# **Research article**

# Characterization of Banana Fibers Extracted with Pectinase from *Staphylococcus sciuri*

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Keywords	Various methods of isolating cellulosic fibers from banana pseudo- stems including mechanical extraction, steam release, steam release
	combined with chemical treatment, and an enzymatic method with
banana pseudo stem;	pectinase from <i>Staphylococcus sciuri</i> , were performed. The fibers
fiber;	were produced by all the methods and characterized. In this work,
pectinase;	we report on the breaking tenacity, breaking strength, fiber length, diameter, toughness, and linear density of enzyme-retted fiber.
morphological properties;	Banana cellulose fiber isolated by the enzymatic retting procedure was comparable in physical properties to cotton fiber and was also
chemical characteristics	shown by FTIR analysis to contain cellulose, lignin, and
	hemicellulose. The banana cellulose fibers had the following
	characteristics: linear density of 161.81 denier, breaking tenacity of
	38.50 g/tex, elongation of 4.03%, fiber diameter of 127.02 $\mu m,$ and
	fiber length of 191.68 mm.

# Abstract

# 1. Introduction

Banana is a major fruit crop grown in tropical regions around the world. Recent studies have shown that approximately 80 million tonnes of pseudo-stem waste are generated annually in India [1]. The banana pseudo-stem can be used as a raw material for manufacturing products such as paper, cardboard, tea bags, and currency notes, and high-quality composite dress materials. Textile manufacturing processes emit chemicals that pollute water and soil resources. The concepts of environmentally friendliness and sustainable development are currently being built into textile manufacturing processes. Natural fibers are becoming more popular as textile raw materials as they are sustainable fibers having numerous advantages such as durability and environmental friendliness. The use of synthetic fibers has caused additional environmental issues [2, 3]. Cotton, jute, banana, and flax are examples of natural fibers. Fiber physical factors such as fiber diameter, length, tensile properties, and nonwoven structure all influence fabric attributes and performance.

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Banana fiber is eco-friendly, has excellent strength and elongation, and can absorb large amounts of water [4]. There has been a lot of research and development aimed at creating eco-friendly techniques for keeping the balance of the ecosystem intact. However, it is also important to use innovative extraction procedures and to think about using other materials [5].

There are two techniques to extract banana fiber, either manually or mechanically, using the Raspador machine. The fiber extracted by these methods is also available in the market: some extracted by hand and some extracted by machine. Machine-extracted fiber is of lower quality and less expensive, whereas hand-extracted fiber is of greater quality and more expensive. Handextracted fiber has been found suitable for manufacturing high-grade paper due to its great purity, while due to the presence of adhering pith, machine-extracted fiber creates lower quality of the product [6, 7]. Despite commendable efforts from groups like the Krishi Vigyan Kendra and the Khadi and Village Industries Commission KVIC, Mumbai, India to develop better machinery for fiber extraction, not even the best machine on the market can completely separate the fiber from the pith. This pith creates problems during its utilization for creating specialist handmade papers and the pitch behaves as a dead load on the fiber, requiring the use of a lot of chemicals, causing a poor quality of output, and raising concerns about environmental contamination. Therefore, a suitable technology is urgently required to enhance the quality of machine-extracted fiber so that it may become a good quality cellulosic fiber for making varieties of handmade paper and other fibrous products, thereby encouraging better utilization of this valuable bioresource. In the field of bast fiber processing and fabric finishing, various innovative technologies have emerged that involve the use of enzymes to alter fiber parameters, produce the needed attributes, enhance processing results, and reduce environmental impact. Flax enzymatic retting, bast enzymatic retting, hemp enzymatic separation, and flax enzymatic processing before wet spinning are all examples of the new class of technologies that have made mechanical treatments more viable [8]. A lot of studies have been carried out on the retting of bast fibers like flax and hemp, and the enzyme treatment features of textile fibers has been explored extensively. However, there are not many reports available in the literature concerned with the utilization of various enzymes for extracting leaf fibers, particularly fiber from the banana plant. Pectinases are crucial for the digestion of 40% of the dry weight of the plant which is made up of bast and leaf fibers [9, 10].

When natural fibers are utilized in composites, they provide additional benefits such as reduced weight, improved insulation, and sound absorption [11, 12]. Due to their similar qualities, natural bast fiber composites have been utilized as alternatives in glass fiber reinforced composites [13]. Banana fiber is a potential natural fiber for use as a composite reinforcement [14-18]. It can be employed as a suitable reinforcement material in composites depending on the physical and thermal characteristics [19-22]. The chemical composition of plant fibers can be affected by various factors including climatic conditions, geographical location, and so on which can weaken its adherence properties [23, 24]. Research on the use of left-over banana trunk as a fiber source in textiles needs to be encouraged. We propose an eco-friendly procedure that utilizes the enzymatic method in producing the fiber.

# 2. Materials and Methods

#### 2.1 Materials

Banana pseudo-stem from plants of the banana species *Musa acuminata* were collected from local cultivators. The stems were about 2.5 m in length, 0.4 m in diameter, and 10.0 kg in weight. Each pseudo-stem was chopped into 5-inch-long pieces with a sharp knife, thoroughly washed with running tap water followed by two washes of sterilized distilled water, and packed. They were then kept at -80°C for storage till further use.

#### 2.2 Extraction of the enzyme from Staphylococcus sciuri

Pectinase-producing bacteria isolated from the tuber in our lab were revived in nutrient broth and cultivated in pectinase screening agar medium [PSAM]. The nutrient agar contained 1.5% pure pectin. The media was sterilized by autoclaving at 121°C for 15 min before bacteria were streaked on plates. Pectin utilization was detected by flooding the culture plates with freshly prepared iodine-potassium iodide solution. These revived pectinase-producing bacteria were utilized for enzymatic retting of the pseudo-stem [25].

# 2.3 The extraction process for banana fiber

#### 2.3.1 The traditional extraction method control

Banana fiber samples that were available from weavers were collected and these were considered as the control fibers in our study. The traditional method of fiber extraction involves immersing the raw pseudo-stems in a 1% acetic acid solution, and then boiling them twice with 2% NaoH solution. The fibers are then cleaned and dried [21].

#### 2.3.2 Steam release followed chemical treatment (SCM)

In this method, pseudo-stems were heated in presence of water at 180°C for 20 min, followed by the release of steam under 15-17 bar pressure at  $200\pm5$ °C for 3-5 s. Next, the fibers were dipped into a solution containing 6 g/L of NaOH, 6 g/L of Na<sub>2</sub>CO<sub>3</sub>, and 4 mL/L H<sub>2</sub>O<sub>2</sub> or without H<sub>2</sub>O<sub>2</sub> at room temperature for 3 h. The fibers were dried on a blotting sheet [24] as shown in Figure 1.

#### 2.3.3 Steam release followed mechanical treatment (SRM)

This method involved heating the pseudo-stem at  $180^{\circ}$ C for 20 min, followed by therelease of steam under 15-17 bar pressure at  $200\pm5^{\circ}$ C for 3-5 s and after cooling the fibers were mechanically processed combing with a steel comb dried and on blotting sheet see Figure 1.

#### 2.3.4 Enzymatic release method (ERM)

Five percent inoculum of a 48 h old culture of *Staphylococcus sciuri* was transferred into a flask containing 100 ml of sterile mineral medium at pH 8.0 containing lactose, and incubated in a rotary shaker at 150 rpm and 25°C for 24 h. A one gram sample of banana pseudo-stem was aseptically placed into the flask with the help of sterile forceps. The flasks were further incubated for 5 days at room temperature in a rotary shaker at 150 rpm. After the experiment, the treated banana stem samples were then thoroughly washed in running water, sun-dried, and mechanical processed by combing with a steel comb and finally dried on a blotting sheet (Figure 1) [25-27].

#### 2.4 Morphological structure analysis

The fibers produced by different methods mentioned above were analyzed by scanning electron microscope using a MODEL Quanta FEG 250, and FESEM images were created. The instrument was operated at 20000 kV in backscattered electron (BSE) mode. Approximately 20 fibers of each type were randomly selected for the analysis. The magnification range was from 400X to 3000X. Different parts of the fibers were carefully cut to expose their inner walls and then the specimen was coated by sputtering with gold-platinum alloy.

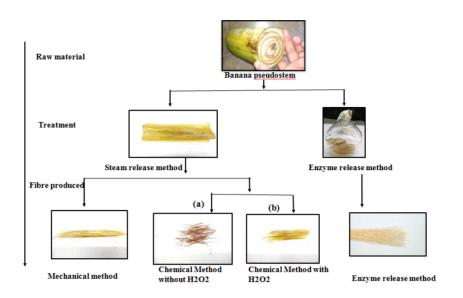


Figure 1. Banana fiber extraction procedures

#### 2.5 Fourier transforms infrared spectroscopy

The fibers obtained by the enzyme release method were sent for further analysis including chemical composition, diameter, length, bundle strength, breaking strength, and elongation to the Uttar Pradesh-based, Government-authorized Northern India Textile Research Association. A PerkinElmer spectrum IR version 10.7.2 spectrophotometer that operated in the 4000-400 cm<sup>-1</sup> transmission range mode was used for the testing.

# 2.6 Fiber diameter, length, strength, and density

#### 2.6.1 Fiber diameter

The fiber was sliced using the microtome, the sections were collected, mounted in cedar wood oil, dispersed with a dissecting needle into the oil to obtain uniform distribution. They were then covered with a coverslip and observed under a microscope at a magnification of 500X. Observed values were converted into micrometers.

#### 2.6.2 Measurement of length

Individual fibers were removed and straightened manually. The length of the fibers was measured against a scale on a sheet of glass oiled with liquid paraffin, and the mean length was calculated.

#### 2.6.3 Measurement of bundle strength

A Pressley type tester is an inclined plane fiber strength tester with a free rolling load cartridge. It is designed to make a flat bundle of fibers, and measures the load required to cause rupture. The load is calculated from a graduated beam scale in pound units. Increasing force is applied on equal sized loose fibers from a composite till they rupture. The ratio of breaking load to the broken fiber weight was determined.

#### 2.6.4 Fiber linear density

Linear density is mass per unit length of fiber. It is expressed in millitex and denier. One denier is the mass in grams of a 9 km length of fiber. The weight of fiber was taken in a microbalance.

Denier = m/n\*l/9000

Where m = mass in mg of the bundle of fiber n = number of fibers l = cut length in mm

The diameters of 20individual fibers were measured with a projecting microscope MP3 with a magnification of 500, and the co-efficient of variation (CV %) was calculated. The lengths of the fibers were calculated by BISFA, and the co-efficient of variation (CV %) was also calculated. Tenacity is defined as the maximum load applied during a tensile test on a bundle of fibers carried to rupture divided by the mass per unit length of the bundle. The strength of fiber was calculated by IS 3675:1966 method. Linear density is defined as the mass per unit length of the fiber. It is usually expressed in denier or millitex. The linear density of fibers was calculated by method IS 10014 Part-2. The results of banana pseudo-stem fiber produced via the enzyme release method were compared with already published data available for cotton, jute, hemp, and flax.

# 3. Results and Discussion

#### 3.1 Extraction of banana fiber with a pure culture of Staphylococcus sciuri

In enzymatic release method, the pseudo-stems were placed in a flask with a revived strain of S. *sciuri* for five days at room temperature. The treated banana stems were then rinsed under running water and dried in an oven at 50°C. The enzymatic treatment was followed by mechanical processing combing with a steel comb, resulting in fibers that were easily separated as individual fibers. Moreover, the fibers then appeared to be of a lighter shade and significantly more visually attractive. When the steam released method was performed, the banana pseudo-stems were treated with steam, followed by either chemical or mechanical procedures. However, it was observed that the fibers could not be separated and the color of the fibers was darker when compared to the fibers that had been enzyme released (Figure 2). The yield of the fiber produced by the enzyme release method was distinctly greater than that produced by the steam release method, as shown in Table 1.

# 3.2 Morphological structure of banana fiber by field emission scanning electron microscopy

Field emission scanning electron microscopy (FESEM) was done to analyze the external properties of the fibers. The cross-section of banana fibers was presumed to be roughly circular and further calculations were based on this presumption [28]. The fibers extracted by the steam release followed by chemical treatment (SCM), the steam release followed by mechanical treatment (SRM) and the enzymatic release method (ERM) were observed under 400X magnification. The ERM method produced smoother banana fibers compared to fibers produced by SRM method (Figure 3).



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Figure 2. Fiber recovered from different extraction procedures

Table 1. Con	parison of the	percent viel	d recoverv o	of banana f	ibers foll	lowing	different treatments

Treatment	Weight of Pseudo-stem Before Treatment	Weight of Pseudo-stem After Treatment	Yield % Final Weight/ Initial Weight *100	Weight Loss %
Steam release chemical method	200g	15.6g	7.82%	92.2%
Steam release mechanical method	200g	5.8g	2.9%	97.07%
Enzyme release method	200g	31.09g	15.5%	84.45%

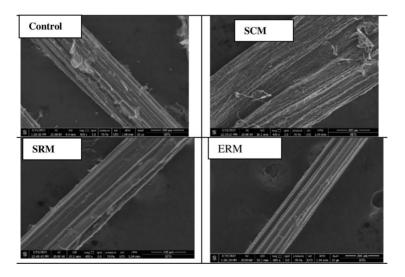


Figure 3. FESEM images of fibers produced by different extraction methods at 400X magnification

#### 3.3 Testing of banana fibers

For the analysis of the properties of the fiber, the ERM fiber sample was sent to NITRA Ghaziabad in order to assess the different parameters of fiber (Table 2).

Table 2. Depiction of characteristics of isolated fiber by enzymatic retting and mechanical method

No.	Test		<b>Covariance CV%</b>
1	Fiber diameter	127.02 μm	41.14
2	Fiber length	191.68 µm	6.07
3	Bundle strength/breaking strength and elongation/ tenacity	38.50g/tex	4.03
4	Fiber denier/ linear density/ fiber fineness	161.81denier	31.17

#### 3.3.1 Fiber diameter

The fiber diameter of banana fiber extracted by the ERM method was found to be 127.02  $\mu$ m. Preethi and Balakrishna [29] previously reported that when fiber was extracted from the pseudo-stems of different cultivars using a semi-automatic machine developed by KVK, C.T.R.I ICAR, Rajamandry, India, the fiber diameter varied from 0.129-0.225  $\mu$ m. Banana fiber extracted biologically by the action of *S. lydicus* MTCC 7505 had a diameter within the range of 150  $\mu$ m-0.15 mm as reported by Jacob and Prema [30].

The diameter of banana fiber when compared with different natural fiber was found to be different. The fiber diameters of cotton, jute, flax, and hemp were found to be 12-45  $\mu$ m, 26-30  $\mu$ m, 20  $\mu$ m, and 16 to 50  $\mu$ m, respectively [29].

#### 3.3.2 Fiber length

The fiber length of the banana pseudo-stem extracted by the ERM method was found to be 191.68 mm, which was quite different to that of other natural fibers. The fiber lengths of cotton, jute, flax, and hemp were reported as 25-60 mm, 0.5-6.0 mm, 6-65 mm, and 2-90 mm [29]. Fibers with greater fiber lengths and diameters but with lower moisture contents are better for use in textile applications than for use in composites.

#### 3.3.3 Bundle strength/breaking strength and elongation/tenacity

Preethi and Balakrishna [29] reported that the tenacity of banana fiber extracted by semi-automatic machine (KVK, C.T.R.I ICAR.) ranged from 5.23-13.51 g/tex. Balakrishnan *et al.* [31] analyzed the tenacity of two varieties of banana fiber whereas the Puwalu banana variety had a tenacity of 7.64-9.34 g/tex and the Ambun banana variety had a tenacity of 8.8-10.9g/tex. Commercially available banana fiber had a tenacity of 24.47 g/tex. Paramasivam *et al.* [32] found that fiber extracted by the action of pectinase had a tenacity of 14.6 g/tex or 14.8 g/tex, but the fiber extracted in the current study had a tenacity of 38.50g/tex which was the highest amongst all. High tenacity means that the fiber has more strength and will not break easily. Some authors also reported the breaking strength of other natural plant fibers which were 30 CN/tex for cotton, 30-45 CN/tex for jute, 54 CN/tex-55 CN/tex for flax, and 47 CN/tex for hemp [2, 29, 33].

#### 3.3.4 Fiber denier/linear density

Banana fibers extracted by the ERM method has linear density of 161.81 denier (Table 2). Balakrishnan *et al.* [34] also reported that linear density depended on the extraction procedure. For example, Sikdar *et al.* [35] reported that when fiber was extracted by a decorticator machine, the linear density was 25-38 tex. The linear densities of other natural plants fiber were previously reported such as cotton with linear density of 1.44 denier [33], jute with 11.7-23.4 denier [31], flax at 3.43-4.18 denier [36], and hemp in the range of 6.3-8.1 denier [34].

#### 3.3.5 Chemical structure of banana fiber by Fourier transform infrared analysis

The FTIR spectra of the banana fibers used in this study seem to be almost the same but slightly different peaks are seen in our result. Main components of lignocellulosic fiber of banana fibers, are cellulose-based materials, as the observed in FTIR spectra There are several absorption bands visible located at 3363.05 cm<sup>-1</sup>, 3336 cm<sup>-1</sup>, 1642.87 cm<sup>-1</sup>, 1322.40 cm<sup>-1</sup>, 1239.75 cm<sup>-1</sup>, 1032.01 cm<sup>-1</sup>, 558.97 cm<sup>-1</sup>, 1373.51 cm<sup>-1</sup>, 983.90 cm<sup>-1</sup>, as shown in Figure 4.

The absorption peaks at 3363 cm<sup>-1</sup> and 3336.94 cm<sup>-1</sup> are assigned to stretching vibrations and other polymeric associations of hydroxyl groups. It was also reported by some researchers that the peak at the absorption band between 3500-3100 cm<sup>-1</sup> was for the hydroxyl group [37-41].

The peak at 1642.87 cm<sup>-1</sup> is due to aromatic C=C stretching and/or asymmetric C-O stretch in COO<sup>-</sup>, and this shows the presence of lignin and other aromatics, or aromatic or aliphatic carboxylates [42, 43]. Sun *et al.* [41] reported that the peak around 1642 cm<sup>-1</sup> is probably due to the bending mode of water, since the hemicelluloses have a strong affinity for water, and these macromolecules in the solid-state may have disordered structures that can be easily hydrated. The band between 1328-1374 cm<sup>-1</sup> is attributed to C-N stretching of aromatic amine, and C–H deformations show the presence of lignin, a component of the plant cell walls.

In this study, we got two peaks in between 1322.40 cm<sup>-1</sup> and 1373.51 cm<sup>-1</sup>, which can be attributed to the C-N stretching of aromatic amines and C-H deformation. We found a peak between 1265-1240 cm<sup>-1</sup>, which is due to C-O-C stretching, C-N stretching and a combination of C-O stretching and O-H deformation at 1030-1080 cm<sup>-1</sup>

Notably, these structures are usually observed in cellulose, hemicelluloses, and lignin, and suggest the aromatic and ethereal nature of the banana fibers. They are significant because the pseudo-stem banana fiber contains functional groups that may permit interaction with metallic ions, and thus banana pseudo-stem fibers can be utilized as biosorbents [44]. The weak broad peaks observed at 558.97, 557.83, and 983.90 cm<sup>-1</sup> are due to deformation vibrations of the C-H bond related to double bond, and C-C stretching [41, 45, 46], but Becker *et al.* [12] found angular deformations of C-H linkages of aromatic groups at 858, 761, 668, and 576 cm<sup>-1</sup>.

# 4. Conclusions

Since banana fibers are strongly bonded together by different chemical components such as lignin, pectin, and hemicellulose, as depicted in the FTIR results, it is difficult to produce banana fibers. However, combinations of various processes at various stages can be applied to produce banana fiber successfully. In this research, banana fiber was extracted by different methods like enzymatic ERM, SRM, and SCM, and the fibers thus produced were then compared with commercially available fiber.

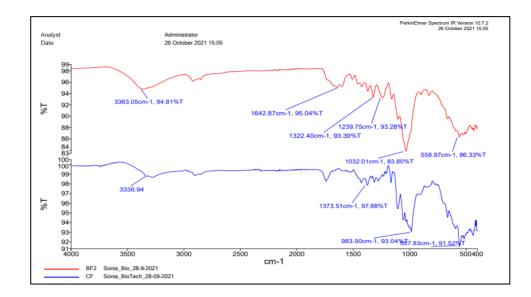


Figure 4. Fourier transform infrared (FTIR) spectra of banana fiber and cotton fiber. The main transmittance peaks of the fibers have been identified.

Fiber obtained by the ERM method was off-white in color, of aromatic and ethereal nature, and had a good tenacity of 38.50g/tex with high fiber length of 191.68 mm and diameter of  $127.02 \mu$ m. Long shiny fibers are preferred as they will usually produce lustrous yarn. Banana fibers need to be graded according to strength, fiber length, diameter, color properties as well as its biochemical nature. A grading system can help in the process of selecting the correct fiber for the relevant industry. Our results of ERM extracted fibers suggest that the fiber could be used in various applications, as a biosorbant for metal remediation, and for packaging. Due to the increasing awareness of environmental issues and demand for eco-friendly raw materials, the demand for sustainable fabrics is expected to grow. The fiber extracted from the banana pseudo-stem shows great potential applicability in the field of organic and ecological textiles, the market for which are predicted to expand tremendously in future years.

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