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Distribution of fungal flora with special reference to *Trichoderma* population in soil samples of Krishi Vigyan Kendra of zone I of Bihar

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Abstract
Fungus is the 2nd largest group in the world after insects. Their antiquity, longevity, ubiquitous distribution in the bio-sphere and their ability to adjust to a wide range of environmental condition had made them the most successful organisms. Enumeration of different physical, chemical and fungal distribution with special reference to population of *Trichoderma* species in soils of Krishi Vigyan Kendra (KVK) Birauli (Samastipur) of Zone I of Bihar was carried out in the present investigation. The fungal flora were isolated from the four soil samples which was collected from two cultivated field, one from orchard and one from vegetable plot of local farmer. Two soil samples of cultivated field was taken from the plot intercropped with turmeric (*Curcuma longa),* arvi (*Acacia cyanophylla*), ginger (*Zingiber officinale),* sole crop of dhaincha (*Sesbania bispinosa)* and one from litchi (*Litchi chinensis)* orchard and one from elephant foot yam or ol (*Amorphophallus campanulatus)* as vegetable soil of local farmer. Soil samples from the intercropped plot and litchi orchard and elephant foot yam or ol (*Amorphophallus campanulatus*) plot had sandy loam to silt loam texture. The pH of all soil samples was near to neutral or alkaline (7.9 to 8.1). Maximum fungal population was recorded in soil of litchi orchard (12.33 X 103 cfu/g soil). *Trichoderma* was recorded frequently in most of the soil samples. The species of *Trichoderma* viz., *Trichoderma harzianum* and *Trichoderma viride* were isolated from the soil samples of KVK, Birauli.

Keywords: KVK, soil factors, fungal diversity, *Trichoderma*.

Introduction
Soil microbial biodiversity is a fundamental element of biosphere. They are critical for the maintenance of soil function in both natural and managed agricultural soils because of their involvement in such processes like soil structure formation, decomposition of organic matter, cycling of carbon, nitrogen, phosphorus and sulphur. They stimulate plant growth and also involved in several biochemical transformation and mineralization activities in soil which directly affects the agriculture. These microorganisms may be fungi, bacteria, actinomycetes etc., and among these, fungi are most dominant group present in the soil.

Fungal diversity encompasses the variety of fungi occurring in nature from the ecosystem to the genetic level as a result of evolution. It includes number and species richness of the concerned taxa and genetic diversity. They play a key role in many essential processes such as organic matter decomposition and elemental release by mineralization. They also play a key role in nutrient cycling by regulating soil biological activity. Agriculture in twenty first century needs to be more productive, environmentally benign, robust in the face of climate change and socially beneficial. The present study was undertaken to study the fungal flora distribution in the soils of Krishi Vigyan Kendra of Birauli, Samastipur of Zone I of Bihar with special reference to *Trichoderma* species. The soil of this Zone is Alluvial which is rich in organic matter and harbours diverse micro-organism that offers several benefits to the crops. The fungal floras were isolated from the soil samples collected from cultivated field, orchard and vegetable plot of nearby local farmer. Certain fungi of peculiar of a particular vegetation type or soil texture and they sometimes acts as indicators of population e.g. *Penicillium*, *Fusarium Aspergillus* and antagonist *Trichoderma* fungi which have been widely used against number of phytopathogens especially soil borne fungi.

Studies on soil microorganism have been carried out by several workers in soils of different parts of India under cultivated field, orchard, forest, grassland and in different cropping system but distribution of fungal flora in agricultural soils of KVK of north Bihar with special reference to *Trichoderma* population is still have little attention. In future it will be imperative to harness beneficial microorganism to plants and bio control agents of plant pathogenic fungi. Keeping in view these facts the present investigation was undertaken

to enumerate fungal distribution in soils of KVK Birauli, Samastipur Bihar.

Materials and Methods

Collection of soil samples: Soils samples were collected from KVK Birauli. Four different soil samples were taken, two from cultivated plot, one from orchard and one from local farmer. V shaped pits were dug in the area to be sampled and one cm thick slice of soil from either side of V shaped pits were drawn from each site in a clean plastic bucket or polythene bags. Collected soil samples was brought in laboratory of Plant Pathology Department of Rajendra Agricultural University, Pusa, Samastipur and dried in shade on paper and after drying it was transferred to clean polythene bag for analysis.

Sampling schedule: Soil samples were collected thrice as per schedule given below:

1) 1st fortnight of November 2012.
2) 1st fortnight of January 2013.
3) 1st fortnight of March 2013.

Soil analysis

Soil analysis was done in the department of soil science of Rajendra Agricultural University, Pusa Samastipur. Soil samples were analysed for texture, pH and organic carbon content. Soil texture was determined by Rapid Feel Method (Prasad *et al.,* 2006) and chemical analysis as soil pH was done by common standered method. Organic carbon content of the soil was determined by Walkley's and Black's method (1934).

Isolation, quantitative estimation and identification of fungal population

Isolation of fungi form soil were carried out by dilution plate method (Warcup, 1950, 1960) [7-8] by using peptone dextrose rose Bengal agar medium and potato dextrose agar medium.

Quantitative estimation of fungal population was done by counting number of colonies that appeared on plate and multiplied by dilution factor to determined fungal population as colony forming units (cfu/g of soil).

Most of the fungi that appeared on medium were identified based on morphological and cultural characters. Two techniques were adopted: (i) Visual observations for colony characters on medium and (ii) By preparing slides using lactophenol or cotton blue as a mounting medium and observed under compound Olympous microscope. Identification was done based on morphological characters of vegetative and asexual reproductive structures of fungus reported in literature.

Percent distribution of dominating fungi and *Trichoderma* isolates:

The number of colonies per plate that appeared from 1 g of soil was calculated. The percentage contribution of each isolate was done by using following formula (Mahalingam *et al.,* 2012).

$$\% \text{ Distribution} = \frac{\text{Colony of particular fungus (CFU) in a sample}}{\text{Total fungal colonies in a sample}} \times 100$$

Morphological study of microflora

Slides were prepared from the 5 days old cultures of fungi and examined under microscope. Morphometric measurements were done with the help of stage and ocular micrometer.

Results and Discusssion

Soil is a complex, dynamic entity with its own physical chemical and biological properties. The biological population depends on physical and chemical properties of soil whereas these properties in turn are continuously modified by the activities of biological population (Kennedy and Papendick 1995).

Analysis of physical and chemical parameters in three different phases is given below in the (Table 1).

Table 1: Soil texture, pH and organic carbon content in soil samples of KVK, Birauli, Samastipur.

Soil sample	Soil texture	Phase I		Phase II		Phase II	
		pH	Organic carbon (%)	pH	Organic carbon (%)	pH	Organic carbon (%)
Intercrop (turmeric, arvi, ginger)	Sandy loam	8.0	0.89	7.9	0.42	8.1	0.93
Dhaincha	Silt loam	7.5	0.63	7.2	0.58	7.6	0.33
Litchi	Sandy loam	6.8	0.75	7.1	0.79	7.6	0.94
Ol	Silt loam	7.2	0.23	7.8	0.28	7.6	0.30

Analysis of fungal diversity with special reference to *Trichoderma* species were estimated in three different phases. It revealed that fungal population consequently increased in successive phases in intercropped soil, litchi and ol but in soil of dhaincha total fungal population slightly decreases from 9.66 $x10^3$ to 9.33 $x10^3$ during first to second phase, as showed in given data (Table 2) but it recognisably boosted in third phase of isolation. In the first phase all the sample showed low population but in third phase there was higher fungal population. The reason for higher fungal population in soil may be due to faster multiplication of organism at higher temperature which prevailed during third phase of isolation. Gauri Rane (2011) [1] also reported higher population of fungi during the study of diversity and seasonal occurrence of soil fungi from two forest soils of Maharashtra. Gulerii *et al.,* (2011) [2] while studying the distribution of fungi during different forest soils of Rajaji National Park had also reported similar result.

The population of *Trichoderm*a was higher in 2nd and 3rd phase of isolation as compared to first phase of isolation. Greater population of *Trichoderma* species in soils have been estimated in the spring and summer months as compared to autumn and winter (Widden and Abitbol, 1980) [9]. Maximum population of *Trichoderma* was found from litchi orchard. *Trichoderma* species prefer and grow well in soil having acidic pH and high organic matter (Upadhyay and Rai, 1978, 1979) [5].

Table 2: Estimation of fungal population in different soil samples of KVK, Birauli, Samastipur

Soil sample	Population of soil microflora X 10^3 cfu/g of soil*			Contribution (%)		Dominating fungus
	Total fungi	Dominating fungi	*Trichoderma* species	Dominating fungus	*Trichoderma* species	
First phase						
Intercrop	10.66	5.66	3.66	55.53 (46.92)**	34.54 (35.90)	*Fusarium*
Dhaincha	9.66	4.66	2.6	48.51 (44.13)	27.77 (31.67)	*Fusarium*
Litchi	9.33	5.0	2.6	53.33 (46.91)	28.51 (32.18)	*Fusarium*
Ol	12.66	7.3	2.0	57.90 (49.34)	15.80 (23.44)	*Fusarium*
CD at 5%	1.08	1.3	0.9	7.5	6.6	-
CV	5.4	12.4	18.18	8.5	11.5	-
Second phase						
Intercrop	11.00	4.3	4.3	39.39 (38.88)	39.39 (38.88)	*Trichoderma* spp.
Dhaincha	9.33	4.3	4.3	46.29 (42.85)	46.29 (42.85)	*Trichoderma* spp.
Litchi	12.33	10.00	10.0	81.19 (64.84)	81.19 (64.84)	*Trichoderma* spp.
Ol	13.00	11.3	11.3	87.72 (72.92)	87.72 (72.92)	*Trichoderma* spp.
CD at 5%	1.21	1.35	1.35	15.6	15.6	-
CV	5.6	9.3	9.3	15.1	15.1	-
Third phase						
Intercrop	20.00	12.33	12.33	61.77 (51.82)	61.77 (51.82)	*Trichoderma* spp.
Dhaincha	18.33	12.66	12.66	69.26 (56.35)	69.26 (56.35)	*Trichoderma* spp.
Litchi	17.33	14.00	14.0	80.71 (64.07)	80.71 (64.07)	*Trichoderma* spp.
Ol	20.33	11.66	11.66	57.38 (49.24)	57.38 (49.24)	*Trichoderma* spp.
CD at 5%	1.3	1.3	1.3	4.90	4.90	-
CV	5.5	5.5	5.5	6.90	6.90	-

*Mean of 3 replications; ** Values in parentheses are Arcsin √percentage transformation

In all the three different phases of isolation *Aspergillus, Penicillium, Fusarium, Alternaria, Trichoderma* and some unidentified colonies occurred frequently.

Trichoderma species were most common genera which occurred frequently in all the soil sample of 2nd and 3rd phase but in first phase *Fusarium* was dominant genus. Two species of *Trichoderma* as *T. harzianum* and *T. viride* were isolated from KVK, Birauli.

As generally observed during investigation higher fungal population were known to be associated with high organic carbon content of soil but in some sample these result were contradictory. Such contradictory results are probably due to the fact that the microbial population of soil is influenced by various factors of soil and just not only by organic carbon content and pH. Low carbon content in soil indicates poor quality of soil which can be altered by enriching and modifying the microbial flora of soil.

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