CHAPTER 5
5. SUMMARY

About 85% of the world's jute cultivation is concentrated in the Ganges delta of India and Bangladesh. China, Thailand, Myanmar, Pakistan, Nepal and Bhutan also contribute to jute production. Jute occupies a unique position in the national economy in terms of its contributions to employment in industry and agriculture, as also in foreign exchange earnings in India and other countries.

Jute is currently attracting more attention than the synthetic fibres which poses stiff competitions to jute in terms of quality and economy. Jute is also facing problems in terms of farmers' non-acceptance as remunerative crop compared to cereal crops. However, improved fibre quality is still in good demand and economically remunerative.

Retting largely determines the quality of fibre. Water-retting is in vogue for extracting jute fibres. Microorganisms are the key agents in controlling the retting process. Efficient pectinolytic microbial inoculums with high xylanase and little or no cellulolytic activity can improve fibre quality, reduce the time of retting and evade environmental pollution that arises due to retting.

Experiment 1: Assessment of jute growing soil quality and water quality parameters at different stages of retting and their cumulative impact on jute fibre quality

Soil, water and jute samples were collected from Sonatikari (22°41'27"N; 88°35'44"E) and Baduria (22°44'24"N; 88°47'24"E), two representatively traditional jute growing belts of N-24 Parganas, West Bengal, India. Soils were collected from the fields prior to sowing of jute. Retting water samples were collected at three different times, viz. pre-retting, after first and second charges of retting. Jute fibre samples were collected after first and second charges of retting. The physico-chemical, micronutrient contents, microbiological and biochemical parameters of the soils of Sonatikari and Baduria sites were more or less similar. The water quality parameters varied significantly between the
different stages of retting. The jute fibre quality (strength and fineness) were much superior at first charge than that of the second charge of retting.

The soil and retting water qualities primarily responsible as determinants of jute fibre quality were scored statistically. Subjecting the various variables with high loading rates to multiple regression analysis it revealed that in addition to the predominant number of retting water quality variables, jute fibre strength and fineness also depended on some soil quality parameters. Important retting water quality parameters that largely determined the fibre quality were pH, COD, Hardness, Fe content and microbial count. Important soil parameters in this respect were pH, OC, micronutrient content, microbial count and enzyme activities.

Experiment 2: Isolation, purification and characterization of pectinolytic bacterial strains from jute retting water

Thirty-eight pure pectinolytic bacterial strains (16 from Sonatikari and 22 from Baduria) were isolated and characterized by conventional and molecular techniques. The strains were rod shaped or coccoid to ovoid in shape and arranged singly or in chains or clusters. Among the 16 strains from Sonatikari, 12 were Gram positive and 4 were Gram negative. Regarding 22 strains from Baduria, 15 were Gram positive and 7 were Gram negative. Most of the Gram positive strains were also spore formers.

All the isolates could utilize starch, sucrose, mannitol, lactose and were catalase positive. The 16S rDNA sequences analysis of the isolates and subsequent comparison with the GenBank database revealed that the genus Bacillus was dominant. Other genera which were not reported earlier as jute retting bacteria included Agrobacterium, Microbacterium, Acinetobacter, Serratia, Stenotrophomonas, Citrobacter and Enterobacter.
Experiment 3: Assessment of pectinolytic, xylanolytic and cellulolytic activities of the isolates for screening of most efficient strains

The pectinolytic bacterial strains were screened for polygalacturonase, pectin lyase, xylanase and cellulase activities. Polygalacturonase, pectin lyase, xylanase and cellulase activities of the strains were within the ranges of 8.74 – 41.00 IU g\(^{-1}\), 2.23 – 75.7 U mL\(^{-1}\), 0 – 1.052 µmol mL\(^{-1}\) min\(^{-1}\) and 0 – 0.316 µmol mL\(^{-1}\) min\(^{-1}\) respectively. In most cases, the isolate producing higher polygalacturonase activity failed to produce higher pectin lyase activity. Four organisms were selected based on their high balanced proportion of pectinolytic activity, high xylanase and little or no cellulolytic activity for future use as retting inocula. The selected strains included: SO7 – *B. pumilus* (GQ891103), SO14 – *B. pumilus* (GQ891105), BA1 – *Bacillus* sp. (GQ891097) and BA15 – *B. pumilus* (GQ891098).

The effects of pH of the assay media and different carbon sources in the culture media on the enzyme activities of the selected organisms were studied. Highest polygalacturonase (12.45 to 41.0 IU g\(^{-1}\)) and pectin lyase (23.66 to 75.7 U mL\(^{-1}\)) activities occurred in the alkaline pH of 8.0 and 8.5 respectively. Maximum xylanase activity (0.404 to 0.735 µmol mL\(^{-1}\) min\(^{-1}\)) occurred at pH 7.0. Acidic pH 5.0 favoured the cellulase activity (0 to 0.168 µmol mL\(^{-1}\) min\(^{-1}\)). The screened isolated in different combinations of consortia in most cases showed synergistic effects resulting in increased enzyme activities than the individual strains constituting the consortia.

Experiment 4: Studies on the effect of inoculation of the selected strains on jute fibre quality

The selected bacterial strains (as shown in Experiment 3) were used either alone or in different combinations of consortia like: C1 – SO14+BA15+BA1+SO7; C2 – SO14+BA15+BA1; C3 – SO14+BA15 and C4 – SO14. The organisms in different combinations of consortia reduced the retting period from 19 days in control to 11 – 13 days in inoculated treatments. Inoculation produced golden-yellow fibres with improved
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fibre strength and fineness compared to control. The consortia C1 (strength- 28.91 g tex\(^{-1}\), fineness- 2.76 tex) and C2 (strength- 28.46 g tex\(^{-1}\), fineness- 2.83 tex) showed the best results and promised to be the good candidates for industrial application in extraction of jute fibre.

The pH of the post-retting water samples became acidic, and the electrical conductivity (Ec), chemical oxygen demand (COD) and hardness markedly increased than the pre-retting stage.

Experiment 5: Assessment of changes in community level physiological profile (CLPP) of bacteria at different stages of jute retting

The CLPP of bacterial communities were determined in jute retting water, collected at pre-retting, after 1\(^{st}\) and 2\(^{nd}\) charges of retting at Sonatikari and Baduria. The CLPP, expressed as net area under substrate utilization curve, was studied by carbon source utilization patterns in BIOLOG Ecoplates. Both between locations and stages of retting, substrate utilization patterns were carbohydrates> carboxylic acids> polymers> amino acids> amines/amides> phenolic compounds. Variation in utilization of substrates was observed both between the locations as well as the stages of retting. The microbial communities from retting ponds of both the locations and different stages of retting exhibited utilization of all six groups of carbon sources. This reflected wide metabolic profile of the bacterial communities.

Experiment 6: Assessment of molecular diversity of bacterial communities at different stages of jute retting

The genomic diversity of bacterial communities was assessed in the same water samples as used in Experiment 5. Genomic DNA from the retting water samples was isolated and the 16S rDNA was PCR amplified with eubacterial universal primers \([357F\ldots]\).
(5'-CCTACGGGAG-GCAGCAG-3') and 907R (5'-CCGTCAATTC(A/C)TTTGA-GTTT-3'). The 357F primer contained a GC clamp (5'-CGCCCGCGCGCGCGGCGGGCGGGGCACGGGGGG-3') at its 5' end. The PCR products were subjected to denaturing gradient gel electrophoresis (DGGE). Irrespective of the locations, bacterial community structures varied between the ponds as well as the stages of retting. Difference in banding pattern at different stages of retting was related to dynamics of bacterial groups.