Jute and Allied Fibres

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Monitoring of microbial status of retting water and soil of some jute growing areas of South Bengal

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ABSTRACT. Retting water and soil samples of highly productive and quality jute fibre producing zones were collected from Nadia, Murshidabad and North 24 Parganas district to isolate efficient retting microbes and to assess the soil microbial properties. The bacterial and fungal population in retting water ranges from 10 x 105 to 12 x1010 cfu/ml and 0.72 to 22.3 x102 cfu/ml respetively. The soil analysis data showed that organic carbon ranged from 0.52-1.11%, microbial biomass carbon 104.7 to 264.5 g C g^{-1} oven dry soil at 24°C and basal soil respiration rate from 0.12 to 0.24 g CO_2-C g-1oven dry soil hr^{-1} at 22°C. The gas formation pattern of the microbes present in retting water walso determined and it was found that organisms present in the retting water samples are mostly Methyl Red (+) i.e. they produced high amount of organic acids. Twenty nos of retting microbes were isolated from the collected retting water samples and were assessed for their pectinolyric and cellulolytic activity.

Introduction

Retting is the major limitation to the production of high quality fibres. Microbes play a significant role in retting process through biochemical reactions producing plant cell dissolving enzymes. It is evident from the work of various researchers involved in retting, that not a single bacteria or fungi are responsible to carry out the retting process. Number of bacteria (aerobic and anaerobic) and fungi in cumulative mode are responsible for the completion of microbial retting. A recent study by Tulika et al., 2008, revealed that jute retting water is a very rich source of large no. of microorganisms, mainly bacteria. So, there was a need to concentrate research on the evaluation of a microbial consortium having efficient retting bacteria and fungi. Hence, this preliminary study was conducted for assessment of microbial properties of soil and retting water for isolation and characterization of efficient retting microbes of jute growing areas of South Bengal to reach the goal.

Materials and Methods

Collection of soil and water samples

Retting water samples were collected during August and September, 07. For the collection of retting water samples, from three districts viz. Nadia, Murshidabad and North 24 Parganas, each district was divided according to the low, medium and high quality and productive zones. The

retting water was collected from the retting jak merged in water. At the same time, surface (15-20 cm) soil samples were also collected from the same site. The water samples were analyzed for microbial population and soil samples for organic Carbon, basal soil respiration rate and Fluorescein diacetate hydrolyzing activity following the standard procedures.

Enumeration of microorganisms from soil samples

The enumeration of microbial population was done on agar plate containing appropriate media following serial dilution technique and pour plate method (Parmer and Schmidt, 1966). Luria agar and Rose Bengal chloramphenicol agar was used respectively for the cultivation of bacteria and fungus.

Analysis of water samples

The collected retting water samples were analyzed for total microbial population. Water samples were serially diluted and then pour-plated on Luria agar (for bacteria) and Rose Bengal chloramphenicol agar (for cultivation of fungus).

Microbial and enzymatic properties analysis for soil quality determination

Soil samples were analysed for organic carbon, SMBC, basal soil respiration rate, and Fluorescein diacetate hydrolizing activity (FDHA). The FDHA was measured by the method of Alef (1995b). The microbial biomass carbon (MBC) of soil samples was determined by the fumigation extraction method (Joergensen, 1995) using a correction factor (Kec) of 0.38 according to Vance et al. (1987). The basal soil respiration rate was estimated according to the method given by Alef (1995a).

Determination of pectinolytic activity of retting microbes

Retting water samples were first serially diluted and plated on Pectin agar. Colonies were the selected (depending on different colony morphology) and replica plated. Then one of the replicas was flooded with Ruthenium Red. Colonies giving detectable halo zone were selected for further study (Qualitative estimation). After purification; theses bacterial cultures were quantitatively estimated by Reducing Sugar estimation (Dinitro salicyclic acid reagent method) method with D-galacturonic acid as calibration standard (Miller, 1959).

Results and Discussion

Bacterial and fungal population in retting water

The data presented in table 1 revealed that, there was a sharp variation in the bacterial and fungal population in the retting water samples collected from various districts of West Bengal. The bacterial population was very high compared to fungal population in each district. The bacterial as well as fungal population was comparatively higher in Nadia district compared to Murshidabad and North 24 Parganas districts. The bacterial and fungal population in retting water ranges from 10 x 105 to 12 x1010 cfu/ml and 0.72 to 22.3 x102 cfu /ml respectively. The higher presence of retting bacteria and fungi may help in quick retting of jute plants by the secretion of desired enzymes needed for retting (Table 1).

Table 1. Bacterial and fungal population in retting water

Sample No.	Bacterial cfu/ml	Fungal cfu (x102)/ml
RW1	32x105	6.9
RW2	26x105	11.2
RW3	46x105	13.8
RW4	38x105	18.5
RW5	48x105	6.3
RW6	42x105	0.72
RW7	32x105	3.8
RW8	22x105	6.5
RW9	32x105	6.9
RW10	46x105	7.3
RW11	23x105	11.7
RW12	10x105	5.2
RW13	69x105	15.8
RW14	25x105	5.9
RW15	17x105	5.1
RW16	60x105	11.6
RW17	38x105	4.6
RW18	69x105	12.8
RW19	15x105	2.7
RW20	49x105	16.5
RW21	12x10 10	7.1
RW22	46x109	11.8
RW23	29x107	8.7
RW24	65x109	16.1
RW25	65x106	15.7
RW26	32x107	8.2
RW27	17x107	22.3

Gas forming organisms

Presences of gas forming organisms are the indication of water pollution by coliform bacteria. Presence of gas forming organisms in retting water samples were determined by using 1cc. vol. capacity Durham's tube dipped in Lactose Broth.

Table 2. Gas formation b microbes present in retting water

Sample No.	Approx. Vol. of Gas Production (in cc.)		Methyl Red test
	24 hrs	48hrs	
RW 1	> 1	b	+
RW 2	0.2	0.5	+
RW 3	0.8	b	+
RW 4	0.1	>0.65	+
RW 5	0.1	0.5	-

Sample No.	Approx. Vol. of Gas Production (in cc.)		Methyl Red test
RW 6	0.1	0.5	+
RW 7	0.1	0.8	+
RW 8	0.1	b	+
RW 9	0.8	b	+
RW 10	0.1	>0.65	+
RW 11	0.1	0.8	+
RW 12	0.2	>0.65	+
RW 13	0.125	0.6	o
RW 14	0.125	0.2	+
RW 15	0.125	0.5	+
RW 16	0.125	0.6	-
RW 17	0.2	0.8	+
RW 18	0.1	0.5	+
RW 19	0.1	0.4	+
RW 20	0.125	0.4	o
RW 21	0.2	0.5	+
RW 22	0.2	0.8	+
RW 23	0.5	0.9	+
RW 24	N.S	0.2	+
RW 25	N.S	0.6	+
RW 26	0.1	0.4	+
RW 27	0.125	0.3	o

* approx. vol. of gas measured using 1 cc. Durham's tube.
N.S = non-significant amount of Gas is produced.
b = amount of gas production is very high i.e. >1 cc.
(+) = Methyl red (+) organisms present in the sample are organic acid producer
(-)=Methyl red (-) organisms present in the sample produce ethanol or acetone
(0)= organisms present in the sample can only degrade peptone instead of lactose present in the media.

Amount of gas produced was measured (approx.) after 24 and 48 hrs interval. Results showed that Sample no.1 (RW1) had a very high no. of Gas forming organisms, because it produced more than 1cc of gas in less than 24 hrs. Organisms present in the retting water samples are mostly Methyl Red (+) i.e. they produced high amount of organic acids. Only two of them showed negative test, the pH of the medium is raised above 6 due to the production of ethanol or acetone by the organisms present.

Pectinolytic activity of retting microbes

Pectinase activity determines potential of any organism for retting efficieny of jute fibres. Thirty five pectinolytic bacterial cultures have been isolated from 27 retting water samples. Out of these 35 isolates, 22 of them have been assayed so far for their pectinase activity (Fig. 1). Results showed that, 9 of them produce significant amount of pectinolytic enzymes and found very promising for pectinolytic enzyme production. These promising pectinolytic bacteria will be tested for their cellulose and hemicellulose degradation capacity.

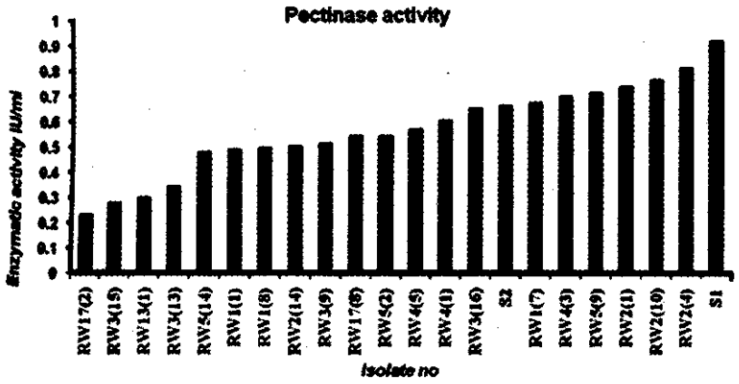

Fig. 1. Pectinase activity of various isolates isolated from retting water.

Soil microbial properties

The soil analysis data showed that organic carbon ranged from 0.52-1.11%, microbial biomass carbon 104.7 to 264.5 ug C /g oven dry soil at 24°C and basal soil respiration rate from 0.12 to 0.24 µg CO_2-C /g oven dry soil /h at 22°C in various districts. The FDHA ranged between 96.5 to 217.0 µg fluorescein /g oven dry soil /h at 24°C. The soil samples collected from Murshidabad district recorded higher values of organic carbon, SMBC, FDHA and BSRR compared to the soil samples collected from Nadia and North 24 parganas districts. The total bacterial and fungal population ranged between 36.2 x 107 to 38.8 x 109 and 1.9 to 15.6 x 106 cfu respectively in various districts.

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