



# Polylactic acid-polycaprolactone nanofiber scaffold for anterior cruciate ligament injury

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Opinion

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## Abstract

ACL (Anterior Cruciate Ligament) injuries are becoming more common in athletes. The use of Anterior Cruciate Ligament Reconstruction (ACLR) surgery to treat ACL injuries resulted in a 94% recurrence rate due to the biomechanically repaired tissue being weaker than the original tissue. As a result, biodegradable artificial ligaments that can withstand mechanical stress during no ligament formation and stabilise the ACL must be developed. The goal of this study is to see how composition variations in Polylactic Acid (PLA) and polycaprolactone used as ACL nanofiber scaffolds affect Ultimate Tensile Strength (UTS) and modulus of elasticity, fibre diameter, cytotoxicity level, degradation level, and the PLA-PCL concentration that provides the best value as an ACL scaffold.

Keywords: Assistive Technology, Amputation, Hemiporectomy, Rehabilitation, Prosthesis, Case Report

## INTRODUCTION

Knee injuries, particularly to the Anterior Cruciate Ligament (ACL), are common in today's world. Over 100,000 medical procedures for ACL cases are performed in the United States each year, with an incidence rate of 1/3000. The anterior cruciate ligament (ACL) is a ligament found in the knee that connects the tibia and femur's anterior bones. In human movement, the ACL's primary function is to stabilise the knee by limiting anterior tibial translation and rotation. Because a torn ACL cannot heal on its own, surgery to replace the ACL is required. ACL injuries are treated with Anterior Cruciate Ligament Reconstruction (ACLR) surgery. This surgery, however, has a 94% recurrence rate because the biomechanically repaired tissue is inferior to the original tissue. This is because the substituted material lacks the natural ligament's biomechanical properties. ACLR materials can be derived from the patient's own tissue (autograft) or from donors (allograft).

The hamstring tendon semitendinosus quadriceps and Bone-Patellar Tendon-Bone Grafts (BPTB) are clinically used autografts and allografts, with the BPTB graft integrating better than the other two materials [1]. A third of the ligament that connects the patella to the tibia is used to create the BPTB graft. The disadvantage of this autograft is that it causes knee pain when the patient kneels. AIAT allografts, on the other hand, have a limited donor pool and a higher risk of infection transmission. The current focus of artificial ligament research is on developing biodegradable biomaterials that can withstand mechanical stress during neoligament formation and restore the primary function of knee stabilisation. Biomaterials used to reconstruct the ACL must be biocompatible, have a high initial strength, be biodegradable, be capable of supplying sufficient nutrients and modulating signals to cells, and promote cellular adhesion and proliferation. The current focus of research is on degradable synthetic polymers that do not cause permanent foreign body reactions. PLA, PCL, PGA, and PLGA are biodegradable synthetic polymers that have recently been developed as fibres, membranes, or patches for a variety of biomedical applications [2,3].

## MATERIALS AND METHODS

Good fellow and Sigma Aldrich supplied the primary ingredients, Poly-Lactic Acid (PLA) (Mn 100.000) and Poly-Caprolactone (PCL) (Mn 80.000). Merck provided the chloroform and Dimethyl Formamide (DMF). Distilled water, Phosphate Buffered Saline (PBS), and 3-(4,5-dimethylthiazol-2-yl) 2,5-diphenyl tetrazolium bromide were provided by Oxoid (MTT). A Genlab HK-7 electrospinning type was used to create nanofiber scaffolds from PLA-PCL. The procedure starts with making a solvent out of a 4:1 mixture of chloroform and DMF. After that, make a 10% w/v solution of each PLA and PCL. The PLA-PCL sample was then dissolved in 10 mL of chloroform-DMF solution with modifications in the composition of the varied PLA-PCL, namely A (100:0), B (85:15), C (80:20), D (70:30), and E (0:100) in wt%, using a 200 rpm magnetic stirrer. The process of dissolving took two hours. The resulting PLAPCL copolymer appeared clear after homogenising the PLA-PCL. When the solution was homogeneous, the electrospinning process began to create fibre. After that, the syringe is connected to the electrospinning apparatus [4,5]. To collect the fibre, the electrospinning process uses a flat-shaped collector covered in aluminium foil. The electrospinning parameters include a spinneret distance to the collector of 15 cm, a needle tip diameter of 0.5 mm, a flow rate of 2 mL/h, and a voltage of 20 kV. The electrospun fibre was referred to as electrospun, while the single nanofiber was referred to as nanofiber. After that, the electrospun fibre was cut to a length of

26 cm before being twisted and braided. Fourier Transform Infrared (FTIR) Spectrophotometer Functional Group Test FTIR (Shimadzu IRTracer-100) at wave number 400 cm<sup>-1</sup> -4000 cm<sup>-1</sup> was used to analyse the functional group. The FTIR analysis was carried out using a cuvette filled with 1 cm sample pieces. The FTIR method was used to assess the spectrum relationships between transmission percentage and wavenumber. Using a Hitachi TM3000 Tabletop Microscope at magnifications of 5000 and 10,000, the nanofiber scaffold was morphologically characterized. The fibre diameter value was determined by analyzing the SEM image with the ImageJ application. The first step in evaluating particle size in a SEM image using ImageJ is to calibrate the image pixel size against the reference size. The reference size is typically displayed on the SEM image as a line with a scale indicating the magnification level used. ImageJ software (Version 1.50b, National Institutes of Health, Bethesda, MD, USA) is used to determine the average fibre diameter, and the data is then processed using Origin lab 2016 to generate a sample diameter distribution plot. Using the ImageJ application, another parameter that can be determined microscopically from the surface structure is the light-dark area fraction; the percentage of dark and light areas, where dark areas represent empty space and light areas represent the fibre formed. To begin, the SEM image is segmented using a threshold to help distinguish between nanofiber and background. The area fraction analysis can be performed using Image J's histogram feature after adjusting the threshold. According to the histogram, a value of 0 represents a dark area and a value of 255 represents a light area. The dark area represents the porosity of the fibre [6,7]. Another parameter that can be determined microscopically from the surface structure is the light-dark area fraction; the percentage of dark and light areas, where dark areas represent empty space and light areas represent the fibre formed, using the ImageJ application. To begin, the SEM image is segmented using a threshold to make the distinction between nanofiber and background more obvious. After adjusting the threshold, the area fraction analysis can be performed using Image J's histogram feature. The histogram shows that a value of 0 represents a dark area and a value of 255 represents a light area. The dark area represents the porosity value of the fibre [8]. Mechanical testing was carried out on a machine that was outfitted wi-

-th an electronic universal system for material testing (AGS-X, Shimadzu, Co., Kyoto, Japan). According to the United States. The sample was formed into a dogbone using Standard Testing and Material (ASTM) type V. shape. The load-deformation curve was captured and used to calculate the ultimate failure load (N). The slope of the load-deformation linear region was used to calculate the UTS. curve at the maximum load-to-failure point [9,10].

## RESULT

We successfully created a PLA-PCL nanofiber scaffold with a variety of compositions, including A (100:0), B (85:15), C (80:20), D (70:30), and E (0:100) in weight %. The electrospun nanofibers we collected on aluminium foil are white in colour. After that, the electrospun fibres are separated from the aluminium foil. The sample was then cut to a size of 26 cm and manually twisted and braided. For tensile testing, the resulting sample is divided into two sections: the braided sample (denoted by A) and the unbraided sample (denoted by B) Scanning Electron Microscopy is used in the morphological technique to determine the structure of a sample. This measurement used 10,000 magnification to ensure that the sample was clearly visible, and fibre diameter measurements were analysed using the software. The SEM observations are the results of the fibre diameter examination. All samples exhibited perfect fibre formation morphologically due to the absence of beads. Except for sample C, the fibre diameter decreases as the PLA composition decreases. PLA has a higher molecular weight than PCL, resulting in a viscous solution. As the PLA composition increases, the solution thickens, increasing the diameter of the resulting fibre. The fibre formed from all samples is of high quality, with an average diameter of 50 nm -1000 nm, which corresponds to the diameter of the fibre in the extracellular matrix. Fiber diameter distribution appears to be non-uniform. This is because of a combination of factors, including the voltage used and the constant distance between the syringe and the collector for all samples. The viscosity of the solution varies with the composition of the sample. To achieve a consistent diameter, several electrospinning process parameters, including the viscosity of the solution, the electric voltage, and the distance between the syringe and the collector, must be combined. Some fibres are fused (fibre fused) to the fibre beneath. The fraction area is another parameter that can be determined microscopically from the surface structure. The ImageJ software can be used to calculate the percentage of dark and light areas, where dark areas represent empty space and light areas represent formed fibre. According to the histogram, 0 represented a dark area (fibre porosity) and 255 represented a light area (fiber density). The Histogram area fraction analysis revealed that the dark area represents the fibre porosity value and the bright area represents the fibre density value. The dark area

represents fibre porosity, while the bright area represents fibre density. The higher the fibre density, the more robust the mechanical properties produced.

## DISCUSSION

The anteromedial and posterolateral bands are two ribbon-like fibres that make up the ACL. The anteromedial band is especially tight during flexion and extension, and it gets even tighter when the knee is bent. The posterolateral bands become moderately relaxed when the knee is bent. The ACL is made up of several intertwined collagen fibres, giving it a high tensile strength. In the tibial and femoral basins, the ACL angle ranges from 60°- 80°. Greater UTS and modulus of elasticity are required to allow for degradation of properly bearing specimens. Male UTS ACL values ranged from 16.3 MPa - 36.4 MPa, while female values ranged from 13.7 MPa -31.5 MPa. Men's modulus of elasticity ranges from 93 MPa -163 MPa, while women's modulus of elasticity ranges from 49 MPa -149 MPa.

## CONCLUSIONS

In this study, variations in the composition of PLA:PCL resulted in the formation of distinct functional groups for each PLA and PCL. According to the results of SEM morphology data, all samples have fibres with diameters less than 1000 nm. In the future, DSC and XRD measurements will be required to determine whether the samples are crystalline or amorphous, which is important for understanding their mechanical properties. The Tensile Test, UTS, and Modulus of Elasticity values remained within the original ACL range for both unbraided and braided samples, and the higher the PCL composition, the lower the UTS and Modulus of Elasticity values.

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