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Investigating The Properties Of Selected Natural Fiber And The Wound Care Performance Of Developed Diabetic Shoe Insole

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ABSTRACT

The textile industry taps into recent science and technological developments to meet the ever-increasing consumer interest. The applications of textiles are not just limited to regular apparel and home textiles but expand much now on functional applications recognized under the technical textiles globally. Among the various categories of technical textiles, medical textile has played a vital role in addressing various needs of practitioners and patients. Treatment for diabetic wounds still remains challenge in the medical industry. Therefore, it is need of the hour to develop a non-woven composite diabetic insole using natural fiber. This paper is about the research of natural fibers and their properties, with their benefits to prepare nonwoven shoe insoles for diabetic foot Ulcers. Natural fiber nonwoven diabetic insoles have been produced and assessed for their wound healing capabilities. Four fibers were selected to construct a nonwoven composite. Extracted Fiber and developed nonwoven composites was evaluated for its FTIR, SEM analysis and wound healing property..

Keywords: *wound healing, sustainable and renewable resource, natural fiber, nonwoven Composites*

1. INTRODUCTION

Sustainable development is a globally acknowledged concept, though there remains considerable debate over the approaches and strategies for achieving it. (1) Today, scientists worldwide are focused on protecting atmosphere and preserving biodiversity by promoting sustainability along with improving the quality of environmentally friendly products. (2) Recently, there has been a growing interest in utilization of naturally derived materials for medicinal and industrial applications. (3) These materials tend to have minimal adverse effects on human health and generate less environmental waste, reducing their contribution to pollution and global warming. (4)

Natural fibers are regarded as environmentally friendly alternatives because of their favorable mechanical properties, especially when contrasted to synthetic fibers, that might be harmful to environment. Among the key advantages of natural fibers are their low cost, easy availability, low density, biodegradability, renewability, high strength, resistance to corrosion, and overall non-toxic, eco-friendly nature. (5) Properties of natural fibres are affected by the cultivation process, environmental conditions, and extraction process. (6) Natural fibres are expanding in several industries, including automobiles, construction, medicine, furniture, packaging, cosmetics, and other biopolymers and fine chemicals. (7) presence of cellulose, hemicelluloses, pectin, lignin, and waxy substances in the structure of natural fibers enables the absorption of moisture from the environment, resulting in weak bindings between fiber and polymerMaterial selection is a critical aspect of the engineering design process, as it is essential for development and production of sustainable products. the materials are employed to investigate their mechanical and physical properties to enhance the product and increase customer satisfaction. Rather than synthetic fibers, natural fibers are implemented to reduce the weight of composites. As a result, several industrial sectors are experiencing a rise in demand for natural fiber-based composites for commercial purposes. Investigation of natural fibers is critical for developing environmentally responsible composites. (9) In general, bio-composite products are created by combining these fibres with a polymer matrix. Some of primary advantages of these polymer composites are their availability, good formability, good thermal insulation properties, sustainability, low cost, and renewable nature, as well as sufficient energy requirements. (10) A metabolic disease, diabetes mellitus, is characterised by a variety of complications, including neuropathy, arterial damage, and chronic wounds, which are a consequence of uncontrolled blood sugar. Diabetes patients experience a delayed wound healing process due to multifaceted and intricate nature of process. (11) Approximately 25% of DM patients has been experiencing inhumane torture of diabetic ulcers, which is one of the most excruciating complications of DM. Additionally, these individuals are at a higher risk of mortality than patients who do not have diabetic ulcers. (4) Research teams worldwide have done studies on factors influencing diabetic footwear, and the results have shown that it considerably decreased the occurrence of ulcerations on the body segments of patients under examination. For this reason, choosing appropriate footwear is crucial for diabetes individuals. (3) It is crucial to ensure that patients are adequately informed about foot care to prevent diabetic foot complications and amputations. To mitigate the prevalence of foot ulcers and complications, it is imperative to implement appropriate foot health care practices

2. MATERIALS AND METHODS

2.1. Collection of raw materials

Okra plant leftover harvest wastes were procured from indigenous farmers of village at Madhampalayam, Coimbatore, Tamil Nadu, India. Tridox procumbens plant stems are collected from the farms located in Periyanaicken Palayam, Coimbatore, Tamilnadu, India. Aloe vera and Kenaf fibers are procured commercially from Epic green OPC Pvt Ltd., Erode.

2.2 Fiber Extraction:

The fibers from the Tridox plant stem and okra plant stem were extracted by soaking plant in Stagnant water retting method. Green bundles of undried Tridox plant stem and okra plant leftovers waste were immersed in a concrete tank for stagnant water retting process. Bundles were submerged in water for 15 days. Material-to-liquor ratio for the stagnant water retting process was maintained at 1:200. Handpicking was employed to separate fiber reeds following conclusion of the retting process. Separated fiber reeds were subsequently rinsed with an adequate amount of water as well as dried in ambient conditions in the shade for 6 to 7 days. Raw fibers are then softened using enzymes.

Fig 1 a) Tridox Procumbens b) Okra c) Kenaf d) Aloevera

2.3 Enzyme Softening:

Extracted fiber is hard in nature, so the fiber is softened with the enzymes. Bio pectinase K is the enzyme selected for softening based on the presence of plant components. Temperature was maintained at 45°C, pH was set at 4.5, having ratio of 1:40 between fiber and bath volume. pH was determined by fiber weight. Time ranges from 1 to 5hrs, and concentration ranges from 1% to 5%. As a result of enzyme treatment, fibers are prepared for subsequent processing.

Fig 2 Enzyme softening process

2.4 Development of nonwoven composites for Shoe insole:

The extracted and enzyme-treated fibers are cut into small pieces manually and then blended with cotton and kapok fiber. After blending fibers are sent to next process. Blended fibers were mixed manually and fed into the pre-opening machine. Then the fibers are mixed, which ensures proper opening and blending of the fibers. The mixed fibers are then fed into finer opener. Opened fibers were pneumatically fed to card feeder. Reason for carding operation was to eliminate any impurities and separate the fibers, which were then aligned and delivered as a web. After the web formation low melting polyester is melted and dispersed which is added to web formation on a fast-moving belt. The developed nonwoven fabrics were carefully removed from the machine and sent for further analysis. Fig 3 . Presents the developed nonwoven composites for Diabetic shoe insole F1- a) Tridox Procumbens, b) Okra, c) Kenaf, d) Aloevera.

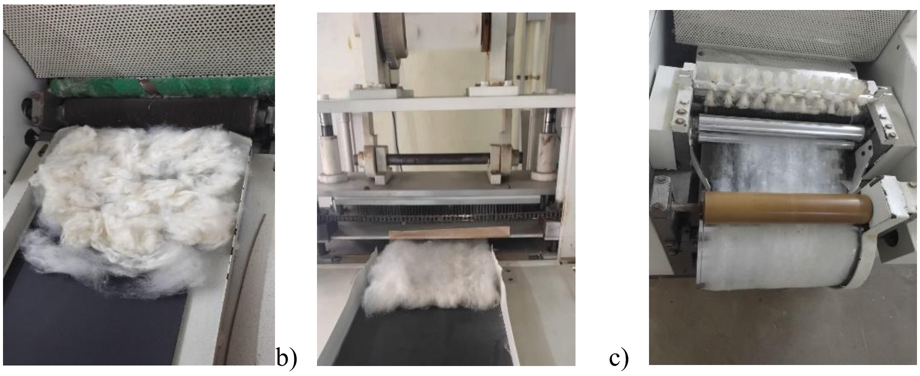


Fig 3 a) fiber blending b) feeding c) web formation

Fig 4 a) Tridox procumbens b) Okra c) Kenaf d) Aloevera

Fig 5 DEVELOPED NONWOVEN COMPOSITES FOR DIABETIC SHOE INSOLE

a) Tridox Procumberns b) Okra c) Kenaf d) Aloe vera

2.5 Characterization of fibers

Characterization methods are more important when selecting natural cellulose fibres to serve as reinforcement in composites. (6)

CHARACTERIZATION OF NON-WOVEN FABRIC FOR DIABETIC INSOLE

2.5.1 SEM analysis

For examining exterior morphology of the extracted natural fibres Tridox procumbens, Aloevera, Kenaf, and okra, field emission scanning electron microscope (FESEM) was implemented. In order to mitigate the impact of electron-beam-charging during the investigation, specimens for FESEM were coated with a gold-palladium layer. A 5 kV Carl Zeiss microscope with electron beam accelerating potential has been employed for recording the micro-graphs of longitudinal view of extracted natural fibres, including Tridox procumbens, Aloevera, Kenaf, and okra. The mean diameter has been randomly estimated from image after morphological observation of micrographs has been conducted.[15]

2.5.2 Wound healing assay: *In vitro* Wound Scratch Assay

The non-woven composite that had been developed for the diabetic insole has been examined for wound healing activities. At density 1X105 cells/ml, L929 murine fibroblast cells has been grown on 24-well plates until they reached around 80% confluency. Following a sterile cell scrapper, small linear scratch has been made in confluent monolayer employing the technique outlined by Liang et al. (2007). Cells treated with produced diabetic insole dissolved in PVA at a 1:1 ratio (25 µl) after being carefully washed utilizing 1 X PBS to eliminate cellular waste. Cell proliferation observed at various time points, including 0, 24, & 48hrs. Digital camera connected to inverted phase contrast microscope(Radical Instruments, India) has been employed to acquire images of migrating cells at all time intervals. Distance traversed by cells migrating into denuded area has been employed for assessing extent of wound healing. [16]

2.4.5 FTIR Analysis

The chemical composition of the developed nonwoven diabetic insole was evaluated utilizing Fourier transform infrared (FTIR) spectroscopy following ASTM E168 standard. The fiber was ground into a powder and combined with analytical – grade potassium bromide (KBr) before being compressed into a disc for measuring. The FTIR spectra were measured using 32 scans in the transmittance mode from 4000 - 400cm^{-1}.

3. 3. RESULT AND DISCUSSION

3.1 SEM Analysis Result

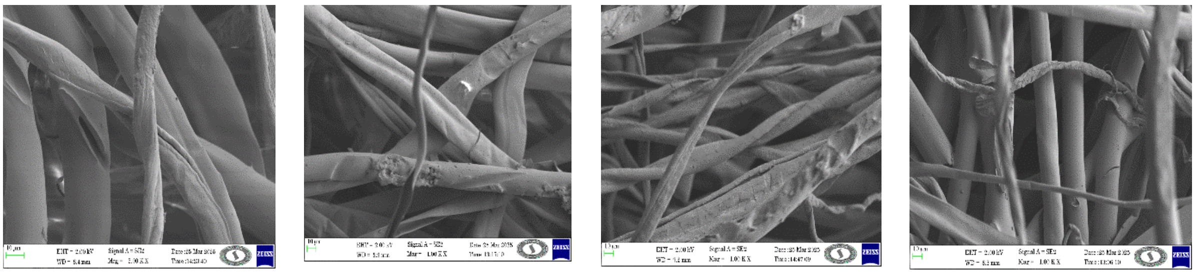


FIG 6 SEM APPEARANCE LONGITUDINAL VIEW OF

Fig 2 a) Aloevera b) Kenaf c) Okra d) Tridox

Scanning electron microscopy demonstrates appearance of raw and treated fiber [17]. SEM reveals surface morphology of fibers found from extracted fiber. The longitudinal views demonstrate respective characteristics, as SEM analysis is an exceptional method for investigating the surface morphology of a variety of fibers. [18,19] Microfibrils are present on surface of aloe vera fibre at 10µm (1.00kx magnitude), as evidenced by presence of vertical lines on longitudinal view. (Fig 6 a) Surface of fiber would have longitudinal pores and a slight roughness, which would increase fiber's thermal capacity. (Fig 6 b) Clearly shows the vertical lines and slight roughness of the kenaf fiber at 10µm (1.00 kx magnitude). The longitudinal fiber section shows that the surface of the fiber is not smooth and has organic compounds and other impurities is shown in (Fig 6 c). The enzyme treated Tridox fiber at 10µm (1.00 kx magnitude) showed the smooth surface and more irregularities at (Fig 2 d).

3.2 Invitro wound scratch Assay

To quantify cell proliferation, cell migration, and wound closure in response to stimulation by specific agents, in vitro wound scratch assay is frequently implemented. In current research *in vitro* wound scratch assay results of developed non-woven shoe insole are shown in the Fig 3. *In vitro* wound scratch assay of developed non-woven composite for shoe insole made of Okra, Aloe Vera, Tridox procumbens, and Kenaf fiber blended with cotton and kapok fiber was assessed at four different timings. Cell proliferation, cell migration, and wound closure were assessed for known concentration of 10µg of developed shoe insole on L929 mouse fibroblast cell lines at four different intervals (0th, 4th, 18th, & 24th hour) following creation of a scratch. Fig 3, which represents the insole's self-wound healing capacity, demonstrated that no cell migration or proliferation has been discovered at zero hour, like control (distilled water).

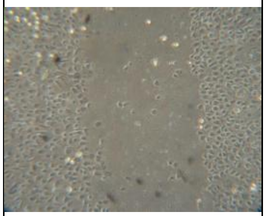
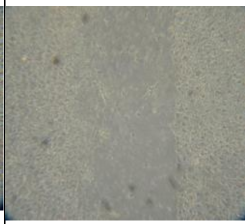
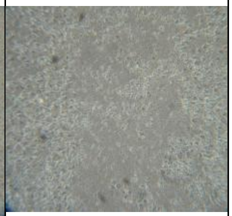
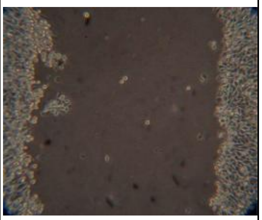
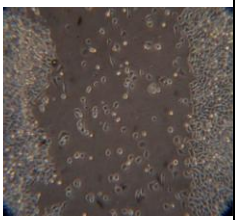
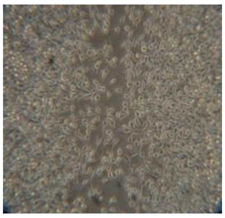
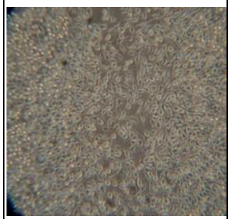
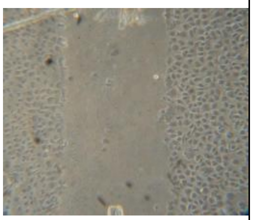
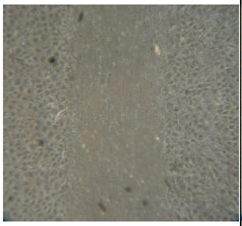
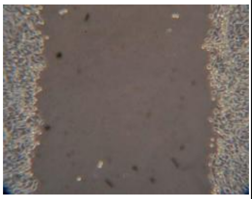
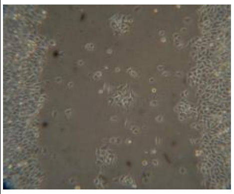
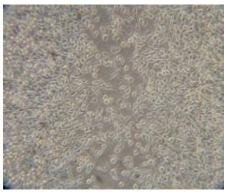
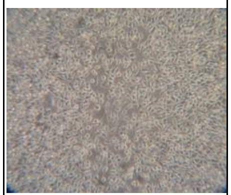
Fiber name	Oth hour	4th hour	18th hour	24th hour
Tridox procumbens				
Okra				
Kenaf				
Aloevera				

Fig 7 In Vitro Wound Scratch Assay

TABLE 1 - INVITRO WOUND SCRATCH ASSAY TEST RESULT

Time Duration (h) Concentration (µg/µl)	**Nonwoven Fabric Aloevera**				
	Wound area (µm)	**0**	**4**	**18**	**24**
25	2660	0	14	63	67
50	2827	0	20	67	72
75	2658	0	24	71	78
100	2683	0	28	75	83
Inference: The given sample showed 83% wound healing activity in L929 cells after 24hrs. Control healed as expected.					
Time Duration (h)	**Nonwoven Fabric Kenaf**				

Concentration (µg/µl)	Wound area (µm)	0	4	18	24
25	1441	0	14	21	23
50	1651	0	25	27	29
75	1783	0	18	23	28
100	1483	0	13	13	20

Inference: The given sample showed 29% wound healing activity in L929 cells after 24hrs. Control healed as expected.

Time Duration (h) Concentration (µg/µl)	Nonwoven Fabric Okra				
	Wound area (µm)	**0**	**4**	**18**	**24**
25	2451	0	15	60	68
50	2588	0	17	65	71
75	2591	0	21	70	77
100	2851	0	30	77	88

Inference: The given sample showed 88% wound healing activity in L929 cells after 24hrs. Control healed as expected.

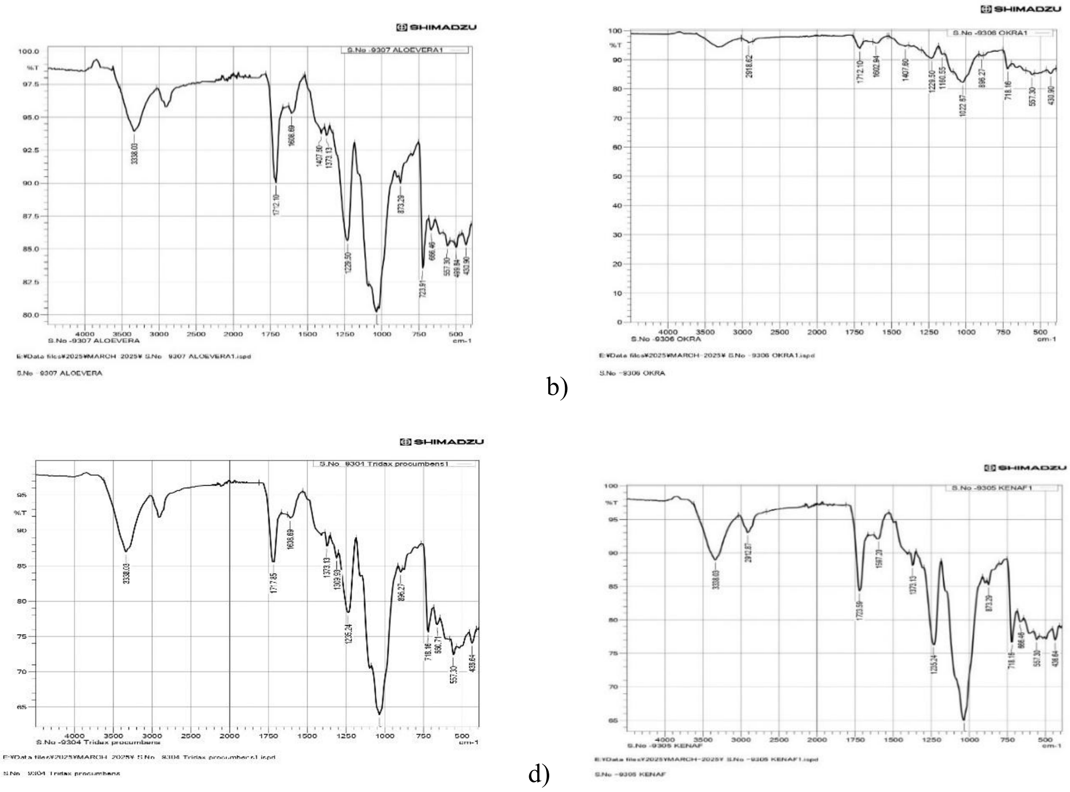
Time Duration (h) Concentration (µg/µl)	Nonwoven Fabric-Tridox procumbens				
	Wound area (µm)	**0**	**4**	**18**	**24**
25	1447	0	7	24	43
50	1151	0	19	23	39
75	1369	0	21	27	30
100	1315	0	13	17	19

Inference: The given sample showed 43% wound healing activity in L929 cells after 24hrs. Control healed as expected.

At the 4th and 18thhour, sample demonsytrated positive cell migration, and cell proliferation was evident, therefore suggesting that the shoe insole that has been developed for wound healing. From the above Table 3, Okra fiber shows the highest wound healing activity of 88% and Kenaf fiber shows 29% lowest wound healing activity.

3.3 FTIR Analysis

Fig. 4(a) demonstrates FTIR of extracted aloe vera fiber. Eleven distinct peaks are observed in the FTIR spectra of aloe vera fiber were visible at 3338.03 indicating the presence of stretching vibrations of OH stretching vibrations and hydrogen bonding, particularly in cellulose-based fiber like cotton. 1712.10 - is often related to C=O (carbonyl) stretching vibration in textile fibers. 1608.69 - indicates the presence of aromatic rings (like benzene rings) or C=C bonds, according to spectroscopy resources. 1407.60 - indicates presence of C-H bending vibrations of aromatic rings. 1373.13 - associated with bending vibration of O-H bonds in water molecules, and can also be related to C-H bonds in cellulose. 1229.50 - indicates presence of amide III, a band related to C-N bond stretching along with N-H in-plane bending. 873.29 - indicates the presence of specific chemical groups or modifications within the fiber structure. 723.91 - indicate the presence of certain molecular structures, such as methylene groups in polymers or bending vibrations of aromatic C-H bonds.666.46 - This peak is often associated with out-of-plane bending vibrations of (CAOH) groups.557.30 - This peak might be associated with the symmetric O-Ti-O stretch in fibers treated with titanium dioxide (TiO_2). 499.84 - generally corresponds to the C-H bending vibration, specifically in the CH_2 group. 430.90 - This suggests the presence of ZnO or a similar compound in the textile sample.



A) b) c) d)

Fig 4 FTIR Test Result a) Aloevera b) Okra c) Tridox Procumbens d) Kenaf

Fig 4(b) shows the Fourier transforms infrared spectrum of the extarcted okra fiber Eleven distinct peak observed in the FTIR spectra of okra fiber were visible at 2918.62 - stretching vibration of C-H bonds, specifically methyl and methylene groups. 1717.10 - is typically associated with the presence of carbonyl (C=O) stretching vibrations, indicating presence of esters, ketones, or carboxylic acids in the fiber. 1602.94 - stretching vibration of C=O double bond within the fiber's components, primarily the ester and amide groups found in cellulose, hemicellulose, and other polysaccharides.1407.60 - stretching of CH_2 bonds in cellulose and hemicellulose components of the fiber. 1229.50 - associated with presence of specific vibrations within molecules, providing information about the chemical structure of the okra fiber. 1160.55 - is associated with stretching vibrations of C-O-C (ether) bond within cellulose and hemicellulose components of the fiber. 1022.67 - corresponds to stretching of a C-O bond in cellulose. 896.27 - this specific wavenumber is commonly associated with C-O-C stretching vibration in cellulose. 718.16 - primarily composed of cellulose, hemicellulose, and lignin. 557.30 - presence of specific functional groups or molecular vibrations. suggests the presence of certain chemical bonds or structures within the fiber's molecular structure. 430.90 - presence of a C-H stretching vibration, specifically a methylene ($-CH_2$) group within the cellulose or other organic compounds of the fiber.

Fig 4(c) shows the Fourier transforms infrared spectrum of the extarcted Tridox Procumbens fiber Eleven distinct peak observed in the FTIR spectra of TPF were visible at 3338.03- OH stretching vibrations and hydrogen bonding, 1717.85- C=O stretching, specifically related to ester or carbonyl groups, 1608.69- C=C stretching within an aromatic ring, such as those found in lignin or other aromatic compounds, 1373.13- presence of C-H bending vibrations in cellulose and hemi cellulose, 1309.93 - It is often referred to as finger print region. It is often associated with vibration of Ch, or CH2 or other relevant groups, depending on the specific fiber type, 1235.24 - is commonly associated with the asymmetric stretching vibration of C-O-C bonds within cellulose and hemicellulose. This peak can also be indicative of cellulose oxidation. , 896.27 - Is typically associated with the amorphous region of cellulose, 718.16 - presence of a C-CI stretching vibration. This type of vibration is characteristic of chlorination, and its appearance in a textile fibers., 660.71 - is often attributed to out of plane bending vibration of C-H bonds in cellulose, 557.30 - associated with the vibration of O-Ti-O bonds as seen in the case of Tio2 treated cotton fiber, 436.64 - associated with biogenic silica. Specifically, this region is noted as a high correlation loading value in PLS – factor 1 for quantitative determination of biogenic silica in marine sediments.

Fig 4(d) shows the Fourier transforms infrared spectrum of the extarcted kenaf fiber Eleven distinct peak observed in the FTIR spectra of Kenaf were visible at 3338.03 - Indicates the presence of stretching vibrations of OH stretching vibrations and hydrogen bonding, 2912.87- stretching vibration of CH2 methylene groups, specifically within the polymer backbone of the fiber 1723.59 - indicates the presence of carbonyl groups C=O. This band is commonly associated with the hemicellulose components of natural fiber, like coir. 1597.20 - indicates the presence of aromatic rings or conjugated double bonds. This peak is commonly associated with C=C stretching vibrations in aromatic rings of polymers. 1373.13- is often attributed to the bending vibration of C-H bonds in cellulose and hemi-cellulose. This peak is generally located within the fingerprint region of the spectrum. 1235.24- Is commonly attributed to the C-O-C asymmetric stretching vibration in cellulose and hemi-cellulose. 873.29 - is often associated with the presence of dehydroalanine groups, which are formed by the cleavage of C-S bonds, or with C-C out-of-plane bending vibrations in benzene rings. 718.16 - indicate presence of C-H bending vibrations in alkyl chains or presence of specific functional groups like those found in polyester or other synthetic polymers. 666.46 - typically indicates out-of-plane bending of (CAOH) group. 557.30 - indicates the presence of a symmetric O-Ti-O stretching vibration, particularly when a textile fiber has been treated with TiO_2. 436.64 - indicate specific chemical bonds or vibrations within the fiber structure.

4. CONCLUSION:

Tridox Procumbens, okra, kenaf, and aloe vera fibers were extracted, and a non-woven composite was developed for Diabetic insoles. All the investigations were done for the characterization of extracted fiber using SEM analysis, FTIR analysis, and wound scratch assay test. By studying the properties of extracted fibers, it is comprehended that all the fiber contains the capability to be used as potential textile fibers and can be converted into nonwoven composites after the evaluation of physical and chemical property fibers are converted into nonwoven composites for diabetic shoe insole and evaluated for wound healing Activity utilizing In Vitro wound scratch assay method. Results of wound scratch test prove that the fibers extracted can be used for Diabetic Shoe Insole

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