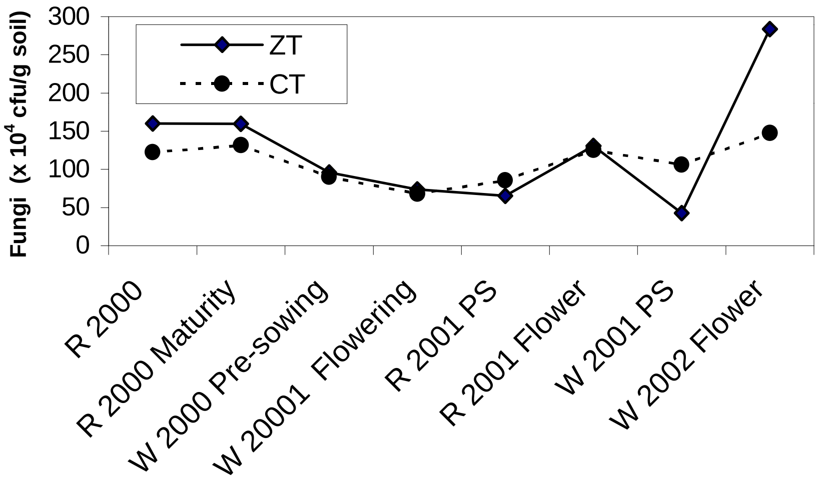

Figure. 2 Mean total fungal counts per gram of soil observed in zero (ZT) and conventional tillage (CT) in Pantnagar area in rice (R) and wheat (W) between 2000 and 2002 (PS = pre-sowing; Flower = flowering))

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Effect of Tillage Practices on Population Dynamics of Soil-borne Antagonists in Uttaranchal State, India

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ABSTRACT

This paper highlights the effect of tillage practices on soil-borne antagonists. The present investigation was carried out to study the effect of zero and conventional tillage on the population dynamics of the soil antagonists *Trichoderma* spp. and fluorescent pseudomonads in the wheat crop using the same fields that were used for the work presented by D. Joshi in the same proceedings.

INTRODUCTION

Any major shift in farming practices, such as reducing or eliminating tillage, may inevitably affect the microenvironment in which crops are grown. This in turn may alter the microbial community in the soil. These communities include both plant pathogens and the microorganisms that are natural antagonists of these pathogens. Many soil borne plant pathogens survive in the soil via the previous years' crop residues. However, no information is available on the effect of zero tillage on the population dynamics of soil antagonists and the relationship between populations of soil antagonists and soil-borne plant pathogens. The present investigation was carried out to study the effect of zero and conventional tillage on the population dynamics of the soil antagonists *Trichoderma* spp. and fluorescent pseudomonads.

METHODOLOGY

The study was undertaken in eight zero tillage fields and eight conventionally tilled fields under a rice-wheat cropping sequence. The fields are located in the Terai in India's Uttaranchal state (see associated paper by Deeksha Joshi *et al.*, Table 1). The population of *Trichoderma* spp. and fluorescent pseudomonads was estimated from soil and rhizosphere samples collected at the grain filling stage in wheat during 2002.

For rhizosphere analysis, a total of ten plants (diagonally selected from the fields) were uprooted. Care was taken to avoid any root system loss while uprooting. Loosely adhering soil particles were removed by gentle tapping; roots were then transferred into polythene bags and the samples taken to the laboratory for analysis. The rhizosphere soil was removed by mechanically scraping the root surfaces. Soil from the ten sample plants in each field was combined and mixed thoroughly to give uniform composite samples. Non-rhizosphere soil samples were also collected from between the rows.

The rhizosphere and non-rhizosphere soil samples were subjected to serial dilutions. One gram of soil was diluted ten times (10^{-1}). The soil suspended in the tube was shaken thoroughly to mix and uniformly disperse soil particles. One ml of the suspension from the first dilution (10^{-1}) was aseptically transferred to another tube and diluted to 10^{-2}. This procedure was repeated until the original sample was diluted to 10^{-3} (for *Trichoderma*) and 10^{-5} (for fluorescent pseudomonads). *Trichoderma* selective medium (TSM) (Elad *et al.,* 1981; Mukherjee, 1991) was used to selectively isolate the fungal antagonists whilst King's B medium (King *et al.,* 1954) was used to isolate the bacterial antagonists. The data was analysed by paired t–test.

RESULTS

Population dynamics reported in Table 1 showed the following:

- In rhizosphere soil, at four of the sites, the fluorescent pseudomonads count was higher in zero tillage fields than conventional tillage fields (with no bacteria being recovered from conventional tillage fields at two sites), while at the other four sites the reverse was true.

- In rhizosphere soil, *Trichoderma* counts were lower in zero tillage fields than conventional tillage fields at six of the seven sites where the fungus was recovered. In fact the fungus was not recovered from seven of the 16 fields investigated, and not at all from one site.

- In the non-rhizosphere soil, pseudomonads counts were higher in zero tillage fields than conventional tillage fields at all eight sites.

- In the non-rhizosphere soil, *Trichoderma* counts were higher in zero tillage fields than conventional tillage fields at five of the eight sites where the fungus was recovered and lower in only two. Again the fungus was not recovered from seven of the 16 fields investigated and not at all from one site.

In the zero tillage fields, the mean population of fluorescent pseudomonads was generally higher in both the rhizosphere and non-rhizosphere soils while the mean population of *Trichoderma* was greater only in the non-rhizosphere soils except in fields G1, H1 and J1 where the population count was zero (Table 1). On the other hand, conventional tillage fields had higher rhizosphere populations of *Trichoderma* except in fields F2 and J2 where the count was zero. Rhizosphere and non-rhizosphere populations of pseudomonads in zero tillage fields and of *Trichoderma* in conventional tillage and zero tillage fields were almost the same. A significant negative correlation was observed between the populations of *Trichoderma* and Pseudomonads in rhizosphere, but was positive in the non-rhizosphere samples.

There was a significant negative correlation between the total fungal population and the population of *Hirschmanniella* sp. with the population of fluorescent pseudomonads.

DISCUSSION

These results present a high level of variation and further studies are needed to understand the trends observed in this research. The finding that fluorescent pseudomonads were generally higher in zero tillage than in conventional tillage fields suggests that tillage practices affect the population of these bacteria. However, more investigation is needed to understand why this result is more observed in non-rhizosphere soil. The pattern of the *Trichoderma* population was quite variable and difficult to analyze. However, there was a significant negative correlation between the population of the bacterial antagonist fluorescent pseudomonads and the fungal antagonist *Trichoderma* in the rhizosphere samples. These pseudomonads are in general antagonistic to *Trichoderma* (Singh, U.S., personal communication). However, there was no negative correlation between the population of these antagonists in the non-rhizosphere soils. This was probably because the two tend not to interact directly with each other in soil.

There was a negative correlation between the population of fluorescent pseudomonads and the population of *Hirschmanniella* sp. Fluorescent pseudomonads have been observed to suppress rice root lesion nematodes. It would be worthwhile to extend this study to see whether fluorescent pseudomonads populations can serve as bioindicators to estimate populations of *Hirschmanniella* and total fungal populations in soil.

Table 1 Population dynamics of fungal and bacterial antagonists (2002 wheat season) in zero tillage (ZT) and conventional tillage (CT)

Field*	Rhizosphere soil		Non-rhizosphere soil	
	Fluorescent pseudomonads (x 10^5)	*Trichoderma* spp. (x 10^3)	Fluorescent pseudomonads (x 10^5)	*Trichoderma* spp. (x 10^3)
B1 (ZT)	34.22	0.66	06.66	3.33
B2 (CT)	00.00	4.33	05.33	0.00
D1 (ZT)	45.00	0.00	48.33	0.33
D2 (CT)	86.33	1.33	08.66	0.00
E1 (ZT)	19.00	0.00	51.00	2.00
E2 (CT)	28.00	1.00	08.33	1.00
F1 (ZT)	43.66	0.00	44.33	7.66
F2 (CT)	27.33	0.00	18.66	1.66
G1 (ZT)	21.66	0.00	21.66	0.00
G2 (CT)	24.00	2.30	20.33	1.66
H1 (ZT)	34.33	0.00	34.33	0.00
H2 (CT)	26.66	3.00	34.00	0.33
I1 (ZT)	25.33	0.33	35.66	0.33
I2 (CT)	45.66	1.33	15.00	0.00
J1 (ZT)	42.00	1.00	26.00	0.00
J2 (CT)	00.00	0.00	14.33	0.00

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Effect of Zero and Conventional Tillage on Fungi in Rice-Wheat Cropping Systems in Pakistan

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ABSTRACT

Soil microflora is important for sustainable agriculture as its activity contributes to increasing agricultural production. Disease susceptibility of crops may be lowered by better understanding the interactions between pathogens and crop residue and then modifying local environmental conditions, crop rotations, tillage practices, and antagonistic mycoflora accordingly. The objective of this study was to examine the effect of zero tillage on soil borne cereal diseases in rice-wheat cropping systems in Pakistan. The study found no distinct difference in the presence of fungi associated with foliar and root diseases of wheat and rice in zero and conventional tillage systems. Isolating fungi from plant parts and soil on potato dextrose agar (PDA) medium showed little variation between zero tillage and conventional tillage. Total fungal colony counts showed that reduced tillage could be enhancing the diversity of soil mycoflora; but the scale of the current study was insufficient to give definitive results on the effect of reduced tillage on the survival of pathogens in soil.

INTRODUCTION

Wheat and rice are the major cereals grown in Pakistan. Official figures report that 21,078 thousand tonnes of wheat were grown over 8,463 thousand hectares and 5,155 thousand tonnes of rice were grown over 2,515 thousand hectares in the year 1999-2000 (Anonymous, 1999). The main growing areas for both crops are the Punjab where these crops are grown one after another in the same fields. Zero tillage is being piloted in these areas as a more sustainable way of growing these crops by saving time and resources. The main purpose of the current study was to see the effect that zero tillage had on fungi associated with the crops and the soil mycoflora. Soil borne pathogens often constrain crop yields by damaging seeds, seedlings, roots, lower leaves, and leaf sheaths. They lower yields by reducing emergence, plant vigour, and tillering. These pathogens seriously affect many crops and cause great losses (Mathur and Cunfer, 1993).

The current study seeks to improve understanding of the dynamics of soil borne fungi, including pathogens and associated competitors and antagonists, under resource conservation technologies in a rice-wheat cropping system.

MATERIALS AND METHODS

Site selection and sampling -- Two sites were selected in the rice-wheat cropping areas of Muridke and Mona (Bhalwal) in the

Punjab of Pakistan. The three-year study took place in one field under conventional tillage and one with either zero or minimum tillage at each site. Sampling was done at the maturity stage of the rice crop in September 2000 and October 2001, and for the wheat crop in March 2000, March 2001 and March 2002. Sampling was carried out at 10 points across the field, five paces apart along a diagonal transit. The nearest plant to each sampling site was dug out with a narrow bladed tool to include tillers, roots and soil to a depth of 20 cm. Soil adhering to the roots was left intact. The plant portion and soil were put in a polythene bag and taken to the laboratory.

Disease assessment -- The plants were assessed for disease by noting the incidence and severity of foliar spots (foliar blight of wheat [*C. sativus* and *P. tritici-repentis*]; brown spot, sheath blight and blast of rice) and root rot of both crops. Foliar disease was assessed on the IRRI 0-5 rating scale where 0 = no symptoms, and 5 = 50% spots on 50% or more leaves (Bridge *et al.,* 1999). For the rice crop, foliar diseases included brown spot (*C. miyabeanus*), bacterial leaf blight (*X. oryzae*), sheath rot (*S. oryzae*), sheath blight (*R. solani*) and blast (*M. grisea*). Root rot was measured on a 0-3 rating scale (Ledingham *et al.,* 1973) where 0 = clean and 3 = severe rot.

Isolation of fungi from foliage and roots -- Pieces of leaves from spotted areas and lesion affected portions of roots were cut up and surface sterilized in 1% chlorox solution for one minute then rinsed in sterilized distilled water. The pieces were plated on potato dextrose agar medium (PDA) after drying on sterilized filter paper. Five pieces were placed in 9 cm sized petri plates and incubated for seven days at 22°C. Fungal cultures isolated from root and foliar portions were then transferred onto PDA slants for further study. Fungal material was mounted on slides in lactophenol and cotton blue stain and examined under stereo and light microscopes. Fungi were identified by reference to different keys (Barnett, 1960; Domsch *et al*., 1980; Gilman, 1945).

Isolation of fungi from soil – About 100 cm^3 of adhering soil was manually removed from the roots. All ten soil sub-samples were combined and well-mixed to form one composite sample. One gram of soil from each sample was then taken to prepare a suspension (10^{-1} dilution) by placing it in 9 ml of sterilized distilled water in a sterile tube. The tube was capped tightly and shaken for 30 min. Second (10^{-2}) and third (10^{-3}) dilutions were then prepared. The 10^{-2} and 10^{-3} dilutions were used to inoculate each of malt extract agar (MEA), malt extract-benomyl-malic acid agar, dichloran-rose bengal-chloramphenicol agar (DRBC), actidione-keratin bait-tap water agar, and tap water agar (TWA) with hemp seed bait (Bridge *et al.*, 1999). This was done by adding 0.1 ml of suspension and immediately spreading it using a sterile spreader. Plates were sealed with tape and incubated at 20-22°C. These samples were monitored daily for the emergence of fungal colonies. Fungal mounts were then prepared from mature colonies which were then identified.

RESULTS

Foliar and root diseases

The disease index was calculated using the following formula:

Disease Index =

$$\frac{(0*(\text{Plants in c. }0)+1*(\text{Plants in c. }1)+2*(\text{Plants in c. }2)+3*(\text{Plants in c. }3))\text{X}100}{\text{Number of total plants}}$$

Results from the Muridke site data (Figure 1) show that there was less foliar disease (foliar spot of wheat, brown spot, sheath blight and rice blast) under conventional tillage than under zero tillage in the first three crops. However, there was less foliar disease in the zero tillage soils in the fourth and fifth crops (Oct-01 and Mar-02). There was about the same amount of root disease in the first crop (wheat) and the second crop (rice) in both types of tillage; but more root disease under conventional tillage than zero tillage in the third and fourth crops. Then, in the fifth season there was much less disease in wheat roots under conventional tillage.

At the Mona site (Figure 2), the foliar disease index for the 2000 wheat and rice crops, and the 2001 and 2002 wheat crops was slightly lower under conventional tillage than under zero tillage. But in the 2001 rice crop the disease index was slightly higher in the conventional than the zero tillage crops. At this site the root disease index was higher (110 and 125) in the wheat crop during first and fifth crops under conventional tillage, whereas it was the same in the roots of the 2000 rice crop under tillage types. But it was a little lower in the 2001 conventional tillage wheat and rice than under zero tillage.

Fungi isolated from crop leaves

A number of fungi were isolated from the foliar parts of the two crops (Table 1). The important rice pathogen *C. miyabeanus* was isolated only in 2001 from both tillage systems. The important wheat pathogen *B. sorokiniana* was isolated in two consecutive years (2001 and 2002) under conventional tillage. *Fusarium* species (*F. equiseti*, *F. graminearum*, *F. oxysporum and F. solani*) are responsible for head scab and root rot in cereals and were isolated from the wheat crop in 2000 and 2002. *Nigrospora* species are mainly saprophytes and occur in crops already weakened by disease, insect pests or poor nutrition. *Nigrospora oryzae*, a weak pathogen of rice that infects leaves and leaf sheaths, was found on leaves from both the zero and conventional tillage fields. In wheat this fungus was present as a saprophyte on leaves in 2001. The soil borne pathogen *Rhizoctonia cerealis*, which causes sharp eyespot in wheat, was isolated from wheat leaves under both zero and conventional tillage during 2002. A *Verticillium* sp. was also isolated from wheat under both systems.

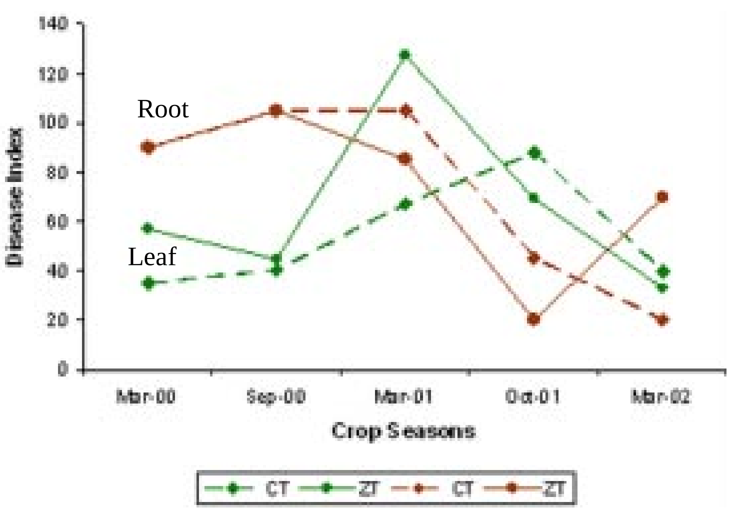

Figure 1: Leaf and root disease index in rice and wheat crops under zero (ZT) and conventional tillage (CT) at Muridke (2000-2002); rice was evaluated in September and wheat was scored in March.

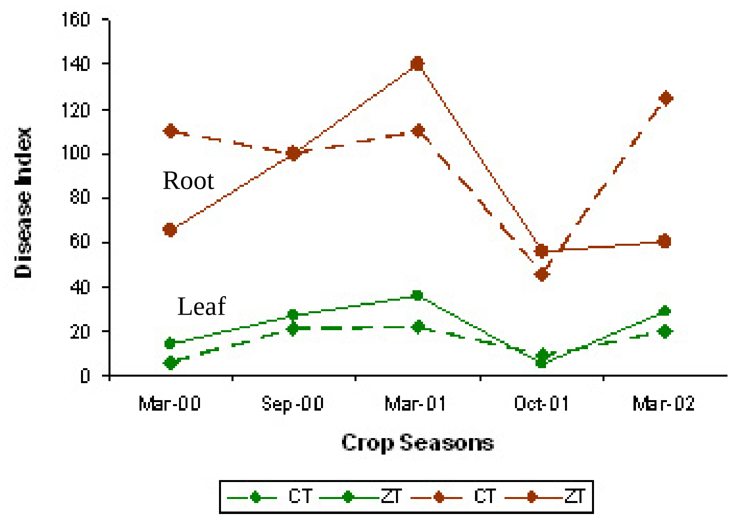

Figure 2: **Leaf and root disease index in rice and wheat crops under zero (ZT) and conventional tillage (CT) at Mona (2000-2002); rice was evaluated in September and wheat was scored in March**

Table 1 **Pathogens isolated from leaves in rice-wheat cropping systems under zero (ZT) and conventional tillage (CT)**

Pathogens	March 2000		Sept. 2000		March 2001		Sept. 2001		March 2002	
	Wheat		Rice		Wheat		Rice		Wheat	
	ZT	CT	ZT	CT	ZT	CT	ZT	CT	ZT	CT
Bipolaris oryzae	-	-	-	-	-	-	+	+	-	-
Drechslera sorokiniana	-	-	-	-	-	+	-	-	-	+
Fusarium equiseti	-	-	-	-	-	-	-	-	+	-
F.graminearum	-	-	-	-	-	-	-	-	-	+
Fusarium oxysporum	+	+	-	-	-	-	-	-	+	-
Fusarium solani	+	-	-	-	-	-	-	-	-	-
Nigrospora oryzae	-	-	+	+	+	+	+	+	-	-
Rhizoctonia cerealis	-	-	-	-	-	-	-	-	+	+
Verticillium sp	-	-	-	-	-	-	-	-	+	+

Fungi isolated from crop roots

The major rice pathogen *C. miyabeanus* was again isolated from rice roots and from foliar parts under both zero and conventional tillage in 2001 (Table 2). *Fusarium* species including *F. equiseti*, *F. graminearum*, *F. oxysporum* and *F. solani* were mainly isolated from the wheat crop under both tillage systems in 2000 and 2002. A few *Macrophomina phaseaolina* fungi, were isolated from the rice roots during 2001 but only under zero tillage. It is not a major pathogen of rice and wheat crops. The weak parasite *Nigrospora oryzae* was also isolated in 2000 and 2001 from rice roots and from foliar samples. *Rhizopus* species, which contribute to black point in wheat seeds, was isolated from roots in 2001 both in zero and conventional tillage. Another seed and soil borne pathogen, *Sclerotium oryzae*, which causes stem rot in rice, was isolated from rice roots during 2000 under conventional tillage (Table 2).

Table 2 Pathogens isolated from roots in rice-wheat cropping systems under zero (ZT) and conventional tillage (CT)

Pathogens	March 2000		Sept. 2000		March 2001		Sept. 2001		March 2002	
	Wheat		Rice		Wheat		Rice		Wheat	
	ZT	CT	ZT	CT	ZT	CT	ZT	CT	ZT	CT
Bipolaris oryzae	-	-	-	-	-	-	+	+		
Fusarium equiseti	+	-	-	-	-	-	-	-	+	-
F.graminearum	-	+	-	-	-	-	-	-	-	-
Fusarium oxysporum	-	+	-	-	-	-	-	-	+	+
Fusarium solani	+	+	-	-	-	-	-	-	+	+
Macrophomina phaseolina	-	-	-	-	-	-	+	-	-	-
Nigrospora oryzae	-	-	-	+	+	+	+	+	+	-
Rhizopus spp.	-	-	-	-	+	+	-	-	-	-
Sclerotium oryzae	-	-	-	+	-	-	-	-	-	-

Fungi isolated from soil and populations on specific media

The pathogenic fungi *Bipolaris sorokiniana*, *Fusarium graminearum*, *Fusarium solani*, *Macrophomina phaseaolina*, *Rhizoctonia solani*, and *Rhizopus oryzae* were isolated from the soil samples (Table 3). All these are seed borne and can therefore survive in soil for long periods. All except *B. sorokiniana*, *M. phaseolina* and *F. solani* were isolated from conventional tillage sites. The beneficial fungi *Trichoderma viride* was isolated only once in the 2002 conventionally tilled crop.

In this study a total of 590 fungal colonies were derived from the conventional tillage samples and 526 from the zero tillage samples on the five different media. Over the three years of the study, the colony counts show only a small difference between the two tillage systems (Figures 3 and 4).

Table 3 Pathogens isolated from soil in rice-wheat cropping systems under zero (ZT) and conventional tillage (CT)

Pathogens	March 2000		Sept. 2000		March 2001		Sept. 2001		March 2002	
	Wheat		Rice		Wheat		Rice		Wheat	
	ZT	CT	ZT	CT	ZT	CT	ZT	CT	ZT	CT
Bipolaris sorokiniana	-	-	-	-	-	-	-	-	+	+
Drechslera sorokiniana	-	+	-	-	-	-	-	-	-	+
Fusarium graminearum	-	-	-	-	-	-	-	+	-	-
Fusarium solani	-	+	-	-	-	-	-	-	+	+
Macrophomina phaseolina	-	+	-	-	-	-	-	-	+	+
Rhizoctonia solani	-	-	-	-	-	-	-	+	-	-
Rhizopus oryzae	+	-	-	-	-	-	-	-	-	-
Trichoderma viride	-	-	-	-	-	-	-	-	-	+

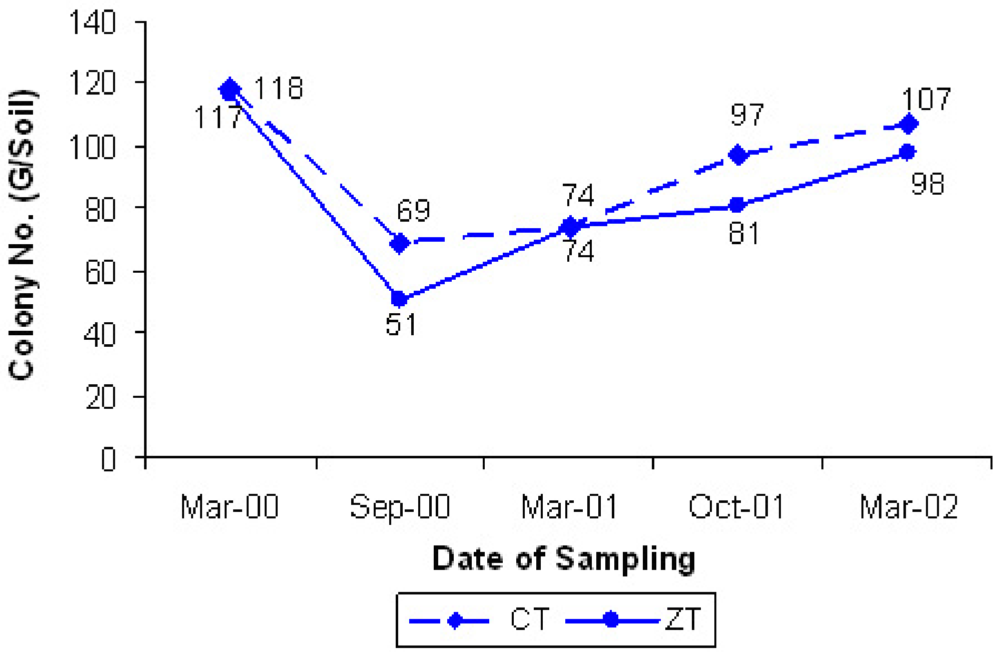

Figure 3: **Total fungal population of soils in rice and wheat crops under zero and conventional tillage (2000-2002); rice was evaluated in September and wheat in March**

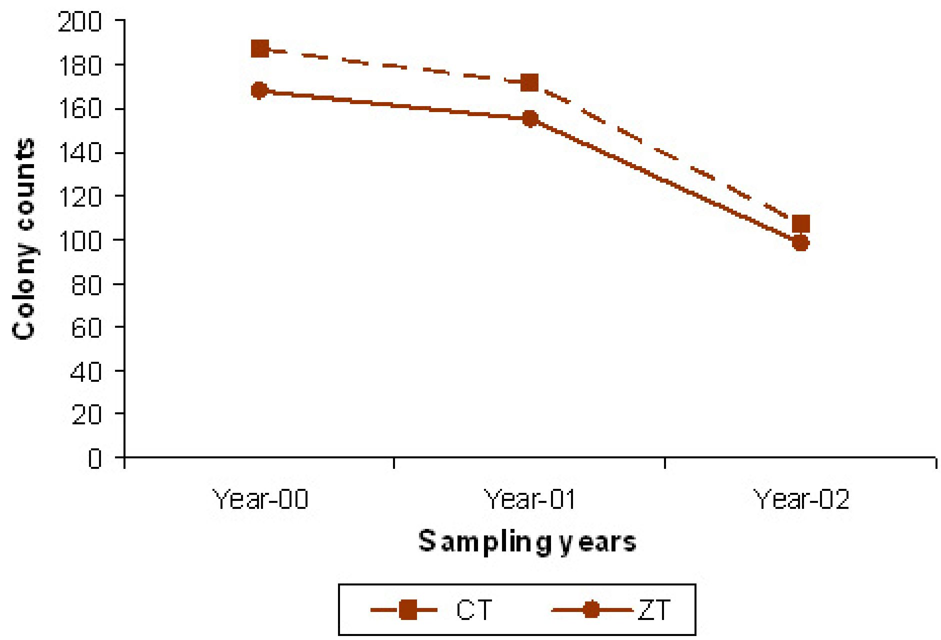

Figure 4: **Year-wise total fungal population in soils of rice- wheat cropping systems under zero and conventional tillage (2000-2002)**

DISCUSSION

Zero tillage is considered a more sustainable way of growing crops as the lower inputs make it more energy efficient. This study assessed disease levels in soil and plant samples and isolated fungi on different media. In the Muridke site the foliar disease index varied over the five cropping seasons with the lowest values recorded under conventional tillage during the first three seasons. In the last two seasons it became higher under zero tillage for both crops cultivated one after another in the same fields. However, from the Mona data it is difficult to see any effect of soil biotic constraints on crop health under the two types of tillage. At this site the root disease index showed no significant effect of zero tillage on crop health as compared to conventional tillage. Similar observations of root necrosis at Muridke also showed no clear effects of the new tillage system. Studies in western Canada have found that reduced tillage lowers the impact of some root diseases but increases the impact of foliar diseases on cereals (Bailey and Duczek, 1996). However, the present study shows no distinct difference between soil-borne fungi associated with foliar and root diseases of wheat and rice in relation to the two tillage systems.

The soil borne pathogenic fungi *C. miyabeanus*, *B. sorokiniana*, *Fusarium* spp., *Macrophomina phaseolina*, *Rhizoctonia solani*, *Rhizoctonia cerealis*, *Sclerotium oryzae*, *Rhizopus oryzae* and *Verticillium* sp. were identified from the plant and soil study samples. *C. miyabeanus* was isolated from both the foliar and root parts of rice plants in the same year; but its absence in the soil in the last three crops suggests that inoculum might come from spores which persist in the soil irrespective of tillage regime. *B. sorokiniana*, known worldwide as the major cause of root and foot rot in cereal crops (Hill *et al.,* 1983; Diehl, 1979; Fedle-Moen and Harrison, 1987), also causes leaf spot in certain conditions. This fungus was isolated from wheat leaves in 2001 and 2002 under conventional tillage and from soil under both tillage systems. This supports Bailey's 1996 findings that reduced tillage lowers the impact of some root diseases on cereals.

Fusarium graminearum was present in a 2001 soil sample at a conventionally tilled site and in the following crop at the same site on foliar parts of wheat. This shows its persistence in the soil under conventional tillage. The other *Fusarium* species of *F. oxysporum*, *F. solani* and *F. equiseti*, generally considered to be less significant causes of root and foot rot (Diehl, 1979 and Iftikhar *et al.*, 1991), were isolated from roots and soil in 2002 in both tillage regimes. Isolation of the soil borne pathogens *Macrophomina phaseolina*, *Sclerotium oryzae*, and *Rhizoctonia solani* from the soil and plants of both crops did not show anything significant on the effect of reduced tillage on these pathogens. The discovery of a *Verticillium* sp. on wheat is a new finding for Pakistan.

Nigrospora and *Rhizopus* species are common saprophytes of rice worldwide. In some regions their association with other fungi causes the discoloration of seeds. Their isolation frequency in the two tillage regimes was similar to other pathogens. The fact that *Trichoderma viride* was only isolated during 2002 under conventional tillage indicates its association with this tillage system in contrast to other pathogens which showed no difference between the two tillage regimes. The low frequency of soil pathogens during the rice crops may be due to rice flooding. Overall no remarkable difference was observed between the two tillage practices.

Season-wise, the total counts of fungi isolated from soil decreased under zero tillage in the second crop (rice) but was about the same during the third crop (wheat) under both regimes. In the fourth crop (rice) and in the following wheat crop the colony counts decreased indicating the positive effect of zero tillage in rice-wheat cropping.

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Nematode Assessment in Minimum and Conventionally Tilled Rice-Wheat Fields in Nepal

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ABSTRACT

Field surveys were carried out in three minimum tilled and three conventionally tilled rice fields in 2001, and wheat fields in 2002, in the Nepalese Terai. The aim was to identify the types of nematodes present in these crops and the associated soil. The study found no difference in the occurrence of root lesions between minimal tillage and conventional tillage field rice and wheat crops except in the Benauli rice crop. Root knots in rice showed a variable response to the two tillage practices. In wheat roots, root knot was observed only in the Belwa fields and the findings showed no relation to tillage type. There was only a small difference between the populations of *Hirschmanniella* spp. in the minimal tillage and conventional tillage fields. *Meloidogyne graminicola* was found only in the Belwa conventional tillage field in rice roots whereas it was found in both minimal tillage and conventional tillage fields in wheat roots at the same site in the same frequency. Among the migratory soil and root nematode pests *Hirschmanniella oryza* and *H. mucronata* were found in rice. These two species and *M. graminicola* were also found in the wheat crop at Belwa. Slightly more phytoparasitic nematodes were found in the minimal tillage than the conventional tillage fields in two of the sites in both the rice and wheat soil. Bacterial feeders and omnivorous nematodes occurred more in the conventional tillage than the minimal tillage rice fields. Fungivorous nematodes were also present in the wheat, with more in the conventional tillage than minimal tillage samples. These findings indicate that nematode genera, species, and trophic groups have a variable response to conventional and minimum tillage practices.

INTRODUCTION

Diversity of form and feeding habits allow soil nematodes to use every type of available substrate. Nematodes alter crop production by parasitizing roots and influencing decomposition and mineralization by interacting with microorganisms. These free living and plant-parasitic nematodes therefore occupy an important position in the soil detritus food web (Pankhurst, 1997). Any changes to the soil habitat of nematodes that influences their food source or environment, such as changing soil or crop management practices, will usually be reflected in changes in the biodiversity of the nematode community (Pankhurst, 1997). Soil nematode communities have thus been used as biodiversity indices in several studies to document the impact of perturbations on soil biological activity (Neher *et al.*, 1995).

The population level of functional groupings of nematodes and the use of indices based upon species level comparisons may be useful qualitative indicators of soil health

(Niles and Freckman, 1996). Freckman (1998) suggested that the productivity of the below-ground systems may be indicated by the number of bacteriophage nematodes present. Wardle (1995) has reported the influence of tillage (conventional compared to no tillage) on the biodiversity of several taxonomic and functional groups of soil organisms including fungi, nematodes, collembola, mites, earthworms, spiders and carabid beetles. Wardle concluded that the effect of different kinds of tillage on species diversity was less for smaller soil organisms (i.e. the fungi and the nematodes) than for the meso- and macro-fauna. Yates and Bird (1994) observed higher numbers of nematodes under minimum tillage compared to conventional cultivation, but the effects were small and seasonally variable. According to Freckman and Caswell (1995), predacious nematodes prefer the upper layers of moist soils. The fact that minimally tilled soils tend to be more moist, especially near the soil surface, may favour this trophic group. In general, the effects of minimum tillage on plant parasitic nematodes are variable (Yates and Bird, 1994).

Long term studies of reduced and conventional tillage systems have revealed distinct differences in the microfauna and in the functional processes in the soil (Roper and Gupta, 1995). Andren and Lagerlof (1983) reported that tillage generally favours organisms with short generation times, high metabolic rates and rapid dispersal. This may be why residue incorporation leads to more bacterial growth and hence an increased abundance of bacterial feeding protozoa, bacterial feeding nematodes and enchytraeids. In contrast, Gupta and van Vliet (1996) reported that fungi dominate over bacteria under no tillage systems resulting in an increased abundance of fungal feeding protozoa and fungal feeding nematodes. However, Parmelee and Alston (1986) found that during the summer a relatively greater number of fungivorous nematodes were recorded from a no-till treatment and bacterial feeding nematodes from conventional tillage. These studies therefore suggest variability in response to tillage changes for different types of nematodes.

This study was carried out to find out the soil health status of minimum and conventionally tilled fields by looking at the presence and population of different types of nematodes, and the extent of damage they caused, in rice-wheat systems in Nepal.

MATERIALS AND METHODS

The study selected a minimally and a conventionally tilled field cropped in a rice-wheat sequence from each of Bara, Parsa and Rauthat districts (Table 1). All survey sites are in the Terai region of Nepal with sub-tropical climates.

Table 1 Location of minimally and conventionally tilled rice and wheat field survey sites in the Nepalese Terai

Code	Tillage practices	Name and Address of farmers
A1	Minimum (surface seeded)	B Choudhary, Santapur, Raughat
A2	Conventional	B Choudhary, Santapur, Raughat
B1	Minimum (direct seeded)	A Choudhary, Belwa, Parsa
B2	Conventional	J Das Tharu, Belwa, Parsa
C1	Minimum (surface seeded)	Prasad Sah, Benauli, Bara
C2	Conventional	RJP Yadav, Benauli, Bara

Sampling was done in both the rice and wheat growing seasons. Sampling for rice was done at the flowering stage in

September 2001, and for wheat at the dough stage in March 2002. Plants were sampled at five pace intervals on a diagonal transect (Bridge *et al.,* 1999). A total of 10 plant-root-soil samples were taken from each field. Each sample was collected in a separate plastic bag and labelled.

The root samples were thoroughly washed to free them from soil and other contamination. Root necrosis and root knot indexing were then done to assess levels of root damage. The levels of root necrosis were scored on the 0-3 scale where:

0 = no root necrosis or lesioning;
1 = up to 25% necrosis;
2 = 25-50% necrosis;
3 = more than 50% necrosis.

Root knot indexing (0-10) was also carried out to evaluate the level of infestation of root knot nematode (Luc *et al.,* 1990), where:

0 = complete and healthy root system with no infestation;
1 = a very few small galls which can only be detected upon close examination;
2 = small galls as in '1' but more numerous and easy to detect;
3 = numerous small galls, some grown together, with root function not seriously affected;
4 = numerous galls, with majority of roots still functioning;
5 = 25% of roots galled but roots functioning;
6 = 50% of roots galled and roots not functioning;
7 = 75% of roots galled and lost for production;
8 = no healthy roots, with nourishment of plant interrupted, but plant still green;
9 = the completely galled root system is rooting, but plant is dying;
10 = plant dead.

Before root washing, the soil from around the roots of each sample was taken and combined for each sampled field. The final composite soil sample from each site was 100cc. The nematodes were extracted in a pie pan using the method suggested by Bridge *et al.* (1999). The suspension containing the nematodes was collected after 24 hours of processing.

Washed roots were dried on tissue paper before being cut into 0.5-1 cm long pieces and mixed together. Two 2g sub-samples were randomly picked out and weighed. Each sampled and weighed root was again cut into small pieces with sharp scissors. The roots were wrapped in fine tissue paper to avoid excess moisture. Two grams of root was placed on a tissue paper on a sieve in a small container and covered with water and left for 48 hours. The nematode suspensions were then collected (Bridge *et al.,* 1999).

Each suspension was carefully poured into graduated tubes and allowed to settle for about three hours. Excess top-water was siphoned off leaving a 20 ml final suspension. Nematodes were counted in 10% (i.e. 2 ml) of aliquot nematode suspension on a nematode counting disc under a binocular microscope. A compound microscope was used for identifying nematode species, genera, and trophic groups using an illustrated key (Hunt, 2000).

Most data were analysed descriptively and presented in mean and percentage figures. A paired t-test was done to test the mean values (Steel and Torrie, 1986).

RESULTS AND DISCUSSION

Root damage assessment

Root necrosis in rice was found to be higher minimally tilled fields than in the conventionally tilled fields at Benauli (Table 2). Samples from the other two sites showed no difference in root necrosis between the two tillage types. Root knot symptoms in conventional tillage fields were found only at the Belwa (B2) and Benauli (C2) sites. Significantly more root knots were found in the Belwa conventional tillage field than in the same site's minimal tillage field. However, there was no difference between the Benauli minimal tillage and conventional tillage sites (C1 and C2).

Wheat root necrosis was higher in the Belwa and Benauli minimal tillage field sites, but there was no difference between the minimal tillage and conventional tillage fields for the Santapur sites (Table 2). Root knot symptoms were only found in both Belwa fields with no significant difference between its occurrence in the minimal tillage and conventional tillage sites.

Root Nematodes

The rice root nematode (*Hirschmanniella* spp.) was the dominant root nematode of rice in the sampled areas (Table 3). There was only a small difference between the populations of *Hirschmanniella* spp. in the minimal tillage and conventional tillage fields. The rice root knot nematode (*Meloidogyne graminicola*) and root lesion nematode (*Pratylenchus* spp.) were found in the Belwa conventional tillage field. A few *Pratylenchus* spp. were found in the rice roots and a few white tip nematodes (*Aphelenchoides besseyi*) were in the wheat.

A few of the following root nematodes were extracted from the wheat roots (Table 3):

- *Hirschmanniella* spp. from minimal tillage fields A1 and C1;
- *Aphelenchoides* spp. from minimal tillage fields A1 and B1; and
- *Meloidogyne graminicola* from both Belwa sites.

Hirschmanniella spp. and *Meloidogyne graminicola* were found on both rice and wheat roots. Their presence in low numbers in wheat is indicating survival of this nematode in the wheat season and its carry over into the rice crop.

Table 2. Root damage [1] in rice and wheat crops at minimum (MT) and conventionally tilled (CT) survey sites

	Survey Sites [3]					
Crop/damage variable [2]	A1 (MT)	A2 (CT)	B1 (MT)	B2 (CT)	C1 (MT)	C2 (CT)
Rice, 2001						
RLI (0-3)	1.5	1.4	1.1	1.1	1.4	1 [4]
RKI (0-10)	0	0	0	0.4 [4]	0	0.2
Wheat, 2002						
RLI (0-3)	1	0.7	1.5	0.9 [4]	1.8	1.1 [4]
RKI (0-10)	0	0	1	1.1	0	0

[1] Mean of 10 observations
[2] RLI = Root lesion indexing (0-3) and RKI = root knot indexing (0-10)
[3] A, B, C = Santapur, Belwa and Benauli, respectively
[4] Significantly different at P = 0.05 (paired t-test)

Table 3. Nematodes extracted[1,2] roots of rice and wheat roots at minimum and conventionally tilled sites, Nepal

Crop	Survey site [3]	*Hirschmanniella oryzae, H. mucronata*	*Meloidogyne graminicola*	*Pratylenchus* spp. *(zeae?)*	*Aphelenchoides besseyi*
Rice 2001	A1	270	0	0	0
	A2	255	0	0	0
	B1	65	0	0	0
	B2	160	165	<10	0
	C1	25	0	0	<10
	C2	70	0	0	<10
	Survey site	*Hirschmanniella* spp.	*Meloidogyne graminicola*	*Pratylenchus* spp.	*Aphelenchoides* spp.
Wheat 2002	A1	<10	0	0	<10
	A2	0	0	0	0
	B1	0	10	0	<10
	B2	0	10	0	0
	C1	20	0	0	0
	C2	0	0	0	0

¹ = Nematodes per 2 gram of root sample
² = mean of two sub-samples
³ A, B, C = Santapur, Belwa and Benauli, respectively; 1 = Minimum tillage , 2 = Conventional Tillage

Soil nematodes and trophic groups

Rice crop

Among the different phytoparasitic nematodes only *Hirschmanniella oryzae* and *H. mucronata* were found in the rice soil. The population of this nematode was higher in the B1 and C1 minimal tillage fields and less in the A1 minimal tillage field. But most were found in the A2 conventional tillage field samples with a count of 1850/litre of soil (Table 4).

Table 4 Phytoparasitic nematodes[1] extracted from soil of minimum and conventionally tilled field in rice and wheat, Nepal

Rice 2001	**Survey sites[2]**					
	A1 [2]	**A2**	**B1**	**B2**	**C1**	**C2**
Hirschmanniella oryzae and *H. mucronata*	1250	1850	300	100	200	50
Meloidogyne graminicola	0	0	0	0	0	0
Tylenchorynchus spp.	0	0	0	0	0	0
Wheat 2002	**Survey sites[2]**					
	A1	**A2**	**B1**	**B2**	**C1**	**C2**
Hirschmanniella oryzae and *H.mucronata*	900	2400	650	100	400	200
Meloidogyne graminicola	0	0	350	100	0	0
Tylenchorynchus spp.	0	0	0	100	0	0

¹ = Nematodes per litre of soil
² A, B, C = Santapur, Belwa and Benauli, respectively; 1 = Minimum tillage , 2 = Conventional Tillage

Phytoparasitic nematodes occurred most in the rice soil, followed by bacterial feeders and then omnivorous nematodes (Table 5). The highest numbers of nematodes were found in conventional tillage fields A2 and C2. In contrast to this Yates and Bird (1994) reported more nematode numbers under no tillage treatments. In the current study the highest proportion of PPN were in the A1 (86.2% of site nematodes) and C1 (50%) minimal tillage fields. Bacterial feeding nematodes were found more in the conventional tillage A2 (28.3%) and C2 (47.1%) fields (the dominance of bacterial feeders in tilled fields has also been reported by Andren and Lagerlof, 1983). Omnivorous nematodes were also more abundant in the conventional tillage B2 (33.3%) and C2 (47.1%) fields. This indicates that there were more saprophytic nematodes in the conventionally tilled fields than the minimally tilled fields.

Wheat crop

Tylenchorhynchus spp., *Meloidogyne graminicola* and *Hirschmanniella oryzae/mucronata* were recovered from the wheat soil with the following trends (Table 4):

- *Tylenchorhynchus* spp. were only present in the Belwa conventional tillage field;
- *Meloidogyne graminicola* was also only found at the Belwa site;
- minimal tillage B1 field had over three times more *M. graminicola* (350/litre of soil) than the conventional tillage B2 field (100/litre of soil); and
- the highest number of *Hirschmanniella oryzae/mucronata* were recovered from the B1 and C1 minimal tillage fields and the A2 conventional tillage field.

Table 5 Nematode trophic groups extracted[1] from rice and wheat soil in minimum and conventional tilled fields, Nepal

Rice 2001	**Survey sites[2]**											
Trophic groups	**A1**		**A2**		**B1**		**B2**		**C1**		**C2**	
	N	%	N	%	N	%	N	%	N	%	N	%
Phytoparasites (not *Tylenchus*)	1250	86.2	1850	69.8	300	30	100	33.3	200	50	50	5.9
Bacterial feeders	100	6.9	750	28.3	500	50	100	33.3	100	25	400	47.1
Fungivores (inc. root hair feeders)	0	0	0	0	0	0	0	0	0	0	0	0
Omnivores	100	6.9	150	5.7	200	20	100	33.3	100	25	400	47.1
Total	1450	100	2650	100	1000	100	300	100	400	100	850	100
Wheat 2002	**Survey sites**											
Trophic groups	**A1**		**A2**		**B1**		**B2**		**C1**		**C2**	
	N	%	N	%	N	%	N	%	N	%	N	%
Phytoparasites (not *Tylenchus*)	900	34	2400	46.6	1000	11.7	300	3.7	400	5	200	3.3
Bacterial feeders	800	30.2	1150	22.3	3750	58.5	4800	58.5	1000	12.5	4200	70
Fungivores (inc. root hair feeders)	650	24.5	1050	20.4	1300	15.2	900	11	6400	80	1400	23.3
Omnivores	300	11.3	550	10.7	2500	29.2	2200	26.8	200	2.5	200	3.3
Total	2650	100	5150	100	8550	100	8200	100	8000	100	6000	100

[1] = Nematodes per litre of soil and as a percentage of total
[2] A, B, C = Santapur, Belwa and Benauli, respectively; 1 = Minimum tillage , 2 = Conventional Tillage

The bacterial feeders were the most commonly occurring type of nematode in wheat, found in much greater quantities than the PPN and omnivorous nematodes (Table 5). The level of fungivorous nematodes in the current study's minimal tillage fields was the same as Gupta and Vliet's (1996) findings. The higher number of nematodes collected from the minimal tillage samples is similar to the findings of Yates and Bird (1994).

CONCLUSION

The effect of minimal tillage and conventional tillage practices on the number of root lesions was observed only from the conventional tillage Benauli site in the rice season. In wheat there was no effect of tillage on the amount of root lesion. Root knot in rice was observed only in the Belwa and Benauli conventional tillage fields. More root knot infestation was observed at the Belwa sites in the wheat than in the rice, but there was no variation according to tillage type. *Hirschmanniella mucronata*, *H. oryzae* and *Meloidogyne graminicola* were found in both rice and wheat and may therefore be a nematode pest in rice-wheat systems. The fact that there was slightly more plant parasitic nematodes in the minimal tillage than the conventional tillage fields might be due to a carry over effect of undisturbed root and rhizospheric soil from the rice to the wheat crop. More free-living nematodes were found in the conventional tillage than the minimal tillage fields in both rice and wheat. This might be due to the incorporation of plant residues into the soil during tillage.

Overall this study shows a variable response in the occurrence of nematodes to the two kinds of tillage practices. Some of the results conflict with the findings of previous studies. This may be due to differences in soil ecology or limitations in this study's data (data was only collected for one year). More studies are needed in different soil environments and time periods to present a more definitive picture of the relation of nematode occurrence to tillage practices.

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Effect of Zero-Tillage on Soil Nematodes under Rice-Wheat Cropping in Haryana State, India

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ABSTRACT

The study presented in this paper was carried out in two sets of fields at Uchana, Karnal district and one set of fields in Teek, Kaithal district in Harayan. These sites had been under zero-tillage for the previous two to four years. The plant parasites *Hirschmanniella oryzae* (rice-root nematode) and *Tylenchorhynchus mashoodi* (stunt nematode) were found at all study sites. The present study, found a definite trend for *T. mashoodi*, with the samples from the conventional tillage fields having significantly higher populations than the zero tillage samples. Also, the mean population of *T. mashoodi* in the March dough stage samples was significantly more than in the November crown root initiation stage samples. These findings were corroborated by the trend in the population of *H. oryzae* over the same period.

INTRODUCTION

Minimum tillage was quite a common practice in early agriculture; but its use was greatly reduced with the increasing mechanisation of agricultural operations. However, it is now becoming popular again in parts of South Asia including in the Indo-Gangetic plains and in Haryana state, Indiain particular. This practice has a number of benefits including increased crop yields, and fuel saving (Mehla *et al.,* 2000). Its adoption is leading to more sustainable and environmentally-friendly food production.

Zero tillage in wheat has a direct impact on soil's physico-chemical properties. It has implications for the types of soil life that influence soil health for crop growth. Little is known about the effect of zero tillage on soil productivity and its long term effects on soil biota especially soil borne pathogens, insects, and nematodes. One of the most important soil-borne biotic factors are the nematode trophic groups. This study looks at the impact of growing wheat under zero tillage on the different trophic groups of nematodes. It was carried out in farmers' fields where zero tillage had been followed for between two and four years.

MATERIALS AND METHODS

The study was carried out in two sets of fields at Uchana, Karnal district (Karnal-I and Karnal-II) and one set of fields in Teek, Kaithal district. These sites had been under zero-tillage for the previous two to four years.

Soil and root samples were collected twice during the *kharif* rice growing season and three times during the *rabi* wheat growing seasons in two districts of Haryana state. The samples were drawn at the crown root initiation (CRI) stage in November and the dough stage in March during the wheat crop, and from the tillering (July) and flowering (September) stages of the rice crop. Soil samples (100cc) and root samples (2g) were analysed for the presence of nematodes. The soil samples were processed using Cobb's sieving and decanting method (Cobb, 1918) and the modified Baermann funnel technique; and roots were processed using the Waring blendor technique. The results are presented in Tables 1 to 12.

RESULTS AND DISCUSSION

The data for the plant parasitic nematodes in the wheat fields (Table 1) shows that:

- the population of plant parasitic nematodes at the CRI (November) stage was slightly higher in zero tilled than conventionally tilled fields in 1999/2000 and 2000/00;
- all the conventional tillage fields had more nematodes than in the zero tillage fields at the dough stage;
- the Karnal-I site had the highest number of nematodes at both sampling stages and in both tillage types with a significantly higher population of nematodes in the conventional tillage fields at the dough stage;

The data for the rice fields (Table 2) shows twice as many nematodes in the conventional tillage fields compared to the zero tillage fields at the tillering stage and more in the zero tillage fields at the flowering stage. Also, more nematodes were recorded in the root samples from the conventional tillage fields at the rice flowering stage.

Table 1 Population counts of plant parasitic nematodes in zero tillage (ZT) versus conventionally tilled (CT) wheat

Farmer (Location)	Sampling timing and tillage practice [2]					
	CRI [1] Stage (November)			Dough Stage (March)		
	ZT	CT	Mean	ZT	CT	Mean
Dharamvir (Karnal-I)	398.2 [2]	369.5	**383.8**	569.0	938.4	**753.7**
Naresh (Karnal-II)	85.6	110.0	**97.8**	402.6	450.0	**426.3**
Nakali (Kaithal)	67.0	53.5	**60.2**	124.6	137.2	**130.9**
Mean [3]	**183.6**	**177.6**		**365.4**	**508.5**	

[1] CRI: Crown Root Initiation
[2] Mean of 1999-2000 and 2000-01 periods from counting occurrence in 100 cc of soil
[3] C.D. (Critical Difference) for sampling time: 6.3; C.D. for tillage practice: 8.9; C.D. for location: 10.9

Table 2 Population of plant parasitic nematodes in zero (ZT) and conventionally tilled (CT) rice

Farmer (Location)	Sampling Timing/Tillage Practice [2]					
	Tillering Stage (July)			Flowering Stage (September)		
	ZT	CT	Mean	ZT	CT	Mean
Dharamvir (Karnal-I)	85.5 [2]	139.0	112.25	480.2 (17.5) [1]	149.5 (20.0)*	**314.85 (18.75)**
Naresh (Karnal-II)	172.2	385.5	278.85	146.3 (6.0)	475.6 (40.5)	**310.95 (23.25)**
Nakali (Kaithal)	65.0	37.9	51.45	326.5 (20)	111.2 (46.5)	**218.85 (33.25)**
Mean [3]	**79.0**	**187.4**		**317.6 (14.5)**	**245.7 (35.6)**	

[1] Figures in parenthesis are nematode population in 2g root
[2] Mean of 1999-2000 and 2000-01 periods from counting occurrence in 100 cc of soil
[3] C.D. (Critical Difference) for sampling time: 5.0; C.D. for tillage practice: 7.1; C.D. for location: 8.7

When the data was disaggregated by species of nematode it was found that the rice-root nematode *Hirschmanniella oryzae*, which causes disease in rice, occurred more in conventional tillage than zero tillage fields at both stages of sampling during the *rabi* wheat season (Table 3). However, during the 2001-02 *rabi* season (March, dough stage of wheat) the conventional tillage fields had more rice-root nematodes than the zero tillage fields in all three sites (Figure 1). However, the population in the soil of rice-root nematodes was more in zero tillage than conventional tillage fields at both stages of *kharif* season crop growth. At the flowering stage of rice the conventional tillage fields harboured more root nematodes than the zero tillage fields (Table 4).

Table 3 Population of *Hirschmanniella oryzae* in zero (ZT) and conventionally tilled (CT) wheat

Farmers (Location)	Sampling Timing /Tillage Practice					
	CRI [1] Stage (November)			Dough Stage (March)		
	ZT	CT	Mean	ZT	CT	Mean
Dharamvir Karnal-I)	77.7 [2]	135.1	106.4	62.0	48.6	55.3
Naresh (Karnal-II)	17.1	55.5	36.3	15.2	37.5	26.3
Nakali (Kaithal)	56.5	41.4	48.9	42.3	82.5	62.4
Mean [3]	**50.4**	**77.3**		**39.8**	**56.2**	

[1] CRI: Crown Root Initiation
[2] Mean of 1999-2000 and 2000-01 periods from counting occurrence in 100 cc of soil
[3] C.D. (Critical Difference) for sampling time: 3.5; C.D. for tillage practice: 5.0; C.D. for location: 6.1

Table 4 Population of *Hirschmanniella oryzae* in zero (ZT) and conventionally tilled (CT) rice

Farmers (Location)	Sampling Timing/ Tillage Practice					
	Tillering Stage (July)			Flowering Stage (September)		
	ZT	CT	Mean	ZT	CT	Mean
Dharamvir (Karnal-I)	29.0 [2]	28.0	28.5	143.0 (6.0) [1]	50.2 (12.9)	96.6 (9.45)
Naresh (Karnal-II)	26.5	0	13.25	60.3 (1.1)	218.5 (5.5)	139.4 (3.3)
Nakali (Kaithal)	52.0	37.9	44.95	226.0 20.0)	101.6 (19.0)	163.8 (19.5)
Mean [3]	**35.8**	**21.9**	**-**	**143.1 (9.03)**	**123.4 (12.4)**	**-**

[1] Figures in parenthesis are nematode population in 2g root
[2] Mean of 1999-2000 and 2000-01 periods from counting occurrence in 100 cc of soil
[3] C.D. (Critical Difference) for sampling time: 3.4; C.D. for tillage practice: 4.9; C.D. for location: 6.0

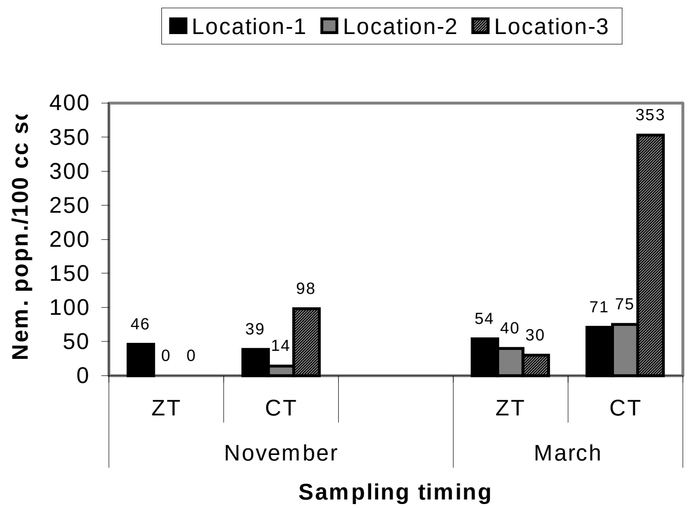

Figure 1. Occurrence of *H. oryzae* in zero tillage and conventional tillage fields (2001-2002); location 1 (Karnal-1), location 2 (Karnal-2), location 3 (Khaital)

The stunt nematode *Tylenchorhynchus mashoodi* showed no definite trend during the 1999-2000 and 2000-01 *rabi* seasons. However, during the 2001-02 season (Figure 2):

- more than twice as many were recorded from the conventional tillage fields than the zero tillage fields;
- they occurred slightly more in the zero tillage fields at the CRI stage of wheat (November) and more in conventionally tilled fields at the dough stage (Table 5);
- the zero tillage fields had slightly more than the conventional tillage fields at both tillering and the flowering stages of rice (Table 6); and
- more were recorded from the Karnal–I and Karnal-II sites than from the Kaithal sites.

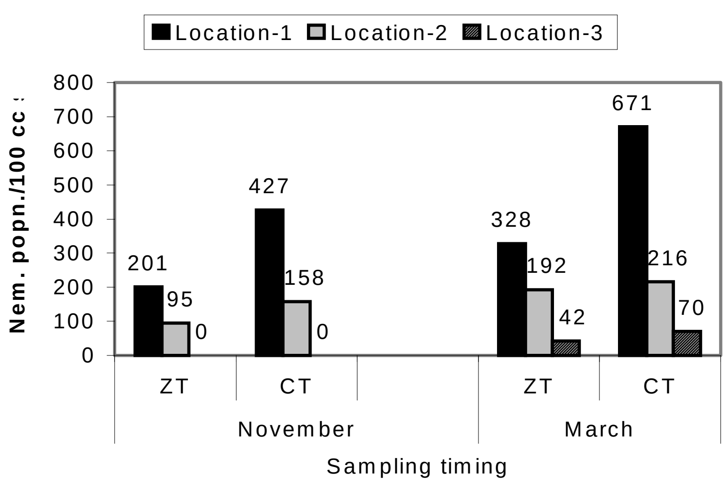

Figure 2 Occurrence of *T. mashoodi* nematode in zero tillage and conventional tillage fields (2001-2002); location 1 (Karnal-1), location 2 (Karnal-2), location 3 (Khaital)

Table 5 Population of *Tylenchorhynchus mashoodi* in zero and conventionally tilled wheat

Farmers (Location)	Sampling Timing /Tillage Practice					
	CRI [1] Stage (November)			Dough Stage (March)		
	ZT	CT	Mean	ZT	CT	Mean
Dharamvir Karnal-I)	320.5 [2]	247.0	283.7	407.0	705.0	556.0
Naresh (Karnal-II)	85.6	110.0	97.8	342.5	412.6	377.5
Nakali (Kaithal)	10.8	13.5	12.1	82.5	54.5	68.5
Mean [3]	**138.9**	**123.5**		**277.3**	**390.0**	

[1] CRI: Crown Root Initiation
[2] Mean of 1999-2000 and 2000-01 periods from counting occurrence in 100 cc of soil
[3] C.D. (Critical Difference) for sampling time: 4.5; C.D. for tillage practice: 6.4; C.D. for location: 7.9

Table 6 Population of *Tylenchorhynchus mashoodi* in zero (ZT) and conventionally tilled (CT) rice

Farmers (Location)	Sampling Timing/Tillage Practice					
	Tillering Stage (July)			Flowering Stage (September)		
	ZT	CT	Mean	ZT	CT	Mean
Dharamvir Karnal-I)	56.5 [1]	111.0	**83.7**	295.0	99.5	**197.2**
Naresh (Karnal-II)	103.7	26.2	**64.9**	86.0	287.0	**186.5**
Nakali (Kaithal)	13.0	0.0	**6.5**	44.0	17.0	**33**
Mean [2]	**57.7**	**45.7**		**143.3**	**134.5**	

[1] Mean of 1999-2000 and 2000-01 periods from counting occurrence in 100 cc of soil
[2] C.D. (Critical Difference) for sampling time: 4.2; C.D. for tillage practice: 5.9; C.D. for location: 7.2

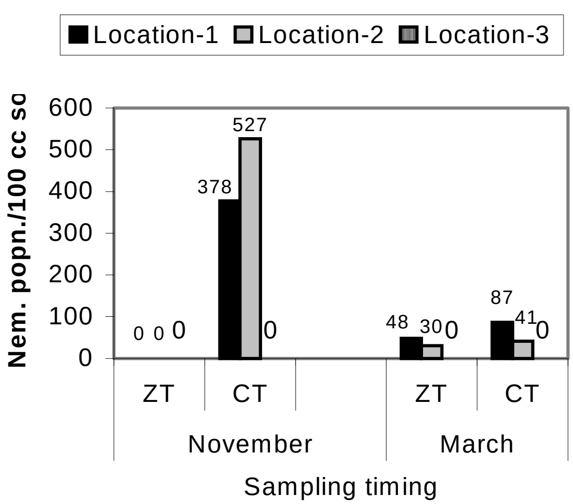

Figure 3 Occurrence of other parasitic nematodes in zero tillage and conventional tillage fields (2001-2002); location 1 (Kanal-1), location 2 (Karnal-2), location 3 (Khaital)

The study also measured the occurrence of other plant parasitic nematodes, including *Psilenchus, Tylenchus* sp., *Hoplolaimus, Helicotylenchus.* Only a few of these were found in the samples with more occurring at Karnal-I and Karnal-II in conventional tillage fields at the CRI and dough stages of wheat (Figure 3).

The population levels findings of non-plant parasitic nematodes are presented in Tables 7 to 12. During the *rabi* wheat season the population of non-plant parasitic nematodes was more in the zero tillage than the conventional tillage fields at both CRI and dough stages (Table 7). During the *kharif* rice season at both stages non-plant parasitic nematodes were found more in conventional tillage fields (Table 8). When the data was separated out by trophic group for the saprophagous nematodes (nematodes nourished on dead or decaying animal matter), it was observed that Dorylaimids occurred more in zero tillage than in conventional tillage fields at both the flowering and dough stages at Karnal-II and Kaithal (Table 9). But, the average population was significant different at both sampling stages. Cephalobids were significantly fewer in zero tillage than in conventional tillage fields in both the September and March *kharif* season samples (Table 10). The data shows that:

- overall there were slightly more Araeolaimid nematodes in zero tilled fields at the flowering stage of rice and the dough stage of wheat (Table 11);
- the Karnal-I site had the most Araeolaimids and Kaithal the least;
- more Alaimids were recorded under conventional tillage in the March samplings and under zero tillage in the September samplings (Table 12).

Similar studies have given differing results. Dabur (2001) observed no definite trends in respect of plant parasitic and saprophagous nematodes related to different tillage practices. But Dabur *et al.* (2002) reported that *H. oryzae* populations were higher in zero tillage than conventional tillage fields at both tillering and flowering stages of rice. The same study showed that the population of stunt nematodes was not significantly different between the two practices.

Table 7 Population of non-plant parasitic nematodes in zero (ZT) and conventionally tilled (CT) wheat

Farmers (Location)	Sampling Timing /Tillage Practice					
	CRI [1] Stage (November)			Dough Stage (March)		
	ZT	CT	Mean	ZT	CT	Mean
Dharamvir Karnal-I)	666.5 [2]	400.0	533.2	542.2	482.0	512.1
Naresh (Karnal-II)	245.6	186.2	215.9	1023.0	798.9	910.9
Nakali (Kaithal)	151.7	109.1	130.4	681.5	567.1	624.3
Mean [3]	**354.6**	**231.7**		**748.9**	**616.0**	

[1] CRI: Crown Root Initiation
[2] Mean of 1999-2000 and 2000-01 periods from counting occurrence in 100 cc of soil
[3] C.D. (Critical Difference) for sampling time: 7.5; C.D. for tillage practice: 10.6; C.D. for location: 12.9

Table 8 Population of non-plant parasitic nematodes in zero (ZT) and conventionally tilled (CT) rice

Farmers (location)	Sampling Timing/Tillage Practice					
	Tillering Stage (July)			Flowering Stage (September)		
	ZT	CT	Mean	ZT	CT	Mean
Dharamvir (Karnal-I)	366.0 [1]	369.5	**367.7**	1463.5	3083.5	**2273.5**
Naresh (Karnal-II)	186.7	286.5	**236.6**	576.0	983.0	**779.5**
Nakali (Kaithal)	130.0	184.0	**157.0**	459.0	488.5	**473.7**
Mean [2]	**227.5**	**280.0**		**832.8**	**1518.3**	

[1] Mean of 1999-2000 and 2000-01 periods from counting occurrence in 100 cc of soil
[2] C.D. (Critical Difference) for sampling time: 10.0; C.D. for tillage practice: 14.2; C.D. for location: 17.4

Table 9 Population of Dorylaimids in zero (ZT) and conventionally tilled (CT) rice fields

Farmers (Location)	Sampling Timing /Tillage Practice					
	Flowering Stage (September)			Dough Stage (March)		
	ZT	CT	Mean	ZT	CT	Mean
Dharamvir Karnal-I)	242.6	560.0	**401.3**	436.3	385.7	**411.0**
Naresh (Karnal-II)	932.6	543.0	**787.8**	292.0	166.2	**229.1**
Nakali (Kaithal)	227.0	184.6	**205.8**	216.6	177.3	**196.9**
Mean	**467.4**	**462.5**	**465.1**	**314.9**	**243.0**	**279.0**

[1] Mean of 1999-2000 and 2000-01 periods from counting occurrence in 100 cc of soil
[2] C.D. (Critical Difference) for sampling time: 4.8; C.D. for tillage practice: 6.8; C.D. for location: 8.3

Table 10 Population of Cephalobids in zero (ZT) and conventionally tilled (CT) rice fields

Farmers (Location)	Sampling Timing/Tillage Practice					
	Flowering Stage (September)			Dough Stage (March)		
	ZT	CT	Mean	ZT	CT	Mean
Dharamvir (Karnal-I)	97.7 [1]	119.7	**108.7**	15.3	138.3	**76.8**
Naresh (Karnal-II)	170.7	281.8	**226.2**	215.8	360.3	**288.0**
Nakali (Kaithal)	96.7	144.2	**120.4**	162.0	209.7	**184.8**
Mean [2]	**121.7**	**181.9**	**152.02**	**131.0**	**236.1**	**183.6**

[1] Mean of 1999-2000 and 2000-01 periods from counting occurrence in 100 cc of soil
[2] C.D. (Critical Difference) for sampling time: 5.8; C.D. for tillage practice: 8.2; C.D. for location: 9.9

Table 11 Population of Araeolaimids in zero (ZT) and conventionally tilled (CT) rice fields

Farmer (Location)	Sampling Timing /Tillage Practice					
	Flowering Stage (September)			Dough Stage (March)		
	ZT	CT	Mean	ZT	CT	Mean
Dharamvir (Karnal-I)	67.9 [1]	114.7	**91.00**	299.0	209.0	**254.0**
Naresh (Karnal-II)	114.0	10.00	**62.00**	230.0	218.0	**224.0**
Nakali (Kaithal)	24.0	17.0	**20.0**	133.0	157.0	**145.0**
Mean [2]	**68.7**	**47.0**	**57.8**	**220.7**	**194.7**	**207.7**

[1] Mean of 1999-2000 and 2000-01 periods from counting occurrence in 100 cc of soil
[2] C.D. (Critical Difference) for sampling time: 3.4; C.D. for tillage practice: 4.8; C.D. for location: 5.9

Table 12 Population of Alaimids in zero (ZT) and conventionally tilled (CT) rice fields

Farmers (Location)	Sampling Timing/Tillage Practice					
	Flowering Stage (September)			Dough Stage (March)		
	ZT	CT	Mean	ZT	CT	Mean
Dharamvir (Karnal-I)	5.03 [1]	1.0	**3.017**	1.00	1.00	**1.00**
Naresh (Karnal-II)	7.47	11.22	**9.34**	1.00	4.75	**2.87**
Nakali (Kaithal)	6.31	1.00	**3.65**	1.00	3.16	**2.08**
Mean	**6.27**	**4.40**	**5.34**	**1.00**	**2.98**	**1.98**

[1] Mean of 1999-2000 and 2000-01 periods from counting occurrence in 100 cc of soil
[2] C.D. (Critical Difference) for sampling time: 0.13; C.D. for tillage practice: 0.18; C.D. for location: 0.22

The present study, found a definite trend for *T. mashoodi*, with the samples from the conventional tillage fields having significantly higher populations than the zero tillage samples (Figure 2). Also, the mean population of *T. mashoodi* in the March dough stage samples was significantly more than in the November CRI stage samples. These findings were corroborated by the trend in the population of *H. oryzae* over the same period (Figure 1). Although wheat is a direct host of *H. oryzae*, its much higher population in March compared to November indicates that either it survived on some alternative host such as weeds, or that the recovery/growth of this nematode's population was more in the period up to March due to more activity or the hatching of eggs.

The differences in the levels of nematode population could, however, be due to the different tillage practices. Garg *et al*. (1995) have demonstrated how tillage practices alter soil structure and induce changes in soil water retention, aeration and thermal gradient which will influence biological activities. Gaur and Singh (2000) reported increases in the populations of *Meloidogyne* spp. and *Tylenchorhynchus* spp. nematodes when zero tillage replaced conventional tillage for growing wheat. However, the authors give no explanation for this. However, the plot size taken by Gaur and Singh (2000) was only $100 m^2$ whereas plot size in the present study is $4000 m^2$ which could account for the difference in nematode populations. Similar increases in nematode populations in zero tillage plots have been reported by Dabur (2001). However, the fact that his study reports no definite trends in nematode populations suggests that this could have been due to different sowing and transplanting dates at the different locations. The latter factors could have masked the similarity in population levels. Another important factor in the Dabur study was that the initial levels of nematodes were not the same in all the study fields.

CONCLUSIONS

The plant parasites *Hirschmanniella oryzae* (rice-root nematode) and *Tylenchorhynchus mashoodi* (stunt nematode) were found at all study sites. The study also found the plant parasitic nematodes *Helicotylenchus* sp., *Hoplolaimus* sp., *Basiria* sp. and *Tylenchus* sp. The study also recorded:

- more *H. oryzae* and *T. mashooddi* from the conventional tillage than the zero tillage fields at the CRI and dough stages of the *rabi* wheat crop;
- less nematodes from the conventional tillage fields during the *kharif* rice season at both stages;

- most *T. mashoodi* at the Karnal I site during *rabi* season, whilst *H. oryzae* showed no trend by location;
- more saprophagous nematodes in zero tillage fields during the *rabi* season at all three locations and in the conventional tillage fields during the *kharif* season;
- more Dorylaimids and Araeolaimids in zero tillage than conventional fields in the September and March samplings;
- more Cephalobids in conventional than zero tillage fields; and
- more Dorylaimids and Alaimids were found, irrespective of tillage practice, in the September compared to the March samplings and more Cephalobids and Araeolaimids in the March than the September samplings.

FUTURE RESEARCH

Further multidisciplinary studies need carrying out on:

- nematode populations, particularly at the peak growing periods of September for rice and March for wheat;
- on the Dorylaimids, Araeolaimids, and Cephalobid nematode groups to investigate their role as bioindicators; their population dynamics in relation to tillage practices, crop type, crop season, interaction with other microfauna; and their role in crop productivity and soil ecology; and
- the trend of rice root nematode populations during wheat seasons.

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Nematodes in Rice-Wheat Cropping under Resource Conservation Technologies in Pakistan

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ABSTRACT

This paper describes the methodology followed to extract and assess the importance of nematodes in rice-wheat cropping under resource conservation technologies in Pakistan. Although there were no definite trends, the average populations indicate that the nematode population was more in the rice than in wheat and that there was no carry over effect from the zero till wheat crop into the following rice crop. The overall data for the pathogenic and saprophytic nematodes in both crops also shows that the wheat crop was not affected by the higher population of these nematodes in the rice crops. The rice root knot nematode, *Meloidogyne graminicola*, was found for the first time in Pakistan but galling was only recorded at two rice sites. The results indicate no significant difference in nematode population between crops or tillage regimes. The three years of data does not support the theory that the nematode population increases in zero tillage fields; rather the nematode population was low in the wheat crop and there was no observable effect of using zero tillage.

INTRODUCTION

Rice-wheat cropping is carried out over almost two million hectares in Pakistan and a large proportion of the population earns their livelihood from it. This rotation of rice and wheat has been practised for a long time in this area. Recent water crises and the use of later maturing varieties of rice have forced researchers to look into conservation technologies that allow for the more efficient use of resources. The current study looked at the effect of zero or minimum tillage on soil-borne microflora. By using zero tillage farmers save the cost of preparing the land and can sow the wheat crop about 15 days earlier. But the concern is that this technique may lead to the build up of soil borne pathogens in the rice stubble. This paper investigates the development of nematode populations in soil under zero and conventional tillage fields.

Nematodes are a major pest of rice and wheat in the rice-wheat cropping systems of the Indo-Gangetic plains. Recent recommendations from the Rice-Wheat Consortium Workshop stated the urgent need to i) characterize the nematode fauna in rice-wheat systems, ii) identify and prioritize nematode induced constraints in rice-wheat systems, and iii) determine the relationships with other soil biota, ecological, and edaphic factors.

Plant pathogenic nematodes separately reported from rice and wheat in Pakistan include *Hirschmanniella* (Khan and Bilqees, 1994), *Ditylenchus* (Gul and Saifullah, 1991), *Helicotylenchus* and *Tylenchorhynchus* (Khan and Shaukat, 1999), *Heterodera* (Maqbool, 1981), *Pratylenchus* and *Xiphinema* sp. (Akhtar, 1962). The current study measures their build up and characterizes the free living nematode fauna in rice-wheat cropping under the different tillage regimes. The rice-wheat growing areas of Muridke and Mona were selected for the study (Table 1).

Initially four farmers were selected for study at each site; but later it was found that available resources only allowed the study to go ahead with two farmers at each site. Farmers 1 and 3 were selected at Muridke and farmers 5 and 6 at Mona.

MATERIALS AND METHODS

The study took place over three years. Samples were collected at the booting stage of wheat (March) and the flowering stage of rice (September). CABI Bioscience developed the protocols for this project to characterise nematodes along with a key for identifying the major nematode groups (Hunt, 2000). Sampling was done in line with the proposed protocols (Bridge *et al.,* 1999). Samples were taken at 10 points, five paces apart along a diagonal transit starting 10 paces into the field. At each sampling site the nearest plant was dug out, to include its tillers, roots and soil, down to a depth of 20 cm. Soil adhering to the roots was left intact. The plant portion and soil were placed in a polythene bag and taken to the laboratory for further analysis.

Extraction of nematodes from soil

A portion of soil was removed from around the roots of each plant. These samples from each site were mixed together to provide a combined bulk sample. Two soil sub-samples of $100cm^3$ each were taken from these bulked samples to extract nematodes. The volumes of sub-samples were measured by the displacement of water in a graduated cylinder. The cylinder was partly filled with water up to a known volume (e.g. 200ml) and soil was added until the water volume increased by 100ml (e.g. up to 300 ml). The soil and water mixture was shaken and washed out into a container of approximately 5 litres. More water was added and the whole suspension thoroughly stirred and left to settle for 30 seconds. The suspension water from the large container was decanted onto a bank of three sieves with aperture sizes of 1.7mm (10 mesh), 90µ (170 mesh), and 38µ (400 mesh).

Table 1 Selected sites and farmers surveyed

Site	Farmer	Farm number	Field number	System
Muridke	Rizwan	1	1	Zero tillage
Muridke	Rizwan	1	2	Conventional
Muridke	Shahbaz	3	5	Zero tillage
Muridke	Shahbaz	3	6	Conventional
Mona (Bhalwal)	Asif	5	9	Zero tillage
Mona (Bhalwal)	Asif	5	10	Conventional
Mona (Bhalwal)	Faiz	6	11	Zero tillage
Mona (Bhalwal)	Faiz	6	12	Conventional

Water was again added to the container and the nematode suspension stirred and again allowed to settle for 30 seconds and poured through the sieves (three times in all). The nematodes were collected on the bottom sieves (90µ and 38µ) and washed off into a beaker each time after the suspension was decanted onto sieves using a spray or jet of water initially from the back of the sieves. The nematode suspension in the beaker was then shaken and poured onto tissue paper on a sieve in a small container. Additional water was added if needed to just cover the tissue. This final extraction was left undisturbed for 24 hours. Then the sieve and tissue were removed. The tissue with debris was carefully taken from the sieve, dried and examined for cysts. If present, cysts were picked off and counted.

The nematode suspension in the small container was stirred, poured into a beaker and allowed to settle for three hours. Excess top water was carefully decanted or siphoned off leaving approximately 10-20 ml behind. This remaining concentrated suspension was poured into a sample bottle and water carefully added to make up to a known volume such as 20ml. The nematodes were heat relaxed (killed) by immersing the sample bottles in hot water just off the boil for 2-3 minutes. The suspension was allowed to cool before the nematodes were fixed. Nematodes in the sample bottle were fixed by adding 4% formalin or double strength TAF (a preservative) to the cooled suspension.

Nematodes were identified and counted by taking an aliquot from the suspension, normally 10% (i.e. 2 ml from a 20 ml final volume), pipetted into a counting dish such as a small plastic petri dish with lines scored on the base of the dish at a width equal to the highest magnification on the stereoscope.

Extraction of nematodes from roots

The sample roots were washed clean of remaining soil. Portions of the roots from each of the 10 plants were randomly cut and removed and mixed together. Small samples were then collected for preservation and later staining, placed in sample bottles, and 4% formalin or 70% alcohol added. Part of the preserved root sample was sent to CABI UK for further examination. The remaining collection of mixed roots was damp dried on tissue paper before being cut into small pieces of 0.5-1 cm and again mixed together to give a single bulked sample. Two sub-samples of 2g each were randomly picked and weighed from this mixed and bulked sample of damp-dried roots. Each of these 2g root samples was cut into smaller pieces with fine scissors and placed on a tissue on a sieve in a small container and just covered with water for 48 hours. The tissue, roots and sieve were then removed and the nematodes counted as for those extracted from the soil samples.

RESULTS

Examination of both rice and wheat roots revealed infestation of the root-knot nematode *Meloidogyne graminicola.* This is the first recording from Pakistan. Other isolated parasitic nematodes were *Aphelenchoides, Hirschmanniella, Pratylenchus, Tylenchorhynchus* and *Tylenchus* spp. The saprophytic nematodes *Dorylaimus, Cephalobus, Mesorhabditis* and *Plectus* sp. were identified from both the rice and wheat fields. Although there were no definite trends, the average populations indicate that the nematode population was more in the rice than in wheat and that there was no carry over effect from the zero till wheat crop into the following rice crop (Figure 1). The overall data for the pathogenic and saprophytic nematodes in both crops also shows that the wheat crop was not affected by the higher population of these nematodes in the rice crops (Figure 2). Root-knot nematode galling was only recorded at two rice sites.

The results indicate no significant difference in nematode population between crops or tillage regimes. The three years of data does not support the theory that the nematode population increases in zero tillage fields; rather the nematode population was low in the wheat crop and there was no observable effect of using zero tillage. This study was, however, too limited in scope to come to definitive conclusions. Further studies of trophic groups and their relationships with cropping pattern and resource conservation technologies need carrying out. Further investigations on the role of entomopathogenic nematode and fungi will be of great interest concerning the balance of soil microflora and fauna. The role of soil borne microflora and microfauna in the control of rice stem borer also needs studying.

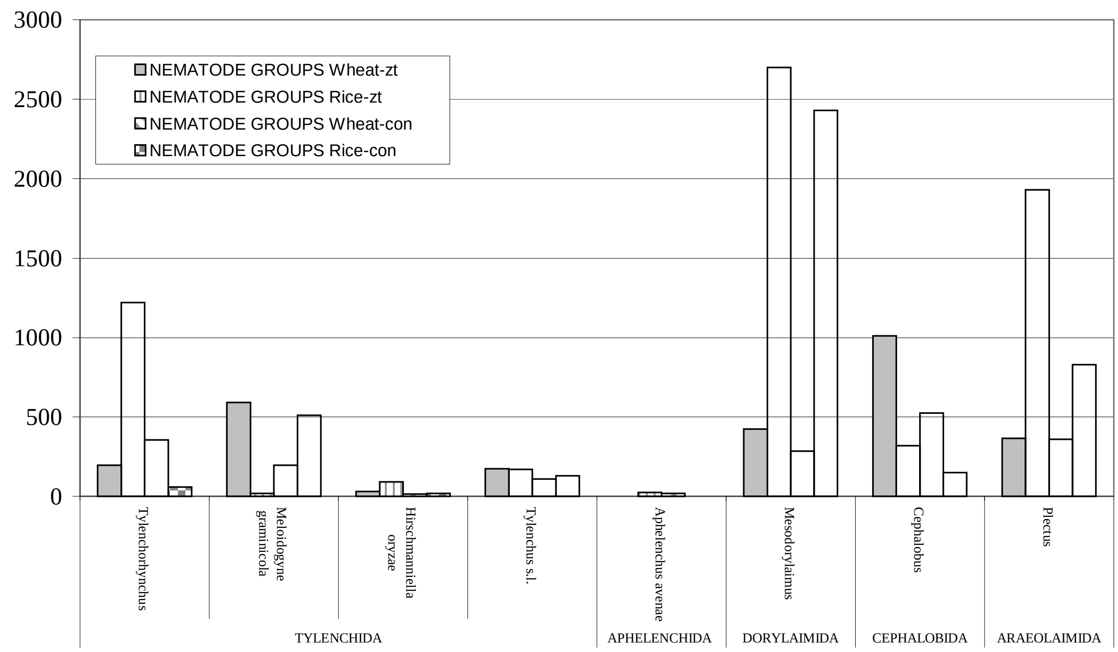

Figure 1. Average population of different trophic groups isolated from rice-wheat system under resource conservation technology (zt: zero tillage; con: conservation tillage)

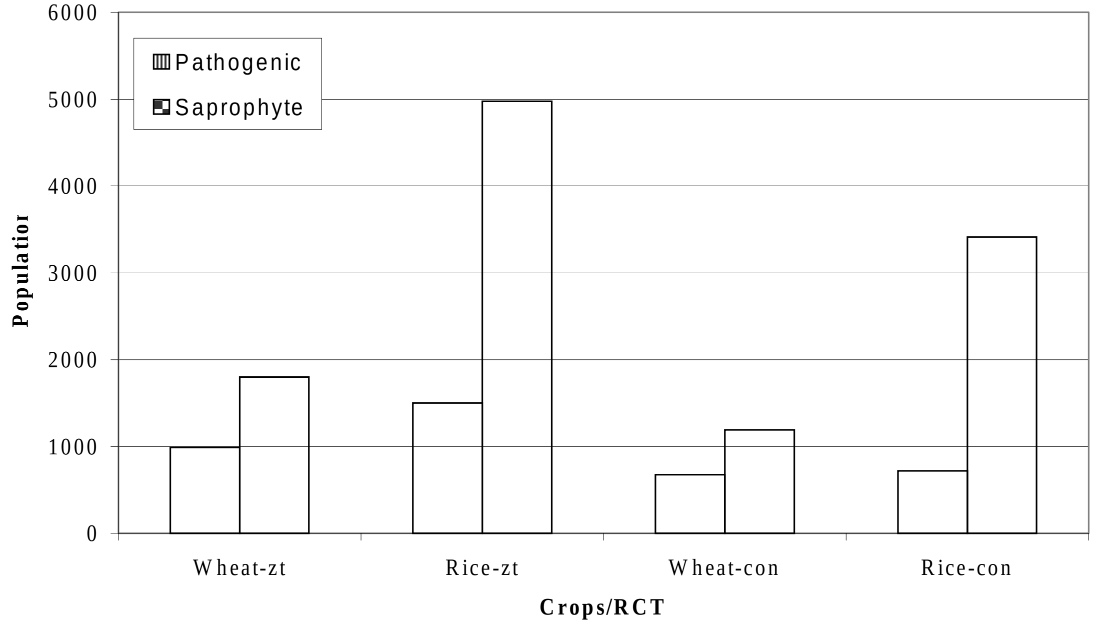

Figure 2. Average nematode population in wheat and rice crops in different resource conservation technologies (zt: zero tillage; con: conservation tillage)

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Rice-Wheat Consortium for the Indo-Gangetic Plains

The Consortium is an Ecoregional Program of the Consultative Group on International Agricultural Research (CGIAR), managed by CIMMYT, involving the National Agricultural Research Systems, the International Agricultural Research Centers, and the Advanced Research Institutions. Its main objective is to promote research on issues that are fundamental to enhance the productivity and sustainability of rice-wheat cropping systems in South Asia.

These objectives are achieved through:

- Setting priorities for focused research on problems affecting many farmers.
- Promoting linkages among rice-wheat research specialists and other branches of research and extension.
- Encouraging interdisciplinary team approach to understand field problems and to find solutions.
- Fostering quality work and excellence among scientists.
- Enhancing the transfer of improved technologies to farmers through established institutional linkages.

Financial support for the Consortium's research agenda currently comes from many sources, including the Governments of Netherlands, New Zealand, Australia and the Department for International Development (DFID), the International Fund for Agricultural Development (IFAD), the United States Agency for International Development (USAID), the World Bank and the Asian Development Bank (ADB).

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