BIORESORBABLE POLYMER BLEND SCAFFOLD FOR TISSUE ENGINEERING

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Tissue engineering merges the disciplines of study like cell biology, materials science, engineering and surgery to enable growth of new living tissues on scaffolding constructed from implanted polymeric materials. One of the most important aspects of tissue engineering related to material science is design of the polymer scaffolds. The polymer scaffolds needs to have some specific mechanical strength over certain period of time. In this work bioresorbable aliphatic polymers (PCL and PLLA) were blended using extrusion and solution methods. These blends were then extruded and electrospun into fibers. The fibers were then subjected to FDA standard in vitro immersion degradation tests where its mechanical strength, water absorption, weight loss were observed during the eight weeks. The results indicate that the mechanical strength and rate of degradation can be tailored by changing the ratio of PCL and PLLA in the blend. Processing influences these parameters, with the loss of mechanical strength and rate of degradation being higher in electrospun fibers compared to those extruded. A second effort in this thesis addressed the potential separation of the scaffold from the tissue (loss of apposition) due to the differences in their low strain responses. This hypothesis that using knit with low tension will have better compliance was tested and confirmed.
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1.1 Tissue Engineering

Tissue engineering is defined as “an interdisciplinary field that applies principles of engineering and life science towards the development of biological substitutes to restore, maintain and improve the function of biological tissue and organs” [1]. It tends to merge different fields of study like cell biology, materials science, engineering and surgery to make new living tissues and scaffolding from different materials and implantation of the tissues. One of the most important aspects of tissue engineering related to material science is design of the polymer scaffolds. Retention of strength and controlled bioresorption are two important functionalities required by the polymer. The polymer scaffold also needs to have similar biological properties to extracellular matrix in order to control cellular behavior. The extracellular matrixes are composed of nanometer size proteins and glycosaminoglycans [2]. The scaffoldings used for tissue engineering needs to function as temporary extracellular matrix until they are repaired or regenerated. The scaffold provides the support to the cells to develop and attach in vitro [3]. Then the scaffold with cells grown in them can be implanted into the injured site, so that the cells can repair or regenerate [4]. The desired property of the scaffold varies from tissue to tissue. But all of the scaffoldings need to be biocompatible with the host body. It should integrate with the host tissues without creating any major immune response to reject the implant [5]. The scaffold should be porous and have a large surface area for the cell to attach and grow. The porosity of the scaffold will allow the
formation of blood vessels in the implant area of vascularized tissues. In addition
matching the mechanical response of tissues in the implant area is a valuable property [6].

Surgical meshes used today as tissue scaffolding meet and exceed several of
the requirements of tissue engineering. They are mostly made from commercial
polymers like polyethylene or polypropylene. They are commonly used in Hernia
Repairs, Pelvic Organ Prolapse (POP) and Stress Urinary Incontinence (SUI). The main
reason for using surgical mesh is to support repaired soft tissues in order to minimize
the reoccurrence of the problem or injury [7,8]. Tissue scaffoldings made from
polyethylene or polypropylene is permanent and they are not reabsorbed by the body.
Should removal be sought, a patient has to receive another surgery to take out the
surgical mesh from the body, with a subsequent long process of healing. There has also
been concern over the retention of a foreign object in the body. In 2008, US Food and
Drug Administration (FDA) issued a warning for use of traditional surgical mesh. It
states that surgical meshes tend to drop from their original position causing injury to
nearby organs, nerve or blood vessels [9].

The reasonable alternative to using traditional non absorbable surgical mesh as
tissue scaffold for tissue engineering is to use bioresorbable polymer mesh. The
polymer could be a single system or a blend of bioresorbable polymers. Blends offer the
possibility of tailoring properties to the needs. Bioresorbable polymer meshes solve the
problem facing surgical meshes used today. Its use would eliminate the need of re-
surgery to remove the implant, as it gets absorbed by the body in a finite period of time
[10].
1.2 Polymer Blends

Polymer blends are mixtures of two or more polymers. [11]. Polymer blending has very long history in commercial polymers especially in rubber, coatings and adhesive industries. It entered the polymer industry around half century ago. It has been growing rapidly since then and now accounts for about 30-40% of total plastic market worldwide [12]. Polymer blending has very strong commercial importance because it offers properties not available in a single polymer. So blending two different polymers is an important tool for industrial production to get tailored material properties [13]. Since it represents a large and important part of industrial production, it is subjected to intensive research.

In early days when plastic chemists started mixing two chemically different polymers, they defined differences in the mixtures as miscible and immiscible polymer blends. Miscible polymer blends are homogenous single phase mixture of polymers with properties proportional to ratio of the blends. But immiscible polymers are heterogeneous mixture of polymer with two or more phases. It need not have properties proportional ratio of blends like miscible blends. Miscible polymer blends have lot of commercial importance, but immiscible polymer blends also have lot of significance by combining polymers each with useful properties in the blends. There are numbers of factors which contribute to the miscibility/immiscibility of the polymer blends [14]. These factors are polarity, specific group attraction, molecular weight, ratio, crystallinity.

Between the two cases of miscibility and immiscibility, there are range in between with partial miscibility and intermediate attractions. These provide a chance to tailor the property by varying different degree of miscibility. It has been found that some
properties are good at complete miscibility, some at intermediate miscibility, some at low miscibility and some at complete immiscibility [15]. In most blended systems, some kind of human intervention is needed to get the optimum properties, which is called compatibilization [16]. The compatibilization of polymer can be done by physical processes, physical additives, polymer modification or reactive blending.

1.3 Biodegradable Polymers

There has been a significant amount of research on polymers synthesized from glycolic and hydroxyl acids during first half of the 20\textsuperscript{th} century but this research was largely abandoned due to environmental instability of polymers for long term use [17]. The degradation of polymers occurs mostly by scission of main or side chains of the polymer molecules. The scission of polymer chains are induced by thermal activation, oxidation, photolysis, radiolysis or hydrolysis [18]. Some polymers also degrade when exposed to the environment in the presence of microorganism. These environments can be anything from sea, land or body of living animals or humans [19,20 ,21 ,22 ,23]. So polymers which undergo chain scission due to microbial activity leading to mineralization are called biodegradable polymers [24]. For biodegradable polymers to degrade, specific conditions like pH, humidity and oxygenation are required.

There is growing interest in biodegradable polymers due to attractive environmental, biomedical and agricultural applications. Biodegradable polymers offer a desperately sought alternative to the high volume of waste generated from non degradable polymers. Development of biodegradable products like fishing nets and, packaging films can greatly reduce the plastic waste accumulation. Since most of the
biodegradable polymers are plant based polymers, this also helps in sustainable material development, as it reduces the carbon footprint of humans and provides an application for agricultural waste [25]. New innovations in biodegradable polymers help preserve fossil based resources and complete the material lifecycle by degrading. The degradation of polymers reduces the volume of the garbage and creates compost in nature. These advantages have resulted in significant academic and industrial interest in biodegradable polymers. Despite these great advantages of biodegradable polymers, their use had been hindered by the problem of limited commercial production of these materials [26]. Many biopolymers have been stuck at lab scale productions. In the long term these are not permanent problems. There has been much research and development for producing monomer from agricultural based materials to support production of biodegradable polymers in an industrial scale and still be economical [27].

1.4 Biocompatibility

In the medical field of implants and tissue scaffolding, biocompatibility of materials is a major concern. Reaction of the host to the foreign material inserted for long period of time cause inflammation and rejection. The term biocompatibility is defined as “the ability of a material to perform with an appropriate host response in a specific application” [28]. So any polymeric material inserted in the body must be biocompatible. Biocompatibility issues start from the first contact of foreign material to the body tissue, its changes over the period of time and its degradation. The host body tries to reject an inserted implant by causing inflammation and irritation of the tissues in
the inserted area. So for any material to be called a biocompatible polymer it should meet and exceed the requirements stated below in the Table 1.1.

Table 1.1. Requirements of biocompatible polymer.

<table>
<thead>
<tr>
<th>Requirements of biocompatibility</th>
</tr>
</thead>
<tbody>
<tr>
<td>Non-toxic</td>
</tr>
<tr>
<td>Non- carcinogenic</td>
</tr>
<tr>
<td>Non-mutagenic</td>
</tr>
<tr>
<td>Non-allergic</td>
</tr>
<tr>
<td>Contaminant free</td>
</tr>
<tr>
<td>Minimal inflammatory response</td>
</tr>
<tr>
<td>Bioresorbable</td>
</tr>
<tr>
<td>Biocompatible degradation products</td>
</tr>
</tbody>
</table>

There are many standards to assess the biological and immunological response to the foreign material as an implant for biocompatibility [29]. These standards serve as the guideline to adequately evaluating biocompatibility [30]. In 1992, the International Standards Organization (ISO) issued an international standard of biological evaluation of medical devices (ISO 10993) which the FDA followed this up in 1995 with its own version.

Biocompatible polymers can be divided into two groups: biostable and bioresorbable. Biostable polymers are simply inert and biocompatible in the host but do not degrade for decades, maintaining their mechanical properties. These polymers are
used mainly for long term permanent aids. The most common biostable polymers are polyethylene, polypropylene and poly(methyl methacrylate) [31]. On the other hand, bioresorbable polymers are also inert and biocompatible, but also degrade over time in the host. So they can be used in short term temporary aids like sutures, tissue scaffolding and drug delivery devices. When bioresorbable polymers degrade in the body, the fragments are completely metabolized to soluble molecules that get excreted out of the body.

1.5 Bioresorbable Polymers

The biodegradable polymer which specifically degrades by hydrolysis is known as hydrolytically degradable polymers. These polymers found their way in medical industry by being first approved as biodegradable sutures in the 1960s [32]. After that many polymers based on lactic acids like polylactide as well as poly(caprolactone), poly(dioxanone) have been accepted for use as medical devices [33]. Further more research is going on the polymers like polyorthoesters [34], polyanhydrides [35], and other materials [36,37].

The prospect of polymers degrading in the body hydrolytically due to enzymatic reaction in the body is very attractive for medical purposes. So the polymers which degrade hydrolytically in the body and its degradable products can be eliminated from the body are called bioresorbable polymer. The bioresorbable polymer degrades to smaller molecular chains and soluble oligomers due to hydrolytic reaction in the body over an extended period of time. These byproducts are eliminated from the body through natural pathways which are involved normally in metabolic pathways [38].
This property of the polymers is now attracting much research and application attention from the medical industry [39,40,41]. The main reason for special interest of medical industry is that the degradation of implants simply means that no further surgical procedure is required to remove the implant at the end of its functional life. This eliminates the need for a second surgery to remove the implant [42]. In the field of tissue engineering, the biodegradable polymer scaffold can be designed according to the tissues which can provide mechanical support and appropriate surface for cell growth and its attachment [43]. It can also be designed to control its degradation rate which will allow the load to be transferred to the new tissue after it heals [44]. Also in the field of controlled drug delivery, the biodegradable polymers have new prospects as a drug delivery system or as an associate to the medical device [45].

1.6 Polymer Degradation

The term degradation simply means cleavage of bonds in the polymer chains. The chains of polymers break by hydrolysis in hydrolytically degradable polymers. The kinetics of the chain scission by hydrolysis is greatly controlled by the anions, cations and enzymes [46]. The degradation of the polymer mainly depends upon its backbone structure [47]. The polymer only degrades if it has hydrolysable or oxidizable linkages in its backbone. But the rate of degradation is dependent on its composition, molecular weight, morphology, hydrophilicity, crystallinity and surface area [48]. The hydrolytically degradable polymers are dominated by hydrolysis but it is also helped by enzymatic activities [49,50]. pH is also an important variable. The hydrolysis process can be catalyzed by both acid and bases [51]. Physically degradation occurs through two
routes: surface erosion and bulk erosion. In surface erosion, the materials are lost from the outer surface facing the environment. The degradation of polymers by surface erosion is proportional to the surface area as larger the surface area faster the degradation. But in case of bulk erosion, the materials are lost from the entire polymer matrix. Both of these processes of biodegradation are affected by the hydrophilicity and crystallinity of the polymers [52]. Water tends to enter the polymer from the amorphous region but crystalline areas are also penetrated slowly [53]. If the polymer is very hydrophobic and does not significantly absorb any water at all, the surface erosion will be the main process of degradation.

1.7 Aliphatic Polyester

Aliphatic polyesters have been known and studied since 1930s [54]. It is one of the most promising polymers to be used as bioresorbable implant to packaging material. This family of polymers is biodegradable and biocompatible. These polymers are mostly used in the degradable drug delivery systems. The first generation of bioresorbable scaffold for tissue engineering was designed using aliphatic polyester [55,56]. These aliphatic polymers degrade by cleavage of ester bonds by hydrolysis [57]. The commonly known polymers in this family are in the Table 1.2 below.
Table 1.2. List of aliphatic polyesters.

<table>
<thead>
<tr>
<th>Aliphatic Polyesters</th>
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<tbody>
<tr>
<td>Poly(glycolide) or PGA</td>
</tr>
<tr>
<td>Polylactide or PLLA</td>
</tr>
<tr>
<td>Poly(lactide-co-glycolide) or PLGA</td>
</tr>
<tr>
<td>Poly(ε-caprolactone) or PCL</td>
</tr>
<tr>
<td>Poly(hydroxybutyrate) or PHB</td>
</tr>
<tr>
<td>Poly(hydroxybutyrate-co-hydroxyvakerate) or P(HB-co-HV)</td>
</tr>
</tbody>
</table>

1.7.1 Poly(ε-caprolactone) (PCL)

PCL is one of the most studied bioresorbable polymers. It is one of the most important polymers in the aliphatic polyester family. These days it is one of the most sought after biodegradable and bioresorbable flexible polymer due to its ease of processability [58]. PCL degrades enzymatically in the human body but non enzymatically in the environment [59,60,61]. It can be processed using injection molding and extrusion without significant loss in molecular weight. PCL is synthesized simply from the crude oil. It is polymerized by ring opening polymerization reaction of ε-caprolactone monomer using catalyst. The chemical reaction of producing PCL is shown in Figure 1.1.
Figure 1.1. Ring opening polymerization of poly(ε-caprolactone).

1.7.2 Polylactide (PLLA)

One of the most promising biodegradable and hydrolytically degradable polymers is polylactide or polylactic acid (PLLA) [62]. The biodegradation of PLLA is very complex process. Its rate of degradation is very slow process. It is produced from agricultural products and biodegrades easily [63]. PLLA is not a new material in the market as it has been studied by various research groups for years. It is known that, in 1845 low molecular weight PLA was made by condensing lactic acid to form cyclic dimer of lactic acid called lactide [64]. Then in 1894, lactide was polymerized to form PLLA [65], but this method was not commercially viable [66]. The monomer lactides were commercially produced by fermentation of petrochemical feedstocks. These days the most prevalent method of producing lactide is direct bacterial fermentation of corn starch. Then these monomer lactides lead to ring opening polymerization to produce high weight molecular PLLA. More recently, Cargill-Dow began using a solvent free method and a new distillation process to produce large variety of PLLAs [67,68]. On the other hand, Mitsui Chemicals has been using a solvent based method where high molecular weight PLLA
is produced by direct condensation using azeotropic distillation [69]. PLLA offers the
balance of biodegradability and mechanical properties. It can be easily fabricated into
parts using traditional polymer fabricating equipment. It is a promising polymer with
many possible applications [70,71]. Its high crystallinity, rigidity and slow degradation
are very attractive for various applications.

1.8 PCL and PLA Blends

Blends of PCL and PLLA are very attractive considering both are biocompatible
and bioresorbable. It is attracting lot of interest with the possibility of tailoring a blend
with the flexible component being the PCL and rigid component being the PLA. Both
polymers have been approved by the FDA for medical and drug delivery systems
[72,73]. Unfortunately PCL and PLLA are immiscible with each other. They are reported
to be immiscible in both molten and bulk state [74]. Even when molten, both of the
polymers remain phase separated and minor components isolate in coarse domains
[75]. Given that its morphology and dispersion sizes will influence the properties of an
immiscible system significantly, it is vital that an efficient mixing system such as a co-
rotating twin screw extruder be employed [76].

1.9 In Vitro Degradation

In vitro degradation tests implants simulation of the human body fluid in the glass
tube. This is usually done by using phosphate buffered saline solution at pH of 7.4
which resembles human body fluid. When the polymeric samples are first subjected to
the in vitro degradation, water uptake by the sample is a preliminary result. From a
diffusion concept, we can say that this process creates a negative water concentration gradient from surface to the center of the sample. However this gradient of water concentration nullifies fast as water or salts diffuse faster than degradation rates. So it can be said that the hydrolysis of the ester bonds during in vitro degradation is homogenous from the start which can be observed by the weight loss [77]. But the rate of diffusion is greatly controlled by the hydrophobicity and crystallinity of the polymers. So highly crystalline or hydrophobic will have slow degradation compared to amorphous and hydrophilic polymers.

During in vitro degradation the aliphatic polyester samples will have increased number of carboxylic acids on the ends of the chains due to degradation process. Then carboxylic acids help in ester hydrolysis since these carboxylic acids on the chain ends are known to autocatalyze the ester hydrolysis process [78]. Then the oligomers which are soluble in the aqueous medium near them escape from the matrix. The soluble oligomers leach out of the surface but those inside the matrix are entrapped and only participate in an autocatalytic effect [79]. The diffusion coefficient of the soluble oligomers depends upon molar mass, swelling of matrix and rigidity. So the degradation rates also depend on the swelling of the matrix and distribution of chiral and achiral units of the polymer chains [80,81]. The release of soluble oligomers depends upon the surroundings like pH, ionic strength, temperature and buffering capacity [82]. But if the degrading material becomes crystalline during degradation due to loss of all the amorphous phase, the degradation of crystalline phase is faster inside than outside if the material swells or absorbs aqueous solution [83]. These issues lead to a size effect where a larger sample of the same material degrades slower than a smaller sample.
CHAPTER 2
MATERIALS AND METHODS

2.1 Materials

2.1.1 Poly(ε-caprolactone) (PCL)

PCL is biodegradable polyester made from synthesis of crude oil. It is a semicrystalline polymer with crystallinity around 50%. It was bought from Union Carbide Corporation (Danbury, CT). It has a molecular weight of 50,000 D. It has low melting point at 60°C and very low glass transition temperature at -60°C. It has very high thermal degradation temperature of 350°C, so it can be easily blended with high melting polymers like polylactide(PLLA) or polypropylene. Solvents used in the study like chloroform and acetone were of analytical grade. The chemical structure of PCL is shown below in Figure 2.1.

![Chemical structure of PCL](image)

Figure 2.1. Chemical structure of PCL.
2.1.2 Polylactide or Polylactic Acid (PLLA)

PLLA is a semi crystalline polymer with melting point \( T_m \) of 174°C and glass transition temperature \( T_g \) of 61°C. It was supplied from Nature Works (MN, USA). The PLLA used had \( M_n: 98,600; M_w: 194,799 \). The crystal structure of PLLA is pseudo-orthorhombic [84]. The chemical structure of PLLA is show below in Figure 2.2.

![Chemical structure of PLLA](image)

Figure 2.2. Chemical structure of PLLA.

2.1.3 Bioresorbable Sutures

The bioresorbable sutures used were polyglactin coated Vicryl Plus™ from Ethicon Inc (Somerville, NJ). It is an undyed braided suture which is a sterile and synthetic absorbable suture widely used in medical field. It has antibacterial coatings to insure no bacterial infection occurs due to use to suture.

2.1.4 Phosphate Buffered Solution (PBS)

The phosphate buffer solution used was a water based salt solution which has a controlled mixture of various salts like ACS reagent grade dibasic sodium phosphate, and monobasic potassium phosphate. The phosphate buffer solution used had a
concentration of 0.1M and pH of 7.4±0.01 at 25°C. The solution was bought from Ricca Chemical Company (Arlington, TX). This solution helps to simulate the pH of the human body and also the osmolarity and ion concentrations of the human body.

2.2 Sample Preparation

2.2.1 Extrusion

The material was blended and extruded using Leistritz twin screw extruder (model # ZSE18HP-400). The extruder has eight heating zones and it was heated from 50-220°C. The temperature of the extruder heating chambers was controlled by water cooling from the chiller. The chiller temperature was set to 26°C set point to control the temperature and protect material from degrading due to high temperature. The vacuum in the zone seven was turned on so any air or vapor released by materials will be sucked out and there will be no bubbles in the fibers. The extruder speed was set to 20rpm for slow and good mixing. The extruder was starve fed which meant only a small amount of material is fed and passes through some zones before another batch of material is fed. This process was necessary because the twin screw extruder has mixing zones which will push materials up and down and front and back. The twin screw extruder is continuously fed, it will be stuck as the material is pushed backward by the screws where new material is present, leading to poor mixing. The material was in the molten state as it comes out of the fiber die which was pulled by the roller setup. The molten fibers pass through the water bath to make sure the fibers are cooled, as air cooling was not sufficient and the fibers would stick to each other. The roller speed was set to 80rpm. The speed of the roller helped to stretch the molten material from the die
exit. The rate of speed of roller determines the thickness of the fiber. The faster the roller, the more the molten materials get stretched before cooling resulting in finer fibers and vice-versa. The fiber dimensions can also be controlled by the speed of the extruder. Higher the extruder speed means higher the feed rate so there is more molten material at the die exit decreasing the ability to stretch the drawn fibers. For all the samples investigated the speed of roller and the extruder speed were kept constant to get similar diameter fibers to compare. The process of extrusion is show below in Figure 2.3.

![Figure 2.3. Fiber extrusion.](image)

2.2.2 Electrospinning

The PCL and PLLA were mixed with chloroform at 30% concentration/weight at 40°C and stirred with magnetic stirrer for an hour. Then 10ml of acetone per 100ml of solution was also added in the solution. The solution was then transferred to the syringe. The syringe used was 6cc Luer Lock from Small Parts (Miramar, FL). The needle fitted on the syringe was 18 gauge(1.27mm) and 1” long stainless steel blunt needle with a Luer polypropylene hub. The Luer Lock of the syringe and Luer
polypropylene hub was properly locked to avoid unwanted spillage due to pressure from the viscous solution inside. The syringe was then pumped by a syringe pump from (Razel Scientific Instruments, St. Albans, VT). The syringe pump dial was set to 18 which is 4.58cc/hr of flow rate for the syringe used. The tip of the needle was charged at 20KV voltage of DC current using high voltage power supply from Gamma High Voltage Research Inc. (Ormond Beach, FL). The roller was covered with non stick aluminum foil which formed the collector. This collector was carefully grounded and kept at distance of 15cm from the tip of the needle. These conditions of electrospinning (20KV voltage, 15cm distance and 4.58 cc/hr of flow rate) were chosen among various variable because all the blends can be formed into fibers using this condition.

As the syringe pump pumps the solution from the syringe to the tip of the needle it forms a cone like structure called Taylor cone due to electrostatic force between the tip of the needle and the roller [85]. Then the syringe pump and high voltage supply is turned on to get PCL fibers on the roller. The diameter of fibers depends on the applied voltage, distance of collector plate, concentration of solution and solution’s electrical properties [86,87,88].The electrical property of the solution is determined by dielectric constant of the solvents used. The higher the dielectric constant the electrical conductivity increases because it disassociates in solution to form positive and negative particles [89]. The higher dielectric constant in the solution results in better electrospun fibers. The fibers tend to spread more and dry quicker with the addition of acetone in the solution. The dielectric constants of chloroform and acetone are 4.8 and 21 respectively at room temperature (20°C). Since PCL and PLLA dissolves in both chloroform and acetone. Acetone and chloroform mixe with each other, so solvents did not phase
separate. So the addition of acetone was used to enhance electrospinning of the chloroform solution to increase the dielectric constant. The process of electrospinning is shown below in Figure 2.4.

![Diagram of electrospinning setup.](image)

Figure 2.4. Diagram of electrospinning setup.

The fibers were made from blend of PCL and PLLA in the ratio of 100:0, 75:25, 50:50, 25:75 and 0:100. The fiber from polypropylene was also made for a control system as it does not degrade in the body. The samples made from extrusion and electrospun methods are listed in the Table 2.1.
Table 2.1. Samples from extrusion and electrospun.

<table>
<thead>
<tr>
<th>Samples</th>
<th>Sample Code</th>
<th>Extruded</th>
<th>Electrospun</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pure PCL</td>
<td>PCL</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>75% PCL + 25% PLLA</td>
<td>75PCL 25PLLA</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>50% PCL + 50% PLLA</td>
<td>50PCL 50PLLA</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>25% PCL + 75% PLLA</td>
<td>25PCL 75PLLA</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>Pure PLLA</td>
<td>PLLA</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>Polyproplene (PP)</td>
<td>PP</td>
<td>Yes</td>
<td>No</td>
</tr>
</tbody>
</table>

2.2.3 Knitting

Pure PCL was used for the mesh which was knitted in a Silver Reed SK155 9mm chunky punch card knitting machine. The extruded PCL fibers in the roll were fed to the machine which was knitted to make the mesh. The fibers were loaded on a tension mast through a tension pole to the feeder of carriage of knitting machine. The meshes were knitted with three different tension settings set by the machine (Tension 5, Tension 7 and Tension 9). The mesh was held by the claw weight during knitting to prevent warping of the mesh. A 4” x 6” mesh was made of for tensile testing. The sample codes are given below in Table 2.2. The knitting machine and its needles are shown below in Figure 2.5.
Table 2.2. Sample codes for knitted mesh.

<table>
<thead>
<tr>
<th>Sample</th>
<th>Code</th>
</tr>
</thead>
<tbody>
<tr>
<td>PCL mesh sample at Tension 5</td>
<td>Tension 5</td>
</tr>
<tr>
<td>PCL mesh sample at Tension 7</td>
<td>Tension 7</td>
</tr>
<tr>
<td>PCL mesh sample at Tension 9</td>
<td>Tension 9</td>
</tr>
</tbody>
</table>

Figure 2.5. Knitting equipment: (A) silver reed knitting machine; (B) needles.
2.3 Methods

2.3.1 Contact Angle

The sessile drop method of measuring contact angle was used to measure the contact angle. The method is simply measuring the angle of pure water on the solid surface using the optical goniometer. The Rame-hart NRL C.A. Goniometer (Model #100-00) was employed for contact angle measurement. The pure water was dropped on the solid polymer film with the syringe in front of the optical lens of the goniometer to measure the contact angle. The contact angle was measured to compare the wettability of the PCL and PLLA and their blends.

2.3.2 Differential Scanning Calorimetry (DSC)

Differential scanning calorimetry was performed using Perkin Elmer DSC 6. The sample was kept at 20°C for five minutes before heating. Then it was heated at the rate of 10°C per minute in an inert nitrogen atmosphere. The amount of heat required to raise the temperature of the sample is measured to the empty container as a reference. The reference empty container has well known heat capacity so it can be compared with the sample. The DSC was done to observe the thermal properties and crystallinity of the polymer blends. The crystallinity ($X_c$) of the polymer was calculated using the formula below.

$$X_c = \frac{\Delta H_{\text{exp}}}{\Delta H} \times 100\%$$

Where, $\Delta H_{\text{exp}}$ = Experimental heat of crystallization

$\Delta H$ = Theoretical experimental heat of crystallization 100% crystalline polymer
2.3.3 Electron Microscopy

An environmental scanning electron microscope (ESEM) FEI Quanta 200 was used for electron microscopy. The environmental mode was used for microscopy in which the pressure was maintained at 0.45 Torr with some water particles. The water particles were in the chamber to reduce the charging of the insulating polymers due to the electron beam in electron microscopy. The low voltage (10KV) was used to reduce the electron beam damage.

2.3.4 In Vitro Degradation

In vitro degradation was performed using ASTM F1635-04a standard. It is the standard test method for invitro degradation testing of hydrolytically degradable polymer resin and fabricated forms of surgical implants. The samples were soaked in the phosphate buffered solution (PBS) at the physiologic temperature i.e. 37°C [90]. The PBS used was 0.1M phosphate buffer with a pH of 7.4. The solution was poured in the test tube containing samples. The test tube used has a cap to ensure no foreign materials go into the solution while soaking and to eliminate the loss of solution due to evaporation. Multiple samples were put in the same test tube for soaking with suitable seperation for fluid contact. During the test the pH was maintained at 7.4±0.2, so the samples were soaked in at least 50:1 ratio of phosphate buffered solution to sample. The standard suggest at least 20:1 ratio but at lower ratios, the pH of the solution tends to change faster. If the pH of the solution is allowed to drift above or below the specified range the test needs to be terminated. So a higher ratio is suggested till 100:1 so that the pH will be more stable. Also to maintain the pH of the solution within acceptable
limits, and to avoid bacterial contamination, the phosphate buffered solution was replaced regularly. The pH of the solution was monitored regularly by a pH meter from Oakton Acorn series pH6 (serial # 1371669). The accuracy of the pH meter was ±0.01pH and resolution is 0.01pH which is better than the ASTM requirement of ±0.02pH accuracy. The pH meter was regularly calibrated by standard 3 point calibration method with standard pH buffers 4.01; 7.00; and 10.00. The test tubes containing the samples and phosphate buffered solution was stored in a heated air oven at the temperature of 37±2 °C which is the physiologic temperature defined by the ASTM standard. Also the solution was monitored visually for any sign of contamination or cloudyness regularly.

2.3.5 Water Absorption

The samples absorbed water in the matrix as they were immersed in the phosphate buffered solution for weeks. The absorption of water shows the bulk hydrophilicity of the polymer. The samples were taken off the tubes containing phosphate buffered solution and were wiped clean with the tissue paper to remove the excess water on the surface. Then the samples were weighed in the weight balance (Adam Lab AAA analytical balance, model# AAA160DL). The percent water absorption was calculated using formula below. Once the sample been removed from the alkaline phosphate buffered solution it was not reimmersed for further testing.

\[
\text{Percent Water Absorption (\%) } = \frac{\text{Wet sample weight}}{\text{Dry sample weight}} \times 100 \%
\]
2.3.6 Weight Loss

The samples were weighed using analytical balance AdamLab AAA (model# AAA160DL). It had a three door windsheild to minimize the effect of blowing air while weighing. Its readability is 0.01 mg. The samples were weighed with an accuracy of 0.1% before soaking in the phosphate buffered solution for in vitro degradation. After a specified time of soaking, samples were removed from the phosphate buffered solution and dried. Each removed samples were rinsed with distilled water before drying so that no residue is left due to phosphate buffer solution. It was dried in the vacuum oven at 30°C until it reached a constant weight. The samples which were removed from the solution for weighing was not resoaked in the phosphate buffer solution for further in vitro tests. According to the ASTM standard F1635 at least three samples were used for weight loss per test period.

\[
\text{Percent Weight Loss (\%) = \left( \frac{\text{Dry sample weight}}{\text{Original sample weight}} \right) \times 100 \%}
\]

2.3.7 Density Measurement

The density of the samples were measured using Archimedes principle, where the sample was immersed in liquid. Since the original density of the samples were close to the density of water, ethyl alcohol was used instead of water. The ethyl alcohol has a density of 0.789g/cm³. The samples were measured using the density measurement plugin of the analytical balance AdamLab AAA (model# AAA160DL). The sample was weighed in air and in the liquid. The weight of sample while immersed in the liquid is
known as apparent weight. The density of the samples were calculated using the formula below.

\[
\frac{\text{Density of sample}}{\text{Density of alcohol}} = \frac{\text{Weight of sample in air}}{\text{Weight of sample in air} - \text{Apparent weight}}
\]

2.3.8 Mechanical Testing

The mechanical testing was done by tensile testing. Tensile testing of extruded and electrospun fibers was done on TA instruments RSA3 (DE, USA). The fiber was tested had a 10mm gauge length. The test was performed at the displacement rate of 1mm/min at room temperature. The samples were completely dried before mechanical testing. But mechanical testing of knitted mesh was done in MTS 810 Material Test System (MN, USA). The knitted fibers were also tested at the displacement rate of 1mm/min with a gauge length of 3mm. The knitted fibers had forty ends at the grip of the equipment.
3.1 Contact Angle

The contact angle of poly(ε-caprolactone) (PCL) and polylactide (PLLA) was measured to compare the wettability of the polymers. The lower the contact angle between water and the polymer means the polymer is hydrophilic. The PCL and PLLA has contact angle of 54.33 ± 2.3 and 64.66 ± 2.5 respectively. The PCL is more hydrophilic compared the PLLA. The hydrophilic nature of PCL also influences the wettability of the PCL/PLLA blend. The blend with higher PCL content like 75PCL 25PLLA, the 50PCL 50PLLA sample also indicated a dominance of PCL had contact angle closer to PCL i.e. 55.1 ± 1.9, 57 ± 2.5 respectively. The higher PLLA content sample 25PCL 75PLLA, had a higher contact angle compared to PCL and the blends i.e. 61 ± 1.5. The ratio of the PCL and PLLA in the blend also affects the contact angle hence its wettability.

3.2 Differential Scanning Calorimetry (DSC)

Since PCL and PLLA are immiscible polymer blends, two different melting and crystallization peaks are observed instead of single peaks as in miscible blends. Two immiscible polymers will retain their own melting and crystallization peak independent of the other material in the differential calorimetry thermograms. The effect of processing can be observed in the first heating and cooling of the sample. The second heating and cooling of the sample nullifies the thermal history effect of processing.
3.2.1 Extruded Fibers

The extruded fibers DSC thermograms show the effect of processing by extrusion. The first heat of the extruded fibers show the glass transition temperature peak of the PLLA, melting point of PCL and PLLA and cold crystallization of PLLA of all blends. The first cooling of the extruded fibers shows the re-crystallization peaks of PLLA and PCL. The first heating and first cooling thermograms are given below in Figure 3.1 and Figure 3.2 respectively.

![1st Heat Extruded Fibers](image)

Figure 3.1. 1st heating of extruded fibers.
Comparing the enthalpy of melting and crystallization of extruded PLLA and its blends with PCL, they both show a decrease by the addition of PCL. Similar comparison of change in melting of enthalpy of PCL cannot be done because the glass transition temperature of PLLA overlaps with the melting point of PCL. But the melting temperature of PCL remains constant. The melting, crystallization and glass transition temperature with enthalpy are given below in Table 3.1.
Table 3.1. Extruded fibers differential scanning calorimetry thermograms.

<table>
<thead>
<tr>
<th>Sample</th>
<th>Melting Temperature ($T_m$) (°C)</th>
<th>Melting Enthalpy ($\Delta H_m$) (J/g)</th>
<th>Glass Transition Temperature ($T_g$) (°C)</th>
<th>Crystallization Temperature ($T_c$) (°C)</th>
<th>Crystallization Enthalpy ($\Delta H_c$) (J/g)</th>
<th>Cold Crystallization Temperature ($T_c$) (°C)</th>
<th>Cold Crystallization Enthalpy ($\Delta H_c$) (J/g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>PCL</td>
<td>58.79</td>
<td>49.88</td>
<td></td>
<td>35.8</td>
<td>-53.32</td>
<td></td>
<td></td>
</tr>
<tr>
<td>75PCL</td>
<td>60.23</td>
<td>69.28</td>
<td></td>
<td>36.62</td>
<td>-66.14</td>
<td></td>
<td></td>
</tr>
<tr>
<td>25PLLA</td>
<td>166.29</td>
<td>9.54</td>
<td></td>
<td>96.04</td>
<td>-3.69</td>
<td></td>
<td></td>
</tr>
<tr>
<td>50PCL</td>
<td>60.23</td>
<td>54.94</td>
<td></td>
<td>36.05</td>
<td>-51.39</td>
<td></td>
<td></td>
</tr>
<tr>
<td>50PLLA</td>
<td>166.32</td>
<td>11.32</td>
<td></td>
<td>116.74</td>
<td>-0.22</td>
<td>96.42</td>
<td>-7.19</td>
</tr>
<tr>
<td>25PCL</td>
<td>59.53</td>
<td>43.68</td>
<td></td>
<td>36.02</td>
<td>-51.1</td>
<td></td>
<td></td>
</tr>
<tr>
<td>75PLLA</td>
<td>165.1</td>
<td>19.51</td>
<td></td>
<td>93.2</td>
<td>-2.26</td>
<td>91.8</td>
<td>-10.12</td>
</tr>
<tr>
<td>PLLA</td>
<td>166.9</td>
<td>51.37</td>
<td>58</td>
<td>92.26</td>
<td>-9.34</td>
<td>95.57</td>
<td>-30.32</td>
</tr>
<tr>
<td>PP</td>
<td>164.43</td>
<td>71.68</td>
<td>110.1</td>
<td>114.32</td>
<td>-95.837</td>
<td>-</td>
<td>-</td>
</tr>
</tbody>
</table>

The enthalpy of melting of pure 100% crystalline PLLA is 93 J/g [91,92]. Similarly, enthalpy of melting of pure 100% crystalline PCL is 139.5 J/g [93,94]. Also the enthalpy of melting of pure 100% crystalline PP is 209 J/g [95,96]. So the percent crystallinity of PP, PCL and PLLA extruded fibers is 34.29%, 35.75% and 55.23% respectively. In the case of 75PCL25PLLA blend, percent crystallinility of PCL and PLLA part was 49.84% and 10.25% respectively. Similarly in the case of 50PCL50PLLA blend, the percent crystallinity of PCL and PLLA was 39.25% and 12.17% respectively. Also in the case of 25PCL75PLLA, the percent crystallinity of PCL and polylactdie was 31.42% and 20.97% respectively. The percent crystallinity of PLLA is significantly affected by the presence of PCL in the blend. Clearly, the crystallinity of PLLA is increasing with decreasing PCL content. But the percentage of PLLA does not seem to affect the crystallinity of PCL as it remains almost constant.
3.2.2 Electrospun Fibers

In the case of electrospun fibers, the remaining chloroform is clearly seen during the first heat of fiber DSC. The boiling point of chloroform is 62°C, so a peak is observed in 1st heat of PLLA fibers at 62°C. But in all other blends due to the presence of PCL whose melting point is at 60°C, the PCL melting endotherm tends to merge with the peak of chloroform. The peak assignment to certain and chloroform is not due to contamination of PCL. As can be seen it vaporizes and thus in the second heat plot no peak is seen at 62°C. The 1st cool and 2nd heat differential scanning thermograms are shown below in Figure 3.3, 3.4 and 3.5 respectively. The melting, crystallization and glass transition temperature with enthalpy are given below in Table 3.2.

![1st Heat Electrospun Fibers](image)

**Figure 3.3.** 1st heating of electrospun fibers.
Figure 3.4. 1\textsuperscript{st} cooling thermogram of electrospun fibers.

Figure 3.5. 2\textsuperscript{nd} heating of electrospun fibers.
Table 3.2. Electrospun fibers differential scanning calorimetry thermograms.

<table>
<thead>
<tr>
<th>Sample</th>
<th>Melting Temperature ($T_m$) (°C)</th>
<th>Melting Enthalpy ($\Delta H_m$) (J/g)</th>
<th>Glass Transition Temperature ($T_g$) (°C)</th>
<th>Crystallization Temperature ($T_c$) (°C)</th>
<th>Crystallization Enthalpy ($\Delta H_c$) (J/g)</th>
<th>Cold Crystallization Temperature ($T_{c'}$) (°C)</th>
<th>Cold Crystallization Enthalpy ($\Delta H_{c'}$) (J/g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>PCL</td>
<td>61.05</td>
<td>65.41</td>
<td>-</td>
<td>36.13</td>
<td>-59.23</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>75PCL 25PLLA</td>
<td>62.47</td>
<td>81.12</td>
<td>34.54</td>
<td>-57.91</td>
<td>165.84</td>
<td>2.81</td>
<td>89.27</td>
</tr>
<tr>
<td>50PCL 50PLLA</td>
<td>62.29</td>
<td>70.26</td>
<td>34.12</td>
<td>-40.05</td>
<td>165.63</td>
<td>11.07</td>
<td>83.3</td>
</tr>
<tr>
<td>25PCL 75PLLA</td>
<td>62.38</td>
<td>8.11</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>PLLA</td>
<td>165.74</td>
<td>45.90</td>
<td>95.72</td>
<td>-4.94</td>
<td>95.53</td>
<td>-20.81</td>
<td></td>
</tr>
</tbody>
</table>

Since the peak of the melting temperature of PCL and remaining chloroform tends to merge. Calculating crystallinity of PCL accurately is impossible. But the crystallinity of PLLA can be calculated accurately. The percent crystallinity of PLLA in PLLA, 25PCL75PLLA, 50PCL50PLLA and 75PCL25PLLA is 49.35%, 38.24%, 11.9% and 3.02% respectively. This is significantly lower than the extruded samples.

3.3 Scanning Electron Microscopy (SEM)

The SEM was employed to observe fibers made by extrusion and electrospinning process and the bioresorbable suture. The microscopy was used to observe the surface of the fibers and the cross section of the fibers. The images were analyzed using ImageJ software. The surface and the cross sectional images were used to calculate the average diameter of the extruded fibers. The average diameter of electrospun fibers was calculated by the surface images.
The images of the extruded and electrospun PCL fibers are shown below in Figure 3.6. The average diameter of the extruded fibers was 498 ± 30 µm. The average diameter of the electrospun fibers was 14 ± 4 µm.

Figure 3.6. Images of pure PCL fibers: (A) surface of extruded fibers; (B) cross section of extruded fibers; (C) electrospun fibers.

The images of the extruded and electrospun pure PLLA fibers are shown below in Figure 3.7. In the electrospun fibers some of the fibers in the background are mixed
while they are in the solution, so they appear to be bigger in the diameter. The average diameter of the extruded fibers was $463 \pm 12 \, \mu m$. The average diameter of the electrospun fibers was $17 \pm 6 \, \mu m$.

![Image of pure PLLA fibers: (A) surface of extruded fibers; (B) cross section of extruded fibers; (C) electrospun fibers.](image)

Figure 3.7. Images of pure PLLA fibers: (A) surface of extruded fibers; (B) cross section of extruded fibers; (C) electrospun fibers.

Similarly, the images of the extruded and electrospun 75PCL and 25PLLA fibers are shown below in Figure 3.8. In the cross section image, the phase separation of PCL
and PLLA can be observed. The separation of PLLA from the PCL matrix is indicated by the appearance of a separate round phase in the cross sectional image of the fibers. In the electrospun fibers some of the fibers in the background are mixed while they are in the solution, so they appear to be bigger in the diameter. The average diameter of the extruded fibers was $454 \pm 22 \, \mu m$. The average diameter of the electrospun fibers was $22 \pm 9 \, \mu m$.

Figure 3.8. Images of 75PCL 25PLLA fibers: (A) surface of extruded fibers; (B) cross section of extruded fibers; (C) electrospun fibers.
The images of the extruded and electrospun 50PCL and 50PLLA fibers are shown below in Figure 3.9. As in 75PCL 25PLLA sample the phase separation of PCL and PLLA can also be observed in the cross section image of 50PCL 50PLLA sample. In the electrospun fibers some of the fibers in the background are mixed while they are in the solution, so they appear to be bigger in the diameter. The average diameter of the extruded fibers was 481 ± 47 µm. The average diameter of the electrospun fibers was 16 ± 7 µm.

Figure 3.9. Images of 50PCL 50PLLA fibers: (A) surface of extruded fibers; (B) cross section of extruded fibers; (C) electrospun fibers.
The images of the extruded and electrospun 25PCL and 75PLLA fibers are shown below in Figure 3.10. As in 75PCL 25PLLA and 50PCL 50PLLA sample the phase separation of PCL and PLLA can also be observed in cross section image of 25PCL 75PLLA sample too. In this case the phase separation of PCL and PLLA is also clearly visible in surface image. The average diameter of the extruded fibers was 422 ± 41 µm. The average diameter of the electrospun fibers was 16 ± 2 µm.

Figure 3.10. Images of 25PCL 75PLLA fibers :(A) surface of extruded fibers; (B) cross section of extruded fibers; (C) electrospun fibers.
The images of the extruded pure polypropylene fibers are shown below in Figure 3.11. The average diameter of the extruded fibers was 472.06 ± 5.6 µm.

![Figure 3.11. Images of pure polypropylene fibers](image)

(A) Surface of extruded fibers; (B) cross section of extruded fibers.

The images of the bioresorbable suture called Vicryl™ are shown below in Figure 3.12. The images clearly show the breaded fibers and its alignment in the suture. The average diameter of the suture was 328.63 ± 24.3 µm.
Figure 3.12. Images of bioresorbable suture: (A) surface of suture; (B) cross section of bioresorbable suture.

All the diameters of the extruded fibers are compared in the Figure 3.13 below. The condition of extrusion (roller and screw speed) for all fiber extrusion is same, the fiber diameter can be compared, but the small variance of diameters is caused by melt viscosity of each blends and polymers and the temperature at which it was processed.

![Extruded Fibers](image)

Figure 3.13. Diameter comparisons of extruded fibers.
The diameters of the electrospun fibers are compared in the Figure 3.14 below. The fibers were prepared using same condition (i.e. distance, voltage and flow rate) which contributes to the size of fibers. The diameters of fibers remain comparable to each other in all blends of different ratio of polymers.

![Electrospun Fibers](image)

Figure 3.14. Diameter comparisons of electrospun fibers.

### 3.4 In Vitro Degradation

The degradation of fibers was observed by the surface and cross sectional morphology of the fibers. This morphology was observed using images of scanning electron microscope. The SEM images to compare the degradation of the sample during eight weeks are given below in Figure 3.15.
<table>
<thead>
<tr>
<th>Samples</th>
<th>0 Week</th>
<th>8 Week</th>
</tr>
</thead>
<tbody>
<tr>
<td>PCL</td>
<td><img src="image1" alt="Image" /></td>
<td><img src="image2" alt="Image" /></td>
</tr>
<tr>
<td>75PCL 25PLLA</td>
<td><img src="image3" alt="Image" /></td>
<td><img src="image4" alt="Image" /></td>
</tr>
<tr>
<td>50PCL 50PLLA</td>
<td><img src="image5" alt="Image" /></td>
<td><img src="image6" alt="Image" /></td>
</tr>
<tr>
<td>25PCL 75PLLA</td>
<td><img src="image7" alt="Image" /></td>
<td><img src="image8" alt="Image" /></td>
</tr>
<tr>
<td>PLLA</td>
<td><img src="image9" alt="Image" /></td>
<td><img src="image10" alt="Image" /></td>
</tr>
</tbody>
</table>
Figure 3.15. SEM images of samples at 0 and 8 weeks.

The PCL has no change of morphology on the surface or in cross sectional images of PCL after eight weeks of degradation study as shown in Figure 3.15. Due to addition of PLLA in 75PCL 25PLLA sample some separation of two phases (PCL and PLLA) appears after eight weeks of degradation. This separation of phases is also seen in 50PCL 50PLLA samples. But in the case of 25PCL 75PLLA there is significant cracking seen in the SEM images.

In the case of PLLA and PP, there is no change of morphology of the surface or in cross sectional images of PLLA and PP after eight weeks of degradation study as shown in Figure 3.15. But the bioabsorbable suture had significant degradation effect clearly seen in the eight week degradation study. The bioresorbable suture broke off its braided structure during eight week of immersion in PBS which is clearly visible from its surface and cross sectional images shown above in Figure 3.15.

3.5 Water Absorption
The water absorption of the sample shows its hydrophilicity. As the sample absorbs water, a significant portion of the surface of the sample is exposed to alkaline phosphate buffered solution which causes hydrolysis in polymers. So high water absorption causes faster degradation of the polymer. In the case of PCL, the water absorption is very low; this complements its hydrophobicity and high crystallinity. The total water absorbed by PCL during the eight weeks of continuous immersion in phosphate buffered solution was 0.9 ± 0.2 %. The trend of water absorption for PCL fibers is shown below in Figure 3.16.

![PCL Water Absorption Graph](image)

**Figure 3.16.** PCL water absorption.

The water absorption of pure PLLA (PLLA) is higher compared to PCL, 75PCL 25PLLA, 50PCL 50PLLA and 25PCL 75PLLA. The total water absorbed by pure PLLA during eight week of continuous immersion in phosphate buffered solution was 10 ± 2%. The trend of water absorption for pure PLLA fibers is shown in Figure 3.17.
The water absorption of 75PCL 25PLLA is slightly higher compared to its pure counter part of PCL. The total water absorbed by 75PCL 25PLLA during eight weeks of continuous immersion in phosphate buffered solution was 1.5 ± 0.5%. The trend of water absorption for 75PCL 25PLLA fibers is shown in Figure 3.18.

Figure 3.17. PLLA water absorption.
The water absorption of 50PCL 50PLLA is slightly higher compared to PCL and 75PCL 25PLLA. The total water absorbed by 50PCL 50PLLA during eight week of continuous immersion in phosphate buffered solution was 3 ± 1 %. The trend of water absorption for 50PCL 50PLLA fibers is shown in Figure 3.19.
The water absorption of 25PCL 75PLLA is higher compared to PCL, 75PCL 25PLLA and 50PCL 50PLLA. The total water absorbed by 25PCL 75PLLA during eight week of continuous immersion in phosphate buffered solution was 6 ± 1%. The trend of water absorption for 25PCL 75PLLA fibers is shown in Figure 3.20.
The water absorption of PP fibers is negligible compared to PCL, 75PCL 25PLLA, 50PCL 50PLLA, 25PCL 75PLLA and PLLA fiber. The total water absorbed by pure PP during the eight weeks of continuous immersion in phosphate buffered solution was 0.5 ± 0.1%. The trend of water absorption for pure PP fibers is shown in Figure 3.21.
The water absorption of bioresorbable suture fibers is very high compared to PCL, 75PCL 25PLLA, 50PCL 50PLLA, 25PCL 75PLLA, PLLA and polypropylene fiber. The total water absorbed by bioresorbable suture fibers during the eight weeks of continuous immersion in phosphate buffered solution was 36 ± 4%. The trend of water absorption for bioresorbable suture fibers is shown in Figure 3.22.
Figure 3.22. Bioresorbable suture water absorption.

The entire sample’s water absorption is compared in Figure 3.23. It shows that bioresorbable suture absorbs the most water absorbed; on the other hand PP absorbed the least. Among the aliphatic polyesters PCL and PLLA), PLLA absorbed more water than PCL. PLLA absorbed almost ten times more water than the PCL. In the blend of these polymers as the ratio of PLLA increased more and more water is absorbed.
Figure 3.23. Water absorption: (A) all samples water absorption; (B) PCL/PLLA blend water absorption; (C) PCL/PLLA blend water absorption for 4 weeks.

3.6 Percent Weight Loss

The weight loss of sample shows the loss of soluble oligomers during degradation. The soluble oligomers are formed due to presence of alkaline phosphate buffered solution. The alkaline phosphate buffered solution causes hydrolysis of the aliphatic polyester through chain scission. The loss of weight is independent to each sample and they degrade at their own degradation rate. The degradation rate of samples depends upon the chemistry of the polymer and type of blends.
The extruded and electrospun PCL loses some weight during eight week degradation. The extruded and electrospun PCL fibers lost about 4.5 ± 0.93% and 5 ± 0.63% of weight respectively during eight weeks of degradation. The trend of loss of weight of PCL fibers is shown in Figure 3.24.

![Figure 3.24. Weight loss of PCL.](image)

The extruded and electrospun PLLA lost some significant weight during eight week degradation. The extruded and electrospun PLLA fibers lost about 9.4 ± 0.96% and 10.61 ± 0.48% of weight respectively during the eight weeks of degradation. The trend of loss of weight of PLLA fibers is shown in Figure 3.25.
The extruded and electrospun 75PCL 25PLLA lost some weight during eight week degradation. The extruded and electrospun 75PCL 25PLLA fibers lost about 5.57 ± 0.92% and 5.79 ± 0.65% of weight respectively during the eight weeks of degradation. The trend of loss of weight of 75PCL 25PLLA fibers is shown in Figure 3.26.
Figure 3.26. Weight loss of 75PCL 25PLLA.

The extruded and electrospun 50PCL 50PLLA lost some weight during eight week degradation. The extruded and electrospun 50PCL 50PLLA fibers lost about 6.98 ± 0.75% and 7.18 ± 0.37% of weight respectively the during eight weeks of degradation. The trend of loss of weight of 50PCL 50PLLA fibers is shown in Figure 3.27.
The extruded and electrospun 25PCL 75PLLA lost some weight during eight week degradation. The extruded and electrospun 25PCL 75PLLA fibers lost about 7.25 ± 0.88% and 7.42 ± 0.98% of weight respectively during eight weeks of degradation. The trend of loss of weight of 25PCL 75PLLA fibers is shown in Figure 3.28.
Figure 3.28. Weight loss of 25PCL 75PLLA.

In the case of extruded PP there was no significant loss of weight during eight weeks of in vitro degradation test. The weight of polypropylene fluctuates slightly but weight loss is negligible. The eight weeks weight loss of polypropylene is show below in Figure 3.29.
Figure 3.29. Weight loss of PP.

The bioresorbable suture fibers lost lot of weight during the eight weeks degradation. The fibers lost about \(37.33 \pm 2.51\%\) during the eight weeks of degradation. The trend of loss of weight of bioresorbable suture fibers is shown in Figure 3.30.
Figure 3.30. Weight loss of bioresorbable suture.

The weight losses of all the samples are compiled in the Figure 3.31. It clearly shows the difference of weight loss of each sample during the eight weeks of degradation. Clearly the bioresorbable suture lost the most of the weight as it is designed to lose weight after three weeks. The polypropylene fibers did not lose any significant weight, which is the main problem in using polypropylene mesh for tissue scaffolding. In the family of aliphatic polyester the PLLA lost more weight than PCL simply because the PLLA absorbed more solution and more of the PLLA chains were in contact with alkaline phosphate buffered solution. More the surface area it has exposed to alkaline phosphate buffered solution more weight it loses by forming soluble oligomers.
Figure 3.31. Weight loss: (A) comparisons of all fibers; (B) PCL/PLLA blend comparison.
3.7 Density Measurement

The density measurement was done to observe the change in density of the sample during degradation. The density of the sample measured using Archimedes principle did not show much change in the density in any samples. The PCL loses weight but it had not absorbed any PBS so it only lost mass in the surface. But the PLLA and its blends lose weight but they absorbed liquid which nullified the density measurements. The PP samples did not lose any weight and did not absorb any liquid. The density measurement is shown below in the Table 3.3.

Table 3.3. Density measurements.

<table>
<thead>
<tr>
<th>Samples</th>
<th>0 Week Density (g/cm$^3$)</th>
<th>4 Week Density (g/cm$^3$)</th>
<th>8 Week Density (g/cm$^3$)</th>
</tr>
</thead>
<tbody>
<tr>
<td>PCL</td>
<td>1.01 ± 0.5</td>
<td>0.99 ± 0.7</td>
<td>1.03 ± 0.6</td>
</tr>
<tr>
<td>75PCL 25PLLA</td>
<td>1.02 ± 0.7</td>
<td>1.07 ± 0.3</td>
<td>1.01 ± 0.4</td>
</tr>
<tr>
<td>50PCL 50PLLA</td>
<td>1.04 ± 0.4</td>
<td>1.09 ± 0.4</td>
<td>1.1 ± 0.7</td>
</tr>
<tr>
<td>25PCL 75PLLA</td>
<td>1.1 ± 0.5</td>
<td>1.07 ± 0.8</td>
<td>1.12 ± 0.4</td>
</tr>
<tr>
<td>PLLA</td>
<td>1.24 ± 0.9</td>
<td>1.17 ± 0.6</td>
<td>1.19 ± 0.7</td>
</tr>
<tr>
<td>PP</td>
<td>0.8 ± 0.2</td>
<td>0.87 ± 0.4</td>
<td>0.9 ± 0.3</td>
</tr>
</tbody>
</table>

3.8 Mechanical Testing

The mechanical testing was done to observe the effect on mechanical properties due to in vitro degradation. The loss of mechanical properties was due to main chain scission in aliphatic polyesters (PCL and PLLA) but in blends of PCL and PLLA the separation of different phases also caused loss of mechanical properties.
3.8.1 Extruded Fibers

The tensile tests of the extruded fibers were done of all blends each week during in vitro degradation for eight weeks except for 75PLLA 25PCL sample. The 75PLLA 25PCL sample was degraded and was not in a condition to be tensile tested after four weeks of degradation. Figure 3.32 below shows the tensile test of the single extruded fiber.

![Tensile testing of the extruded fiber sample.](image)

The complete reports of the tensile test of all the extruded samples are shown in the Table 3.4 to Table 3.11 below.
Table 3.4. Extruded fibers week 1 report.

<table>
<thead>
<tr>
<th>Sample</th>
<th>Elastic Modulus (MPa)</th>
<th>Tensile Strength (MPa)</th>
<th>Strain at Failure (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pure PCL</td>
<td>2.4 ± 0.38</td>
<td>18.15 ± 2.12</td>
<td>-</td>
</tr>
<tr>
<td>75PCL 25PLLA</td>
<td>4.56 ± 0.97</td>
<td>21.82 ± 1.46</td>
<td>7.08 ±0.22</td>
</tr>
<tr>
<td>50PCL 50PLLA</td>
<td>3.29 ± 0.6</td>
<td>14.99 ± 4.56</td>
<td>4.63 ± 0.84</td>
</tr>
<tr>
<td>25PCL 75PLLA</td>
<td>13.14 ± 1.41</td>
<td>11.04 ± 1.65</td>
<td>1.15 ± 0.13</td>
</tr>
<tr>
<td>Pure PLLA</td>
<td>19.52 ± 2.42</td>
<td>51.97 ± 1.42</td>
<td>37.9 ± 30.63</td>
</tr>
<tr>
<td>Pure PP</td>
<td>9.95 ± 0.865</td>
<td>30 ± 1.33</td>
<td>-</td>
</tr>
</tbody>
</table>

Table 3.5. Extruded fibers week 2 report.

<table>
<thead>
<tr>
<th>Sample</th>
<th>Elastic Modulus (MPa)</th>
<th>Tensile Strength (MPa)</th>
<th>Strain at Failure (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pure PCL</td>
<td>4.36 ± 0.34</td>
<td>20.2 ± 1.02</td>
<td>-</td>
</tr>
<tr>
<td>75PCL 25PLLA</td>
<td>5.73 ± 0.52</td>
<td>18.59 ± 0.75</td>
<td>5.77 ± 0.7</td>
</tr>
<tr>
<td>50PCL 50PLLA</td>
<td>5.86 ± 0.54</td>
<td>16.86 ± 2.6</td>
<td>3.67 ± 0.42</td>
</tr>
<tr>
<td>25PCL 75PLLA</td>
<td>6.29 ± 2.4</td>
<td>5.3 ± 2.5</td>
<td>0.81 ± 0.24</td>
</tr>
<tr>
<td>Pure PLLA</td>
<td>17.35 ± 1.77</td>
<td>46 ± 1.74</td>
<td>39.94 ± 25</td>
</tr>
<tr>
<td>Pure PP</td>
<td>9.09 ± 0.99</td>
<td>31.54 ± 1.54</td>
<td>-</td>
</tr>
</tbody>
</table>

Table 3.6. Extruded fibers week 3 report.

<table>
<thead>
<tr>
<th>Sample</th>
<th>Elastic Modulus (MPa)</th>
<th>Tensile Strength (MPa)</th>
<th>Strain at Failure (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pure PCL</td>
<td>2.62 ± 0.35</td>
<td>19.18 ± 0.59</td>
<td>-</td>
</tr>
<tr>
<td>75PCL 25PLLA</td>
<td>5.4 ± 0.37</td>
<td>20.93 ± 1.4</td>
<td>5.95 ± 0.61</td>
</tr>
<tr>
<td>50PCL 50PLLA</td>
<td>4.9 ± 0.44</td>
<td>16.32 ± 3.5</td>
<td>3.95 ± 0.8</td>
</tr>
<tr>
<td>25PCL 75PLLA</td>
<td>13.9 ± 3.89</td>
<td>8.02 ± 1.32</td>
<td>0.92 ± 0.23</td>
</tr>
<tr>
<td>Pure PLLA</td>
<td>16.35 ± 1.11</td>
<td>46.19 ± 3.09</td>
<td>56.49 ± 55.19</td>
</tr>
<tr>
<td>Pure PP</td>
<td>9.95 ± 0.75</td>
<td>29.14 ± 0.98</td>
<td>-</td>
</tr>
</tbody>
</table>
Table 3.7. Extruded fibers week 4 report.

<table>
<thead>
<tr>
<th>Sample</th>
<th>Elastic Modulus (MPa)</th>
<th>Tensile Strength (MPa)</th>
<th>Strain at Failure (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pure PCL</td>
<td>3.43 ± 0.24</td>
<td>24.98 ± 1.68</td>
<td>-</td>
</tr>
<tr>
<td>75PCL 25PLLA</td>
<td>5.84 ± 3.39</td>
<td>15.24 ± 3.31</td>
<td>6.11 ± 0.58</td>
</tr>
<tr>
<td>50PCL 50PLLA</td>
<td>6.03 ± 1.34</td>
<td>11.96 ± 2.68</td>
<td>3.26 ± 0.05</td>
</tr>
<tr>
<td>25PCL 75PLLA</td>
<td>4.63 ± 0.82</td>
<td>1.96 ± 0.22</td>
<td>0.399 ± 0.12</td>
</tr>
<tr>
<td>Pure PLLA</td>
<td>14.87 ± 2.71</td>
<td>50.53 ± 3.38</td>
<td>23.02 ± 17.27</td>
</tr>
<tr>
<td>Pure PP</td>
<td>10.82 ± 1.03</td>
<td>29.34 ± 0.74</td>
<td>-</td>
</tr>
</tbody>
</table>

Table 3.8. Extruded fibers week 5 report.

<table>
<thead>
<tr>
<th>Sample</th>
<th>Elastic Modulus (MPa)</th>
<th>Tensile Strength (MPa)</th>
<th>Strain at Failure (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pure PCL</td>
<td>2.36 ± 0.34</td>
<td>21.72 ± 1.71</td>
<td>-</td>
</tr>
<tr>
<td>75PCL 25PLLA</td>
<td>7.06 ± 1.36</td>
<td>19.89 ± 2.9</td>
<td>4.7 ± 0.95</td>
</tr>
<tr>
<td>50PCL 50PLLA</td>
<td>5.69 ± 1.29</td>
<td>10.46 ± 2.57</td>
<td>2.28 ± 0.82</td>
</tr>
<tr>
<td>25PCL 75PLLA</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Pure PLLA</td>
<td>8.79 ± 0.49</td>
<td>22.27 ± 0.414</td>
<td>31.19 ± 30.28</td>
</tr>
<tr>
<td>Pure PP</td>
<td>6.04 ± 0.85</td>
<td>22.82 ± 0.89</td>
<td>-</td>
</tr>
</tbody>
</table>

Table 3.9. Extruded fibers week 6 report.

<table>
<thead>
<tr>
<th>Sample</th>
<th>Elastic Modulus (MPa)</th>
<th>Tensile Strength (MPa)</th>
<th>Strain at Failure (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pure PCL</td>
<td>2.46 ± 0.24</td>
<td>20.82 ± 0.36</td>
<td>-</td>
</tr>
<tr>
<td>75PCL 25PLLA</td>
<td>6.96 ± 1.93</td>
<td>22.83 ± 3.88</td>
<td>5.56 ± 0.46</td>
</tr>
<tr>
<td>50PCL 50PLLA</td>
<td>4.05 ± 0.45</td>
<td>7.45 ± 0.54</td>
<td>2.14 ± 0.21</td>
</tr>
<tr>
<td>25PCL 75PLLA</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Pure PLLA</td>
<td>7.5 ± 0.18</td>
<td>19.33 ± 4.85</td>
<td>22.58 ± 11.94</td>
</tr>
<tr>
<td>Pure PP</td>
<td>5.95 ± 0.43</td>
<td>20.57 ± 1.02</td>
<td>-</td>
</tr>
</tbody>
</table>
Table 3.10. Extruded fibers week 7 report.

<table>
<thead>
<tr>
<th>Sample</th>
<th>Elastic Modulus (MPa)</th>
<th>Tensile Strength (MPa)</th>
<th>Strain at Failure (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pure PCL</td>
<td>3.13 ± 0.17</td>
<td>23.3 ± 0.965</td>
<td>-</td>
</tr>
<tr>
<td>75PCL 25PLLA</td>
<td>5.28 ± 0.37</td>
<td>16.09 ± 0.55</td>
<td>4.62 ± 0.23</td>
</tr>
<tr>
<td>50PCL 50PLLA</td>
<td>5.22 ± 0.322</td>
<td>6.73 ± 0.27</td>
<td>1.62 ± 0.08</td>
</tr>
<tr>
<td>25PCL 75PLLA</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Pure PLLA</td>
<td>4.17 ± 0.35</td>
<td>15.23 ± 0.95</td>
<td>23.76 ± 10.08</td>
</tr>
<tr>
<td>Pure PP</td>
<td>5.79 ± 0.35</td>
<td>20.98 ± 1.67</td>
<td>-</td>
</tr>
</tbody>
</table>

Table 3.11. Extruded fibers week 8 report.

<table>
<thead>
<tr>
<th>Sample</th>
<th>Elastic Modulus (MPa)</th>
<th>Tensile Strength (MPa)</th>
<th>Strain at Failure (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pure PCL</td>
<td>2.87 ± 0.29</td>
<td>22.35 ± 1.5</td>
<td>-</td>
</tr>
<tr>
<td>75PCL 25PLLA</td>
<td>4.97 ± 0.15</td>
<td>15.97 ± 0.36</td>
<td>4.76 ± 0.23</td>
</tr>
<tr>
<td>50PCL 50PLLA</td>
<td>5.03 ± 0.16</td>
<td>6.47 ± 0.38</td>
<td>1.26 ± 0.05</td>
</tr>
<tr>
<td>25PCL 75PLLA</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Pure PLLA</td>
<td>4.02 ± 0.1</td>
<td>13.45 ± 2.72</td>
<td>21.84 ± 10.27</td>
</tr>
<tr>
<td>Pure PP</td>
<td>5.39 ± 0.45</td>
<td>19.56 ± 1.45</td>
<td>-</td>
</tr>
</tbody>
</table>

The stress vs. strain plot of the tensile tests of extruded PCL fiber during eight weeks of degradation is shown below in Figure 3.33. The PCL fiber property does not change at all as its weight loss and water absorption were also to the minimum. Also there were no visible defects seen in the electron microscope images during eight weeks of degradation. The PCL fiber keeps on stretching to almost 500% and more so the test was stopped before that.
The tensile strength and elastic modulus of the PCL fibers also remained almost constant during the eight weeks of degradation with minor variance with acceptable standard of deviation. The tensile strength and elastic modulus of PCL fibers during the eight weeks are shown below in Figure 3.34 and Figure 3.35 respectively.

Figure 3.33. Stress vs strain plot of extruded PCL.

Figure 3.34. Tensile strength of extruded PCL.
The stress vs. strain plot of the tensile tests of extruded PLLA fiber during the eight weeks of degradation is shown below in Figure 3.36. The PLLA fiber property degrades a lot after four weeks in vitro degradation. The degradation is not clearly seen in the electron microscope images since there was no PCL to create gaps or cracks.

Figure 3.36. Stress vs strain plot of extruded PLLA.
The PLLA fibers lost tensile strength and elastic modulus significantly during the eight weeks of degradation. The tensile strength and elastic modulus of PLLA fibers during the eight weeks are shown below in Figure 3.37 and Figure 3.38 respectively.

![Tensile Strength of PLLA](image)

**Figure 3.37.** Tensile strength of extruded PLLA.

![Elastic Modulus of PLLA](image)

**Figure 3.38.** Elastic modulus of extruded PLLA.
The stress vs. strain plot of the tensile tests of extruded 75PCL 25PLLA fiber during the eight weeks of degradation is shown below in Figure 3.39. The 75PCL 25PLLA fiber property degrades slightly during in vitro degradation. This is clearly seen by the appearance of gaps between the two components in the extruded fibers in the electron microscopic images. Since the blend with more PLLA was covered with PCL, so much of the alkaline solution of phosphate buffered solution did not reach the PLLA for degradation to occur.

![75PCL 25PLLA](image)

Figure 3.39. Stress vs strain extruded 75PCL 25PLLA.

The tensile strength and elastic modulus of the 75PCL 25PLLA fibers fluctuated but almost remained constant during the eight weeks of degradation with some variance. The tensile strength and elastic modulus of 75PCL 25PLLA fibers during the eight weeks are shown below in Figure 3.40 and Figure 3.41 respectively.
The stress vs. strain plot of the tensile tests of extruded 50PCL 50PLLA fiber during the eight weeks of degradation is shown below in Figure 3.42. The 50PCL 50PLLA fiber property degrades significantly after four weeks in vitro degradation. This
is clearly seen by the appearance of significant gaps between two blends in the extruded fibers in the electron microscope images. Since only some of the PLLA in the blend was covered with PCL, much of the alkaline solution of phosphate buffered solution did reach the PLLA for degradation.

Figure 3.42. Stress vs strain extruded 50PCL 50PLLA.

The 50PCL 50PLLA fibers started losing its tensile strength significantly after four weeks of degradation. But the elastic modulus of the 50PCL 50PLLA remained almost constant with some degree of variance. The tensile strength and elastic modulus of 50PCL 50PLLA fibers during the eight weeks are shown below in Figure 3.43 and Figure 3.44 respectively.
Figure 3.43. Tensile strength of extruded 50PCL 50PLLA.

Figure 3.44. Elastic modulus of extruded 50PCL 50PLLA.

The stress vs. strain plot of the tensile tests of extruded 25PCL 75PLLA fiber during the eight weeks of degradation is shown below in Figure 3.45. The 25PCL 75PLLA fiber property degrades a lot after four weeks in vitro degradation. The degradation was so severe that after four weeks of in vitro degradation, the fibers were
in no shape of tensile testing for fifth week and broke during handling. The degradation is clearly seen by appearance of cracks between the two components in the extruded fibers in the electron microscope images. The cracks were propagated due to uneven water absorption of two polymers in the immiscible blend system. The PLLA absorbed more water than PCL which caused more stress and the polymer blend cracked. Since only some of the PLLA in the blend was covered with PCL the alkaline solution of Phosphate buffered solution came in contact with the PLLA for degradation and hence the blends cracked.

![25PCI 75PLLA](image)

Figure 3.45. Stress vs strain extruded 25PCL 75PLLA.

The 25PCL 75PLLA fibers started losing its tensile strength significantly after four weeks of degradation. But the elastic modulus of the 25PCL 75PLLA was not stable due to degradation of PLLA. The tensile strength and elastic modulus of 50PCL 50PLLA fibers during the eight weeks are shown below in Figure 3.46 and Figure 3.47 respectively.
The stress vs. strain plot of the tensile tests of extruded PP fiber during eight weeks of degradation is shown below in Figure 3.48. The PP fiber property does not degrade at all after eight weeks in vitro degradation.
The PP fibers lost very little of its tensile strength and elastic modulus significantly during the eight weeks of degradation. The tensile strength and elastic modulus of PP fibers during the eight weeks are shown below in Figure 3.49 and Figure 3.50 respectively.

Figure 3.48. Stress vs strain plot of extruded PP.

Figure 3.49. Tensile strength of extruded PP.
Figure 3.50. Elastic modulus of extruded PP.

All of the tensile test results during the eight week of degradation is shown below in Figure 3.51 and 3.52.

Figure 3.51. Elastic modulus of extruded fibers.
Figure 3.52. Tensile strength of extruded fibers.

Figure 3.53. Strain to failure of extruded fibers.
When we compare tensile test results of pure components, all the blends and, it is observed that the tensile property of the material can be tailored according to the need by varying the ratio of PCL and PLLA. The PLLA clearly has high elastic modulus and high tensile strength than PCL. But its blends have much less tensile strength and elastic modulus compared to PLLA due to its immiscibility.

3.8.2 Electrospun Fibers

The tensile tests of electrospun fiber were done for all blends during the eight weeks of in vitro degradation. The tensile testing of electrospun fibers is shown below in Figure 3.54.

![Electrospun Fiber](image)

Figure 3.54. Tensile testing of electrospun fiber.

The complete results of the tensile tests of the extruded samples are shown in the Table 3.12 to Table 3.19 below.
Table 3.12. Electrospun fibers week 1 report.

<table>
<thead>
<tr>
<th>Sample</th>
<th>Elastic Modulus (MPa)</th>
<th>Tensile Strength (MPa)</th>
<th>Strain at Failure (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pure PCL</td>
<td>1.15 ± 0.23</td>
<td>5.36 ± 0.82</td>
<td>15.33 ± 0.99</td>
</tr>
<tr>
<td>75PCL 25PLLA</td>
<td>0.31 ± 0.11</td>
<td>1.4 ± 0.46</td>
<td>14.9 ± 1.22</td>
</tr>
<tr>
<td>50PCL 50PLLA</td>
<td>0.75 ± 0.21</td>
<td>3.05 ± 0.66</td>
<td>12.7 ± 1.74</td>
</tr>
<tr>
<td>25PCL 75PLLA</td>
<td>1.38 ± 0.41</td>
<td>4.69 ± 1.65</td>
<td>58.59 ± 8.13</td>
</tr>
<tr>
<td>Pure PLLA</td>
<td>5.29 ± 0.62</td>
<td>19.1 ± 1.72</td>
<td>9.05 ± 1.63</td>
</tr>
</tbody>
</table>

Table 3.13. Electrospun fibers week 2 report.

<table>
<thead>
<tr>
<th>Sample</th>
<th>Elastic Modulus (MPa)</th>
<th>Tensile Strength (MPa)</th>
<th>Strain at Failure (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pure PCL</td>
<td>0.55 ± 0.2</td>
<td>4.54 ± 0.75</td>
<td>23.84 ± 2.3</td>
</tr>
<tr>
<td>75PCL 25PLLA</td>
<td>0.18 ± 0.09</td>
<td>1.25 ± 0.25</td>
<td>13.21 ± 1.7</td>
</tr>
<tr>
<td>50PCL 50PLLA</td>
<td>0.71 ± 0.11</td>
<td>2.34 ± 0.6</td>
<td>8.91 ± 1.02</td>
</tr>
<tr>
<td>25PCL 75PLLA</td>
<td>0.71 ± 0.14</td>
<td>4.03 ± 1.5</td>
<td>52.79 ± 4.24</td>
</tr>
<tr>
<td>Pure PLLA</td>
<td>4.93 ± 1.07</td>
<td>17.89 ± 1.81</td>
<td>6.22 ± 1.25</td>
</tr>
</tbody>
</table>

Table 3.14. Electrospun fibers week 3 report.

<table>
<thead>
<tr>
<th>Sample</th>
<th>Elastic Modulus (MPa)</th>
<th>Tensile Strength (MPa)</th>
<th>Strain at Failure (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pure PCL</td>
<td>0.47 ± 0.11</td>
<td>3.85 ± 0.45</td>
<td>19 ± 1.7</td>
</tr>
<tr>
<td>75PCL 25PLLA</td>
<td>0.16 ± 0.03</td>
<td>0.63 ± 0.24</td>
<td>8.46 ± 1.61</td>
</tr>
<tr>
<td>50PCL 50PLLA</td>
<td>0.71 ± 0.2</td>
<td>2.55 ± 0.33</td>
<td>5.1 ± 1.03</td>
</tr>
<tr>
<td>25PCL 75PLLA</td>
<td>0.81 ± 0.17</td>
<td>4.13 ± 1.12</td>
<td>110.9 ± 9.23</td>
</tr>
<tr>
<td>Pure PLLA</td>
<td>1.95 ± 0.91</td>
<td>8.23 ± 2.09</td>
<td>13.05 ± 4.19</td>
</tr>
</tbody>
</table>

Table 3.15. Electrospun fibers week 4 report.

<table>
<thead>
<tr>
<th>Sample</th>
<th>Elastic Modulus (MPa)</th>
<th>Tensile Strength (MPa)</th>
<th>Strain at Failure (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pure PCL</td>
<td>0.46 ± 0.24</td>
<td>3.71 ± 0.68</td>
<td>29.54 ± 3.4</td>
</tr>
<tr>
<td>75PCL 25PLLA</td>
<td>0.13 ± 0.03</td>
<td>0.61 ± 0.21</td>
<td>12.43 ± 2.58</td>
</tr>
<tr>
<td>50PCL 50PLLA</td>
<td>0.89 ± 0.24</td>
<td>2.4 ± 0.48</td>
<td>3.38 ± 0.25</td>
</tr>
<tr>
<td>25PCL 75PLLA</td>
<td>0.96 ± 0.12</td>
<td>3.9 ± 0.42</td>
<td>42.63 ± 4.12</td>
</tr>
<tr>
<td>Pure PLLA</td>
<td>1.95 ± 0.65</td>
<td>8.23 ± 1.24</td>
<td>9.36 ± 3.7</td>
</tr>
</tbody>
</table>
Table 3.16. Electrospun fibers week 5 report.

<table>
<thead>
<tr>
<th>Sample</th>
<th>Elastic Modulus (MPa)</th>
<th>Tensile Strength (MPa)</th>
<th>Strain at Failure (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pure PCL</td>
<td>0.67 ± 0.22</td>
<td>4.31 ± 1.52</td>
<td>25.01 ± 3.9</td>
</tr>
<tr>
<td>75PCL 25PLLA</td>
<td>0.19 ± 0.06</td>
<td>0.81 ± 0.21</td>
<td>4.53 ± 1.1</td>
</tr>
<tr>
<td>50PCL 50PLLA</td>
<td>0.48 ± 0.25</td>
<td>1.73 ± 0.51</td>
<td>7.37 ± 2.22</td>
</tr>
<tr>
<td>25PCL 75PLLA</td>
<td>0.6 ± 0.14</td>
<td>2.73 ± 0.62</td>
<td>109.16 ± 10.79</td>
</tr>
<tr>
<td>Pure PLLA</td>
<td>1.73 ± 0.61</td>
<td>7.88 ± 1.4</td>
<td>9.36 ± 3.81</td>
</tr>
</tbody>
</table>

Table 3.17. Electrospun fibers week 6 report.

<table>
<thead>
<tr>
<th>Sample</th>
<th>Elastic Modulus (MPa)</th>
<th>Tensile Strength (MPa)</th>
<th>Strain at Failure (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pure PCL</td>
<td>0.67 ± 0.28</td>
<td>4.49 ± 1.61</td>
<td>32.99 ± 4.21</td>
</tr>
<tr>
<td>75PCL 25PLLA</td>
<td>0.17 ± 0.07</td>
<td>0.77 ± 0.28</td>
<td>6.23 ± 1.55</td>
</tr>
<tr>
<td>50PCL 50PLLA</td>
<td>0.31 ± 0.05</td>
<td>1.39 ± 0.45</td>
<td>10.21 ± 2.19</td>
</tr>
<tr>
<td>25PCL 75PLLA</td>
<td>0.45 ± 0.17</td>
<td>2.59 ± 0.78</td>
<td>150.18 ± 11.23</td>
</tr>
<tr>
<td>Pure PLLA</td>
<td>1.48 ± 0.49</td>
<td>7.07 ± 1.55</td>
<td>6.2 ± 1.59</td>
</tr>
</tbody>
</table>

Table 3.18. Electrospun fibers week 7 report.

<table>
<thead>
<tr>
<th>Sample</th>
<th>Elastic Modulus (MPa)</th>
<th>Tensile Strength (MPa)</th>
<th>Strain at Failure (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pure PCL</td>
<td>0.72 ± 0.17</td>
<td>4.22 ± 0.99</td>
<td>30.57 ± 6.25</td>
</tr>
<tr>
<td>75PCL 25PLLA</td>
<td>0.16 ± 0.08</td>
<td>0.68 ± 0.25</td>
<td>4.53 ± 1.29</td>
</tr>
<tr>
<td>50PCL 50PLLA</td>
<td>0.47 ± 0.07</td>
<td>1.4 ± 0.5</td>
<td>5.63 ± 1.08</td>
</tr>
<tr>
<td>25PCL 75PLLA</td>
<td>0.49 ± 0.11</td>
<td>2.47 ± 0.53</td>
<td>101.83 ± 15.21</td>
</tr>
<tr>
<td>Pure PLLA</td>
<td>1.14 ± 0.41</td>
<td>4.25 ± 0.45</td>
<td>7.27 ± 1.08</td>
</tr>
</tbody>
</table>

Table 3.19. Electrospun fibers week 8 report.

<table>
<thead>
<tr>
<th>Sample</th>
<th>Elastic Modulus (MPa)</th>
<th>Tensile Strength (MPa)</th>
<th>Strain at Failure (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pure PCL</td>
<td>0.72 ± 0.23</td>
<td>4.19 ± 1.01</td>
<td>31.83 ± 10.21</td>
</tr>
<tr>
<td>75PCL 25PLLA</td>
<td>0.12 ± 0.07</td>
<td>0.59 ± 0.31</td>
<td>5.09 ± 1.19</td>
</tr>
<tr>
<td>50PCL 50PLLA</td>
<td>0.48 ± 0.16</td>
<td>1.26 ± 0.47</td>
<td>6.23 ± 1.5</td>
</tr>
<tr>
<td>25PCL 75PLLA</td>
<td>0.61 ± 0.19</td>
<td>2.47 ± 0.49</td>
<td>72.24 ± 9.75</td>
</tr>
<tr>
<td>Pure PLLA</td>
<td>1.22 ± 0.35</td>
<td>3.44 ± 0.75</td>
<td>4.14 ± 1.25</td>
</tr>
</tbody>
</table>
The stress vs. strain plot of the tensile tests of electrospun PCL fiber during eight weeks of degradation is shown below in Figure 3.55. The PCL fiber properties does not degrade at all after eight weeks in vitro degradation. The tensile strength of the electrospun PCL fibers drops slightly from 5.36 ± 0.82 MPa to 4.19 ± 1.01MPa. Unlike the extruded PCL fiber ($\varepsilon_f=500\%$), the electrospun fibers tend to break as. This early failure of electrospun PCL is caused by the electrospinning process itself. The electrospinning process diposites fibers randomly in any direction on the collector plate with many resulting pores. The fiber junction points create a stress concentration which causes it to fail early. With the randomness of junction points the strain to failure of tends to fluctuate during degradation.

![Electrospun PCL](image_url)

Figure 3.55. Stress vs strain plot of electrospun PCL.

The stress vs. strain plot of the tensile tests of electrospun PLLA fiber during eight weeks of degradation is shown below in Figure 3.56. The PLLA fiber property does degrade significantly after eight weeks in vitro degradation. The tensile strength of
the electrospun PLLA fibers drops significantly from $19.1 \pm 1.72$ MPa to $3.44 \pm 0.75$ MPa. The strain to failure tends to fluctuate during degradation period.

Figure 3.56. Stress vs strain of electrospun PLLA.

The stress vs. strain plot of the tensile tests of electrospun 75PCL 25PLLA fiber during the eight weeks of degradation is shown below in Figure 3.57. The 75PCL 25PLLA fiber property does degrade significantly after eight weeks in vitro degradation. The tensile strength of the electrospun 75PCL 25PLLA fibers drops from $1.4 \pm 0.46$ MPa to $0.59 \pm 0.31$ MPa. The strain to failure also dropped from $14.9 \pm 1.22$ to $5.09 \pm 1.19$ during degradation.
Figure 3.57. Stress vs strain of electrospun 75PCL 25PLLA.

The stress vs. strain plot of the tensile tests of electrospun 50PCL 50PLLA fiber during the eight weeks of degradation is shown below in Figure 3.58. The 50PCL 50PLLA fiber property does degrade significantly after eight weeks in vitro degradation. The tensile strength of the electrospun 50PCL 50PLLA fibers drops significantly from $3.05 \pm 0.66$ MPa to $1.26 \pm 0.47$ MPa. The strain to failure also drops to half from $12.7 \pm 1.74$ to $6.23 \pm 1.5$. 
Figure 3.58. Stress vs strain of electrospun 50PCL 50PLLA.

The stress vs. strain plot of the tensile tests of electrospun 25PCL 75PLLA fiber during the eight weeks of degradation is shown below in Figure 3.59. The 25PCL 75PLLA fiber property does degrade significantly after eight weeks in vitro degradation. The tensile strength of the electrospun 25PCL 75PLLA fibers drops significantly from 4.69 ± 1.65 MPa to 2.47 ± 0.49 MPa. The strain to failure tends to fluctuate during degradation period as noted for all electrospun fibers.
Figure 3.59. Stress vs strain of electrospun 25PCL 75PLLA.

The eight weeks of tensile test data of all the pure PCL, PLLA and its blends is cumulatively shown in Figure 3.60 and 3.62.

Figure 3.60. Elastic modulus of electrospun fibers.
Figure 3.61. Tensile strength of electrospun fibers.

Figure 3.62. Strain to failure of electrospun fibers.
When we compare all the blends and its pure components tensile test, we observe that tensile property of the material can be tailored according to the need by varying the ratio of PCL and PLLA. The PLLA clearly has high elastic modulus and high tensile strength than PCL. But its blends have much less tensile strength and elastic modulus compared to PLLA due to its immiscible blend property.

3.8.3 Knitted Mesh

The tensile tests of knitted mesh were done for just PCL before eight weeks of in vitro degradation. The digital camera images were used to calculate the effective area of the knitted mesh which was used to back calculate the true stress of the knitted mesh.

The Tension 9 sample had biggest pore size when compared to other samples followed by Tension 7 and Tension 5 respectively. The pore size of Tension 5, Tension 7 and Tension 9 samples are 0.12 ± 0.04, 0.2 ± 0.01 and 0.3 ± 0.02 cm$^2$ respectively. The comparison of pore sizes is given in Figure 3.63 below.
Since the Tension 9 mesh samples had biggest pore size, so it had less number of pores and junction per square centimeter compared to Tension 5 and Tension 7 samples. The numbers of pores per square centimeter of Tension 5, Tension 7 and Tension 9 samples are $7.5 \pm 0.8$, $6 \pm 0.4$ and $3 \pm 4 \text{ cm}^2$ respectively. Also the numbers of junctions per square centimeter of Tension 5, Tension 7 and Tension 9 samples are $15 \pm 2$, $12 \pm 1.3$ and $6 \pm 2 \text{ cm}^2$ respectively. The comparison of number of pores and junction is shown below in Figure 3.64 and 3.65 respectively.
The tensile test of the Tension 5 sample is done in both warp and weft direction of the mesh. The tensile test of the Tension 5 sample is show below in Figure 3.66. The
tensile testing was done in both warp and weft direction. The tensile strength of mesh in warp and weft direction was $13.46 \pm 1.24$ MPa and $10.38 \pm 1.56$ MPa. But initially the warp side shows much more compliance by stretching without taking much of the stress. The stress vs strain plot of Tension 5 sample is shown below in Figure 3.67. The sample keeps on yielding as extruded PCL sample to 500% or more.

Figure 3.66. Tensile testing of Tension 5 sample.

![Tension 5](image)

Figure 3.67. Stress vs strain of Tension 5 sample.
The tensile test of the Tension 7 sample is done in both warp and weft direction of the mesh. The tensile test of the Tension 7 sample is shown below in Figure 3.68. The tensile testing was done in both warp and weft direction. The tensile strength of mesh in warp and weft direction was 13.38 ± 1.31 MPa and 13.19 ± 1.36 MPa. But initially the warp side shows much more compliance by stretching without taking much of the stress. The stress vs strain plot of Tension 7 sample is shown below in Figure 3.69. The sample keeps on yielding as extruded PCL sample to 500% or more.

Figure 3.68. Tensile testing of Tension 7 sample.

![Tension 7 tensile testing](image)

Figure 3.69. Stress vs strain of Tension 7 sample.
The tensile test of the Tension 9 sample is done in both warp and weft direction of the mesh. The tensile test of the Tension 9 sample is show below in Figure 3.70. The tensile testing was done in both warp and weft direction. The tensile strength of mesh in warp and weft direction was 12.42 ± 1.62 MPa and 9.47 ± 1.89 MPa. But initially the warp side shows much more compliance by stretching without taking much of the stress. The stress vs strain plot of Tension 9 sample is shown below in Figure 3.71. The sample keeps on yielding as extruded PCL sample to 500% or more.

Figure 3.70. Tensile testing of Tension 9 sample.

Figure 3.71. Stress vs strain of Tension 9 sample.
The distinct difference is observed between testing warp and weft direction. The warp direction tends to stretch more initially before it takes much stress. This phenomenon is caused by the design of the mesh or simply the design flaw of the mesh from knitting. But in weft direction it is almost behaves like the single strand of fiber since it does not get to stretch as warp side due to design.
4.1. Effect of Blend Composition on Bioresorption

With retention of the glass transition and melting points corresponding to the pure components, the polylactide (PLLA) and poly(ε-caprolactone) (PCL) blends indicate that they are thermodynamically immiscible. The result of immiscibility is a likely phase separated structure that produces a tortuous path for the transfer of water from the bath through the fiber. Thus the ratio of the pure component compositions in the blend affects the bioresorption. Since the amorphous area is the lower density phase in a polymer, water is first absorbed through the amorphous area followed by the crystalline area. So the water absorption rate is increased or decreased by the degree of crystallinity of the material. Hydrolytic degradation is the primary mechanism of degradation in PLLA and it occurs at a faster rate than PCL. Higher the water absorption, the more likely is the presence of water around the biopolymer chains. Due to increase in the reaction of the polymer chains in the matrix and surface more polymer chains are broken down into oligomers which escape from the structure. The process of chains breakage and conversion of polymer chains into oligomers to escape from the matrix is quantified in the weight loss. As the structure loses its mass it is clearly shown in the weight loss. This loss in weight indicates the degradation of polymer or bioresorption in the phosphate buffered solution. Thus both intrinsic solubility of the polymer in water, propensity for hydrolytic degradation influence the bioresorption. While this is easily deduced in single component systems, the results in this thesis
showing a trend of increasing water absorption with increased PLLA presence show that phase separation contributes in the mechanism of transport and consequent increased polymer-water interaction. PLLA shows a higher bioresorption rate than PCL for the entire time period of the measurement. Blends however show phase separation influencing the availability of the weakest link (PLLA) to the water for higher degradation. At short times (<4 weeks) the PLLA blends have a higher bioresorption rate than the pure polymer because the immiscible interface with PCL allows more transport. At long times, the polymer is influenced by the proportionate area (volume fraction) of the PLLA and trends are more linear. That is increased PCL means decreased bioresorption.

4.2 Effect of Blend Composition on Mechanical Breakdown

The tensile behavior of the PCL and PLLA are very different. As shown in the tensile data the PCL strain to failure is much higher (+500%) than of PLLA strain to failure (40%). High strain to failure of PCL is due to ductility of PCL. Also the glass transition temperature of PCL is -60°C and the tensile test were done at room temperature, the PCL chains are mobile and during tensile test they align themselves in the direction of the force. But in case of PLLA the glass transition temperature is 58°C and test was done at room temperature which below the glass transition temperature. So the PLLA is brittle at room temperature. Before tensile testing the PLLA sample is clear, but during tensile test the sample becomes hazy or white color indicating a crazing mechanism. This transformation of PLLA is due to formation of stress bands or cracks are developed perpendicular to the direction of the force, which diffracts light. In
the blends the compositions of the blends dominate the type of failure. While pure PCL with a high strain to failure shows classic necking deformation together with chain alignment and fibrillar microstructures on stretching, the PLLA with low strain to failure and a classic 90 degree to the tensile axis, crazing deformation. Remarkably when a small fraction of PLLA is introduced into PCL (75PLLA 25PCL) or when a small amount of PCL is introduced into PLLA, the deformation mechanism is dominated by fibrillar fracture corresponding to the pure PCL. At the 50:50 compositions failure is neither fibrillar nor crazing and cleavage fracture dominates. During the degradation, the mechanism of failure was retained.
Figure 4.1. SEM images of tensile tested extruded samples: (A) PCL; (B) 75PCL 25PLLA; (C) 50PCL 50PLLA; (D) 25PCL 75PLLA; (E) PLLA; and (F) PP.

Microscopic examination of each pure component and blend over the period of mechanical breakdown was found to be of value. In the case of PCL, when the yielded part of the sample is observed in the SEM, there are fibrils created in the direction of the force. The creation of a groovy structure in the yielded region is due to the alignment of
PCL in the direction of force. The SEM images of PCL tensile tested samples over the period of bioresorption measurements are shown below in Figure 4.2. The fibrillar structure during degradation was retained for the entire 8 week period. This supports the lower water absorption and lower bioresorption of the PCL sample.
Figure 4.2. Tensile tested extruded PCL fibers: (A) 0 week; (B) 2 week; (C) 4 week; (D) 6 week; (E) 8 week.

The SEM images of 75PCL 25PLLA tensile tested samples are shown below in Figure 4.3. In the case of 75PCL 25PLLA, the sample shows transformation from the highly ductile failure in the pure component to a more brittle fracture surface similar to pure PLLA. Over the period of the bioresorption, the deformation mechanism changed to diminishing ductility. In the early weeks (0 and 2 week) the PCL and PLLA show ductility by stretching to form a neck and then failure of the sample. This can be concluded by the observation of a stretched neck-like structure on the cross section of the failed sample. But as degradation proceeded, (6 and 8 week) the sample simply failed in a brittle manner with a failure cross section perpendicular to the tensile axis.
Figure 4.3. Tensile tested extruded 75PCL 25PLLA fibers: (A) 0 week; (B) 2 week; (C) 4 week; (D) 6 week; (E) 8 week.
The SEM images of 50PCL50PLLA tensile tested samples are shown below in Figure 4.4. In 50PCL 50PLLA the fracture is brittle compared to the pure PCL and ductility is limited to the center of the fiber surface. (i.e. 0 and 2 week). But towards the end of the degradation period, (> 4 weeks) the sample has a brittle fracture with a clean cross sectional surface.
Figure 4.4. Tensile tested extruded 50PCL 50PLLA fibers: (A) 0 week; (B) 2 week; (C) 4 week; (D) 6 week; (E) 8 week.

The SEM images of 25PCL 75PLLA tensile tested samples are shown below in Figure 4.5. In the case of 25PCL 75PLLA, the extruded sample was only tested to the fourth week because the sample lost almost all its tensile strength after four weeks, limiting the ability to grip the sample. Crazing coupled to fibrillar deformation are observed in the fracture surface of the as processed sample. As the degradation started, even a small fraction of PCL (25%) in PLLA caused separation. The higher ductile phase, PCL, formed fibrillar structures as in its pure component and bridged the crack plane to hold the two surfaces together even at high deformations. So PCL fiber stretching is clearly seen at the failure cross section. With increased time, the PLLA structure breakdown and increased PCL fibrillation is observed. This clearly states indicates that phase separation causes unique contributions to structural breakdown not observed in pure components.
Figure 4.5. Tensile tested extruded 25PCL 75PLLA fibers: (A) 0 week; (B) 2 week; (C) 4 week.

The SEM images of PLLA sample is shown below in Figure 4.6. In the case of PLLA, the initial sample has cracking on the crack plane perpendicular to direction of force. This shows limited ductility. Throughout the degradation process, a highly brittle deformation mechanism is observed.
In case of PP, the sample yields the same way during eight weeks of degradation. There is not much change in the surface of the yielded sample. The SEM images of PLLA sample is shown below in Figure 4.7.
Figure 4.6. Tensile tested extruded PLLA fibers: (A) 0 week; (B) 2 week; (C) 4 week; (D) 6 week; (E) 8 week.
Figure 4.7. Tensile tested extruded PP fibers (A) 0 week; (B) 2 week; (C) 4 week; (D) 6 week; (E) 8 week.
In summary, the blends with higher PLLA content are seen to form cracks. These cracks would form surface imperfections that would increase the transport of water through the fiber. In addition, hydrodynamic stress from higher water absorption in the PLLA, created internal stress in the matrix causing it to crack or in the case of phase separated blends introducing cracking between the components. We can deduce that this combination of events caused the mechanical breakdown of the blends.

4.3 Effect of Processing on Bioresorption

The effect of different processing is a one of dominating reason for rate of bioresorption. Both electrospun and extruded PLLA started to lose their mechanical strength after four weeks of degradation, so did the blends. The blend was much more vulnerable to PBS than pure PLLA due to stresses caused by uneven water absorption. Due to this reason the 25PCL 75PLLA formed huge cracks and was not able to continue mechanical testing after four weeks of immersion in PBS. In other blends too the mechanical strength was falling after four weeks of immersion in PBS.

The processing conditions of the material influenced the crystallinity of the blend. The crystallinity of blend is controlled by the cooling rate for extruded fibers and evaporation of solvent for electrospun fibers. Since all the extruded fibers were cooled in a bath set at room temperature water after being extruded from twin screw extruder and electrospinning was done at same condition, the crystallinity of the blend was affected by the composition. But while comparing the similar composition fibers two processing condition (extrusion and electrospinning), the extruded fibers were higher crystalline. This difference of crystallinity is clear in the differential scanning calorimetry
results shown above. The higher crystallinity simply means more amorphous content. These amorphous are the gateway for water absorption, as water get absorbed through amorphous part and then followed by the crystalline part. Since electrospun fibers due to processing are largely amorphous compared to extruded counterpart and also has more surface area. So much more surface of electrospun fibers are direct contact with alkaline PBS than extruded fibers due its higher amorphous content and higher surface area. Due to higher surface area and higher amorphous part more alkaline water gets in contact with the material to convert them into soluble oligomers. So more weight lost by the electrospun fibers compared to similar composition extruded counterpart. Hence the electrospun are much faster reabsorbed by the body than extruded fibers.

Also due to different processing of the same composition blend, the fibers have different mechanical strength. The extruded fibers have higher mechanical strength than electrospun fibers which is shown in the results. But due to immersion of fibers in PBS for eight weeks both of the fibers loose significant mechanical strength. This loss of mechanical strength can be explained by loss of polymer chains in the form of soluble oligomers due to water absorption. The water absorption is again controlled by percent crystallinity which is controlled by processing. So during the eight weeks of in vitro degradation test, the fibers lose weight and has mechanical breakdown.

The effect of processing in mechanical strength is shown below in Figure 4.8. Due to difference in processing condition the mechanical strength of extruded fibers are much higher than electrospun fibers. The extruded PLLA has tensile strength about 63% more than of electrospun PLLA. Similarly the PCL has tensile strength about 70% more than electrospun PCL. The trend is also followed by the blends as extruded
75PCL25PLLA, 50PCL50PLLA and 25PCL 75PLLA has tensile strength about 93%, 79% and 57% respectively higher than electrospun fibers. Similarly the elastic modulus of fibers also drops due to processing condition. The elastic modulus of extruded PCL is 52% higher than electrospun fibers. Similarly the elastic modulus of PLLA is 72% higher than its electrospun counterpart. Also this trend is continued by the blends too as extruded 75PCL25PLLA, 50PCL50PLLA and 25PCL 75PLLA has tensile strength about 93%, 77% and 89% respectively higher than electrospun fibers.
Figure 4.8. Comparison of extruded and electrospun fibers through eight week degradation (A) tensile strength; (B) elastic modulus.

4.4 Effect of Process of Making Mesh

The two processes, knitting and electrospinning, of making meshes for tissue scaffolding lead to differences in deformation. The knitting process resulted in higher compliance in the warp direction of knitting compared to transverse. The compliance is due to slippage of junction and accommodation of stress by making the pores smaller leading to higher stretch for low resistance stresses. This compliance is useful in preventing any kind of tear of injured tissue while extreme stresses are applied. The inbuilt compliance of knitted fiber favors warp direction more than weft direction. The warp direction strains much more in lower stress than in weft direction. The stress-strain curve of the single fiber, the weft direction and electrospun fibers show a similar
stress-strain relationship. The material when stretched resists immediately. The electrospun mesh has junction points bonded by the solution and therefore are not compliant similar to the knitted samples. There is no slippage of fibers between each other to accommodate the applied stress. However, electrospun meshes are random meshes and so while there is breakage in some locations, the fibers still hold the sample together during deformation. As we have shown in our earlier publication [97] the process by which electrospinning produces a fiber entanglements [98]. Entanglements once formed are stretched in the direction of the jet but when they hit the jet there is no retention of the orientation of the fiber and therefore a largely amorphous structure can result. As seen in the comparison of mechanical properties and bioresorption rates, processing by electrospinning results in a higher degradation rate as well as lower mechanical properties. This is largely associated with the higher amorphous structure in the solvent cast, electrospun deposited meshes. That is entanglements enable the charged polymer jet to retain its dimensional stability to the point of contact with the charged plate.

The cyclic testing of tissue is shown below in Figure 4.9. In the testing we can see the tissues are very compliant as they have high strain at very low stress. This characteristic of high compliance is achieved by the knitting of extruded PCL fibers. The comparison of single extruded fiber and electrospun fiber versus knitted fibers is shown below in Figure 4.10. The single monolithic extruded fiber and electrospun mesh do not have much compliance where the knitting of extruded PCL introduces the very compliance needed to make good tissue scaffolding.
Figure 4.9. Cyclic testing of tissues.
Figure 4.10. Comparison of PCL mesh.
5.1 Material Issues

Blending affects the bioresorbability, mechanical properties and water absorption differently from the single components. Weight loss over time of blends and water absorption was related to the fraction of Polylactide (PLLA) in blended compositions. The differences in the absorption of water affected the changes in mechanical performance over time.

Both extruded and electrospun fibers containing PLLA lost mechanical strength during the eight weeks of in vitro degradation. The extent of loss was influenced by the material erosion during the degradation experiments. For extruded fibers, pure PLLA lost 74% of tensile strength, since fiber integrity was maintained. In the case of extruded 75PCL 25PLLA, 50PCL 50PLLA and 25PCL 75PLLA the loss of tensile strength correlated to increased PLLA presence in the blend. The extruded 75PCL 25PLLA, 50PCL 50PLLA, 25PCL 75PLLA, and PP lost about 26.81%, 56.83%, 82.24%, and 34.8% tensile strength respectively. Poly(ε- caprolactone) (PCL) extruded fibers gained about 23.14% of strength by losing its outer shell of amorphous PCL during the eight weeks of degradation.

After eight weeks of degradation the electrospun fibers for both single components and blends lost their mechanical strength significantly with increasing PLLA content. The electrospun PCL, 75PCL 25PLLA, 50PCL 50PLLA, 25PCL 75PLLA and PLLA lost mechanical strength about 21.8%, 57.85%, 58.68%, 47.33% and 81.98%
respectively indicating slightly higher loss. Electrospun PCL, 75PCL 25PLLA, 50PCL 50PLLA and PLLA lost 44.94%, 31.04%, 1.85% and 7.86% respectively more than extruded fibers. The difference in the extent of degradation was related to the increased amorphous content in electrospun fibers over extruded.

5.2 Design Issues

Knitting introduces a degree of compliance into the structure that monolithic single fibers do not. Tissues have a high compliance at low strains, while fibers by themselves are highly resistant at low strains. The introduction of the highly compliant junctions in a knitted structure enabled the mesh to match the pattern of strain response of a tissue. The properties of knitted mesh would vary in the warp and weft directions but corresponding electrospun fibers did not show these differences. However, the stress-strain curve of electrospun meshes matched that of the fiber constituent and thus did not give the high compliance at low strains.
REFERENCES


[65] C.A. Bischoff, P. Walden, 1894, 279, 45.


[70] Q. Fang, M.A. Hanna, Ind Corps Prod, 1999, 10, 47.


