TECHNICAL UNIVERSITY OF LIBEREC
Faculty of Textile Engineering

Wetting behavior of electrospun biodegradable materials

Master Thesis

Study programme: N0723A270002 Textile Engineering
Study branch:

Author: **Yash Chandrashekhar Shah, B.Eng.**
Thesis Supervisors: doc. Ing. Eva Kuželová Košťáková, Ph.D.
Department of Chemistry

TECHNICAL UNIVERSITY OF LIBEREC
Faculty of Textile Engineering

Master Thesis Assignment Form

Wetting behavior of electrospun biodegradable materials

Name and surname: **Yash Chandrashekhar Shah, B.Eng.**
Identification number: T19000391
Study programme: N0723A270002 Textile Engineering
Study branch:
Assigning department: Department of Nonwovens and Nanofibrous materials
Academic year: **2020/2021**

Rules for Elaboration:

1) Prepare a literature review focused to electrospun biodegradable nanofibrous materials and wetting characterization of them
 2) Design and set materials and methods parameters
 3) Electrospun biodegradable nanofibrous material by needless electrospinning
 4) Study wetting characteristics by Washburn wicking method
 5) Discuss results and draw a conclusion

Scope of Graphic Work:
Scope of Report: 40-60
Thesis Form: printed/electronic
Thesis Language: English

List of Specialised Literature:

1. SZEWCZYK, Piotr, Daniel URA, Sara METWALLY, Joanna KNAPCZYK-KORCZAK, Marcin GAJEK, Mateusz MARZEC, Andrzej BERNASIK a Urszula STACHEWICZ, 2018. Roughness and Fiber Fraction Dominated Wetting of Electrospun Fiber-Based Porous Meshes. *Polymers* [online]. **11**(1), 34. ISSN 2073-4360. Available from: doi:10.3390/polym11010034
2. HOU, Lanlan, Nü WANG, Jing WU, Zhimin CUI, Lei JIANG a Yong ZHAO, 2018. Bioinspired Superwettability Electrospun Micro/Nanofibers and Their Applications. *Advanced Functional Materials* [online]. **28**(49), 1801114. ISSN 1616301X. Available from: doi:10.1002/adfm.201801114
3. HUANG, F. L., Q. Q. WANG, Q. F. WEI, W. D. GAO, H. Y. SHOU a S. D. JIANG, 2010. Dynamic wettability and contact angles of poly(vinylidene fluoride) nanofiber membranes grafted with acrylic acid. *Express Polymer Letters* [online]. **4**(9), 551–558. ISSN 1788618X. Available from: doi:10.3144/expresspolymlett.2010.69

Thesis Supervisors: doc. Ing. Eva Kuželová Košťáková, Ph.D.
                             Department of Chemistry

Date of Thesis Assignment: November 1, 2020
Date of Thesis Submission: August 30, 2021

L.S.

doc. Ing. Vladimír Bajzík, Ph.D.   Ing. Jiří Chvojka, Ph.D.
            Dean   Head of Department

Liberec   November 1, 2020

Declaration

I hereby certify, I, myself, have written my master thesis as an original and primary work using the literature listed below and consulting it with my thesis supervisor and my thesis counsellor.

I acknowledge that my bachelor master thesis is fully governed by Act No. 121/2000 Coll., the Copyright Act, in particular Article 60 – School Work.

I acknowledge that the Technical University of Liberec does not infringe my copyrights by using my master thesis for internal purposes of the Technical University of Liberec.

I am aware of my obligation to inform the Technical University of Liberec on having used or granted license to use the results of my master thesis; in such a case the Technical University of Liberec may require reimbursement of the costs incurred for creating the result up to their actual amount.

At the same time, I honestly declare that the text of the printed version of my master thesis is identical with the text of the electronic version uploaded into the IS/STAG.

I acknowledge that the Technical University of Liberec will make my master thesis public in accordance with paragraph 47b of Act No. 111/1998 Coll., on Higher Education Institutions and on Amendment to Other Acts (the Higher Education Act), as amended.

I am aware of the consequences which may under the Higher Education Act result from a breach of this declaration.

August 26, 2021   Yash Chandrashekhar Shah, B.Eng.

ACKNOWLEDGEMET

It pleasures me immensely to recount the people, who have helped me through my endeavours, sometimes by just "being a friend or a dedicated ear that listened", sometimes by helping me technically or philosophically to overcome the hurdles in my research. I would like to thank my supervisor Doc. Ing. Eva Kuželová Košťáková, Ph.D., who had been lending her valuable time and knowledge to my research work whenever it was possible. This research work would not have been possible without her assistance. Also I would like Thank Ing. Hana Musilova For the cooperation and motivating me for the diploma thesis.

Liberec

30.08.21

Yash Chandrashekhar shah

Abstract

Water wetting behaviour of electrospun materials is important mainly for their medical application, where in testing and application is the optimal surface hydrophilicity necessary. Mostly used polycaprolactone electrospun layer are known as hydrophobic materials. The contact angle measurements are the most used analysis. The thesis brings in the theoretical part introduction to electrospinning and its application and testing as biomaterials for medical applications. There are introduced the most used biodegradable polymers and their properties with concentration to polycaprolactone as the material used in experimental part. The literature review in testing of wettability is presented here to with respect to contact angle analysis of polycaprolactone electropun materials. Experimental part is focused to wettability tests (wicking of water into polycaprolactone electrospun layers) in different time after electrospinning. The aging of electrospun material in time and different storing conditions are the main changed parameters which influence to final nanofibrous material water wettability is study here.

Keywords: Electrospinning, wetting property, polycaprolactone, Dynamic wetting, Fiber diameter, Surface density.

Abstrakt

Smáčení elektrostaticky zvlákněných materiálů vodou hraje důležitou roli při jejich aplikaci v medicíně, a to jak při jejich testování tak při vlastním použití. Široce využívaný polymer – polykaprolaton, elektrostaticky zvlákněný do podoby vrstvy je v odborné literatuře označován za hydrofobní polymer. Měření kontaktního úhlu je metoda, která se nejvíce používá k popisu smáčitelnosti povrchu kapalinami. Tato diplomová práce přináší v teoretické části úvod do technologie elektrického zvlákňování a využití a testování elektricky zvlákněných materiálů jako biomateriálů pro medicínské aplikace. Zároveň představuje nejvíce používané biodegradabilní polymery a jejich vlastnosti se zaměřením se detailněji na polykaprolakton jako materiál využívaný v experimentální části. Literární rešerše se soustředí na hodnocení smáčitelnosti zejména reprezentované měřením kontaktního úhlu pro studované materiály. Experimentální část je pak zaměřena na testy vzlínání vody do elektricky zvlákněných nanovlákenných materiálů z polykaprolaktonu v různých časech po výrobě. Stárnutí materiálů za různých okolních podmínek vykazuje změny ve smáčitelnosti a právě vliv doby po výrobě a vliv teploty skladování s ohledem na vzlínání vody do elektricky zvlákněných materiálů z polykaprolaktonu je zde detailně studován.

Klíčová slova: Elektrospinning, smáčecí vlastnosti, polykaprolakton, dynamické smáčení, průměr vlákna, povrchová hustota.

Table of Contents

List of figuers

List of Tables

1.Introduction

Nanofibrous structures are among the most advanced textile materials utilized in a wide range of applications across the world, some of which are delicate and important, such as medical textiles or protective textiles, which can put someone's life at danger. As a result, accurate measurements of these structures' particular characteristics are required to be measured. Wetting property of electrospun materials gained very crucial role in applications such as medical textiles, protective clothing, filtration of liquids, etc. Which are possible to measure by different methods such as contact angle , wicking property, goniometry etc.

It is known that electrospun materials are relatively non-uniform, rough from the surface and porous, therefore these structures are extremely difficult to test with respect to their wetting properties. However, contemporary technologies like as the sessile drop technique and the Washburn method can provide us with some significant results that can be used to describe these structures based on their wetting properties.

Materials stored at different conditions may changes their wetting property, so by storing the material at different ambient conditions it is easy to observe.

Thorough study of wetting properties of Electrospun materials which are stored in different conditions been conducted. In this thesis where the properties are tested mainly by Force tensiometer-K1000 (Krüss),which uses Washburn as the principle method. By this study it is easy to see that how electrospun material changes their properties as aging increases. And also how the ambient parameters like Temperature and humidity affects the wetting of the electrospun materials.

2. Theoretical part

The Theoretical part consists of the detailed description of electrospinning technology like principal, process and material parameters, modifications, and applications of nanoparticles. The next part will be focused on biodegradable polymers and at last in the literature review, the part focus is on wetting properties of PCL.

2.1 Introduction to electrospinning Technology

Electrospinning technology is utilized for making Nanofibers with the use of electrical forces. In this process, there is a possibility of manufacturing fibres with different diameters from the range of micrometre to the nanometres. Production of Nanofiber is considered possible by using both natural and synthetic polymers. This process of fibre spinning possesses a vital significance because of two reasons one depicting its versatility making the possibility of using a wide range of polymers in this process and another is it produces fibres of very small diameter which is not possible in other conventional spinning processes such as ring spinning etc. [1]. Electrospinning is a broadly used method for producing the Nanofiber on a large scale because it is easy to handle, uses very few solutions, controllable fibre diameter, is easy to process, economically convenient, reprocessing of fibres [2].

Fibres that are produced from the electrospinning process possess several good characteristics like high surface to volume ratio that is a very greater surface area per unit mass as well as is nearly about 1-100 square Meter per gram, high porosity, lightweight, good relatively mechanical strength, high permeability [1], [3]. Furthermore, because of this property's electrospun nanofibers are considered as the ideal material for a broad scope of applications like in Tissue engineering for drug transportation, wound healing, tissue engineering scaffolds, etc., Filtration application, in chemistry application, for reinforcement material in composite, in electronics and space application [2],[3]. Electrospinning within the fibres may be done with the help of two distinctive methods AC Electrospinning and DC Electrospinning.

2.2. AC electrospinning process

Experimental setup of the AC electrospinning as proved in figure 1. The high point voltage terminal initiating from the step-up transformer is interconnected to the spinning needle, and

connecting to the ground with a flat ground electrode. Collectors are situated amid ground electrodes and needles [44]
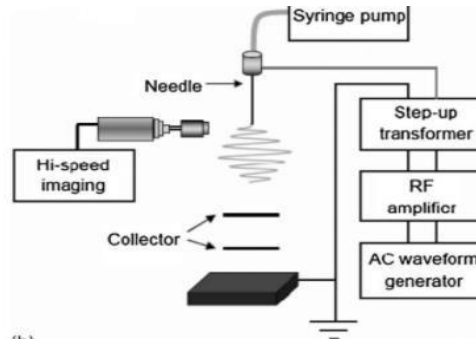
Figure 1.Schematic diagram of AC electrospinning [7].

AC electrospinning is considered extremely useful and very much useful in the concerned sector. It provides an inexpensive needle tip and straightforward method for producing fibre threads accompanied by specified morphology that may be straight connected within the regulated fabrics. This approach can be used to generate a wide range of morphologies, ranging by threads to mesh networks, by simply changing the collection location, as well as the applied voltage and frequency. Because the fibre threads and mesh networks are considered highly interwoven, they should be particularly beneficial for certain applications such as fabric weaving and filtration, and biomedical use, because of intrinsic mechanical strength. In Dc electrospinning the productivity is less, because of some reasons such as the creation of a single jet from a nozzle; as a result, this production technique is inefficient. The latter uses an electric field to destabilize free liquid surfaces, which increases throughput by producing many jets discharged from the surfaces of rollers, spheres, strings, and spirals. Although there has been considerable development in total producibility, the DC method's efficiency still is poor [7].

2.3. DC Electrospinning process

Electrospinning technique is considered a distinctive process used for producing exceptionally fine fibres through the polymeric solution or melts solution and fibres generated possess thinner diameter in nanometre and high surface to volume ratio. In DC electrospinning voltage is the important parameter to generate the electrospinning and it is in the range of several tens of

kilovolts. This technology is quite similar to pesticide sprayers, and electrostatic precipitates, its main principle is strong electrostatic repulsive forces mastering the weaker surface tension force from the charged polymer solution [1]. The solution which is used for the electrospinning process must carry the electric charge and contain sufficient viscosity so that it can be stretched without breaking into small drops [3]. The setup of the DC electrospinning system entails three main configurations as shown in figure 2.
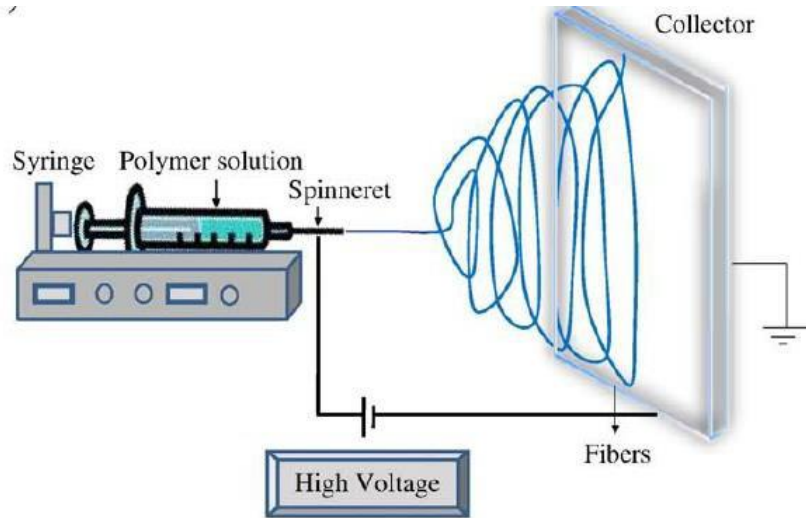
Figure 2.Schematic diagram of horizontal electrospinning [1].

The working principle of the DC electrospinning process is mainly formed on the rule of "electrostatic attraction". The pump pushes the solution due to the high voltage of the supply. Then individual droplet comes at the needle tip where the drop gathered at an oppositely charged electrode called a collector, and it is observed from here that electrostatic attraction results in the formation of fibre [2]. When the drop is at the tip of the needle at that time by increasing the electric field drop becomes highly electrified, and because of the electric charge the shape of the drop is changed into a conical shape, referred to as "Taylor cone". When electrostatic repulsive force is greater than the surface tension of the solution then it is ejected and cached by the collector. Before reaching the collector the solution undergoes different instabilities and gradually becomes thinner because of the evaporation of solvent and fibre elongation [3].

2.4. Effect of process and material criteria on the electrospinning process

There exist different parameters that may impact the electrospinning procedure that is causally related to the morphology and fabrication of the fibre. These factors are mainly classified as material parameters, process parameters, ambient parameters, and other supporting parameters. It is necessary to understand these factors to get a clear acknowledgement of the electrospinning procedure and fabricating the Nanofibers from polymers.

2.4.1. Material parameters

Material Parameters such as Concentration, molecular weight, viscosity, and surface tension of the mixture affects the electrospinning process and shows an impact on the fibre morphology as well, these parameters are listed below:

A) Concentration

The size and the shape of the formed fibres are sprightly dependent on the concentration of the mixture. It mainly focuses on three distinctive parts, like when the concentration is extremely low at this time the nanoparticles get formed and due to low concentration the viscosity is low and surface tension is high, so electrospray occurs. When the concentration of the solution is appropriate then smooth nanofibers are formed and when the concentration is too high, a helical-shaped ribbon structure is observed [4]. Figure3. shows the effect of concentration on the nanofiber formation. The absorption of the mixture also affects the flow rate of the mixture through the spinnerets, if the concentration of the mixture is less then it breaks into small droplets before reaching the collector, and this small braking forms the beads. As the concentration of the solution is increased then due to proper flow of solution the beadle's fibres are formed but when it exceeds the limit above this it affects the flow rate of the solution, the solution gets dried on the needle tip and it blocks the chain [5]. In the electrospinning procedure of the gelatine, it is found that as concentration increases the diameter of the fibre also increases [1].
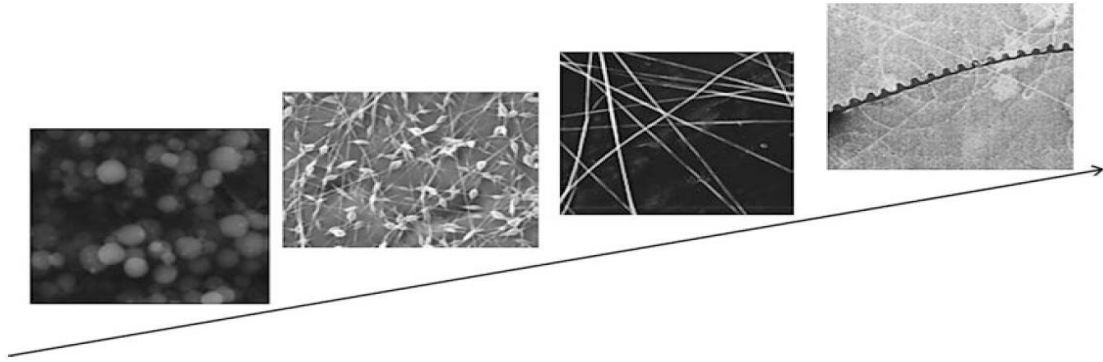
Figure 3.SEM image of the effect of concentration on the fibre formation [4]

The image above shows that effect of concentration on the fibre formation, from left to right the concentration of the solution is increasing image on the extreme left is of beads with low concentration, in the middle the concentration is an appropriate and extreme right ribbon-like structure due to high concentration [4].

B) Molecular weight

The molecular weight of the solution is not considered important in the spinning of the fibres if the intermolecular forces engagement provided by the oligomers is sufficient. The polymer's molecular weight impacts the solution viscosity, if the concentration of the mixture keeps the constant and molecular weight of the polymers is reduced then beads are formed. after an increase in the molecular weight when proper fibres are obtained and there is again an increase in the molecular weight and ribbon-like structure will be formed as shown in figure 4. [4].
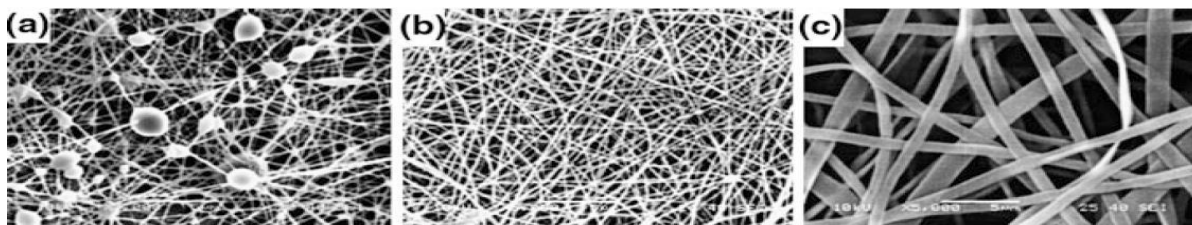
Figure 4.SEM image of the effect of molecular weight increasing from left to right [4].

The images presented above are as follows:(a) less molecular weight with bead formation(b)Appropriate molecular weight with fibre formation (c)More molecular weight with ribbon-like structure [4]. The molecular weight of the polymer impacts the properties like

viscosity, conductivity, surface tension, and dielectric strength. In electrospinning technology generally greater molecular weight polymers are utilized because they provide incredibly good viscosity for the spinning of the solution. The molecular weight of the polymer gives the entanglement of the polymer chain inside the solution [1].

C) viscosity

The viscosity of the mixture decides the morphology of the fibre and it is very much difficult in getting continual smooth fibres with very little viscosity. On the contrary, if the viscosity of the mixture is remarkably high then there is difficulty in ejecting the jet from the tip so there is a need for suitable viscosity for the electrospinning. The viscosity of the solution, concentration, and molecular weight are interdependent of each other and they together impact the morphology of the fibres [4]. The shape of droplets coming from the needle depends on the viscosity of the mixture when the viscosity of the mixture is less the shape of beads is spherical and as viscosity increases the shape changes at high viscosity providing nanofibers with good orientation [5]. In a study of polyethene oxide, it is found that the range between 1 to 20 poise is suitable for the electrospinning process [1]. Solution with the optimum viscosity is the best for the electrospinning, whereas the solution with high and low viscosity both forms the beads [2]. Figure 5.shows that change in the shape of droplets with increasing viscosity of the mixture (a) small droplets with low viscosity (b and c) further elongated droplets (d) Nanofibers are formed with optimum viscosity.
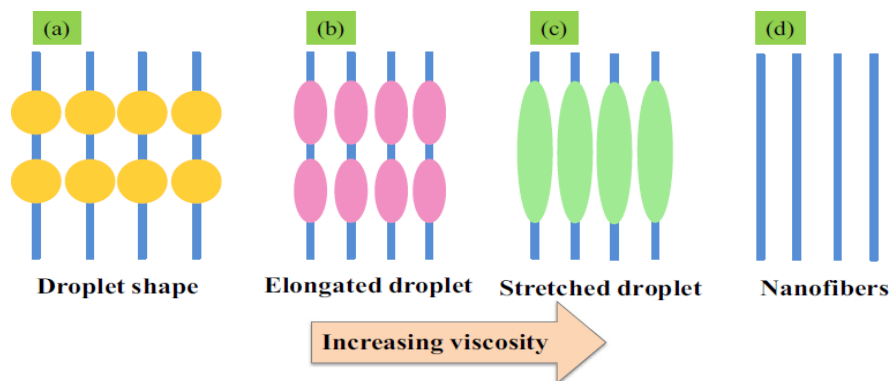
Figure 5.This shows that the change in the shape of droplets with increasing viscosity from left to right [5].

D) Surface tension

The surface tension of the solution is depicted as the force applied on the plane of the surface per unit length. In the electrospinning procedure, the value of applied voltage should be adequate in overcoming the surface tension so that the spinning mixture can form the nanofibers. Different solutions have different surface tension values and keeping the concentration of the solution constant and decreasing the surface tension of the mixture embedded fibres may get converted to level fibres [2]. Surface tension is the property of the solvent part from the solution which affects the electrospinning process, through a decrease in the surface tension of the solution fibres without beads can be produced. Casually, the greater surface tension of the mixture hinders the electrospinning process due to jet instabilities and spreading of the droplets. The surface tension of the solution decides the formation of beads, droplets, and fibres [1], [4].

2.4.2. Process Parameters

A) Applied Voltage

In the electrospinning process voltage applied is considered a significant parameter. The ejection of jets through the Taylor cone is possible only when the voltage applied crosses the value of the threshold voltage. There are several contradictions regarding the impact of the voltage applied on the diameter of the fibre. Some scientists say there is not so much impact of the voltage applied on the fibre diameter in the electrospinning of polyethene oxide. On the contrary, polyvinyl alcohol scientists said that due to high voltage the large diameter fibres are produced. On the contrary, another group discovered that due to high voltage there is an increase in the electrostatic repulsive force, and because of this narrow diameter fibres are produced, while it is observed that bead formation takes place at high voltage.

Conclusively it is seen that applying voltage throws impact on the diameter of the fibre [4]. As voltage is raised beyond the critical value bead formation takes place along with that the diameter of the fibre also increases and it results in a decrease in the size of the Taylor cone at the end of the needle and jet velocity also increases at the same rate of flow [5]. In many research papers, it is found that higher applied voltage provides more stretching in the solution and thin fibres are

formed. If the solution of low viscosity is used with higher applied voltage then it forms the secondary jets and thin fibre diameter is produced, and by the high voltage rate, the stretching of the solution droplets increases and it may provide a rapid acceleration towards the collector due to greater potential difference [6]. Figure 6 shows that if the voltage is less than the Taylor cone will not form but if the voltage exceeds then droplets are stretched.
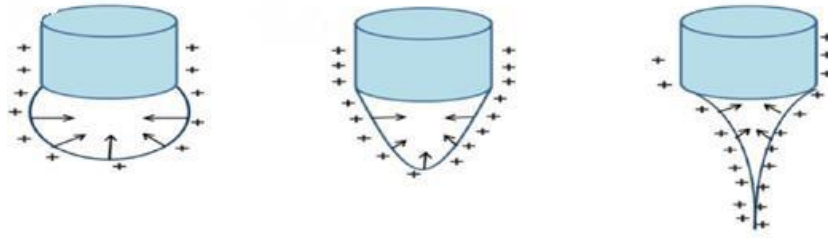
Figure 6.Formation of the Taylor cone because of the charge on the solution [5]

B) Solution flow rate

Solution flow rate is another vital element as it affects the velocity of the jet and the transfer rate of the material. The lower flow rate is good for the electrospinning process because it allows more time for the evaporation of the solvent. In the study of polyester, the diameter of the fibre increases with a lower rate of flow, and the morphological framework also gets slightly changed by a change in the rate of flow of the solution and a high flow rate gives the bead because of not having proper drying time [1]. To form the uniform and beadles' fibres it is recommended to use a critical flow rate for the polymer solution and this property ranges with polymer solutions. In some studies, it is observed that in an increase in the rate of flow the unspun droplets and ribbon-like structure is formed this is due to less stretching of solution and no evaporation of the solvent in the time between needle and collector [5].

C) Tip to collector distance

Distance within the collector and tip have an impact upon the path of the jet and the duration required for travelling before resting on the collector. Electric field intensity is inversely proportional to the electric field strength at a constant voltage. If voltage is continual and the distance is changed then the properties of the fibre are quite similar to the change in voltage and maintain a constant distance. An increase in the distance first reduces the fibre diameter and then

increases due to the weakening of electric field strength [6]. If the distance in midst of the collector and tip is less then fibre would not have adequate time to get dry before moving to the collector and if the distance is very much stretched then bead formation may happen. Therefore optimum distance is required for the smooth formation of fibres [4].
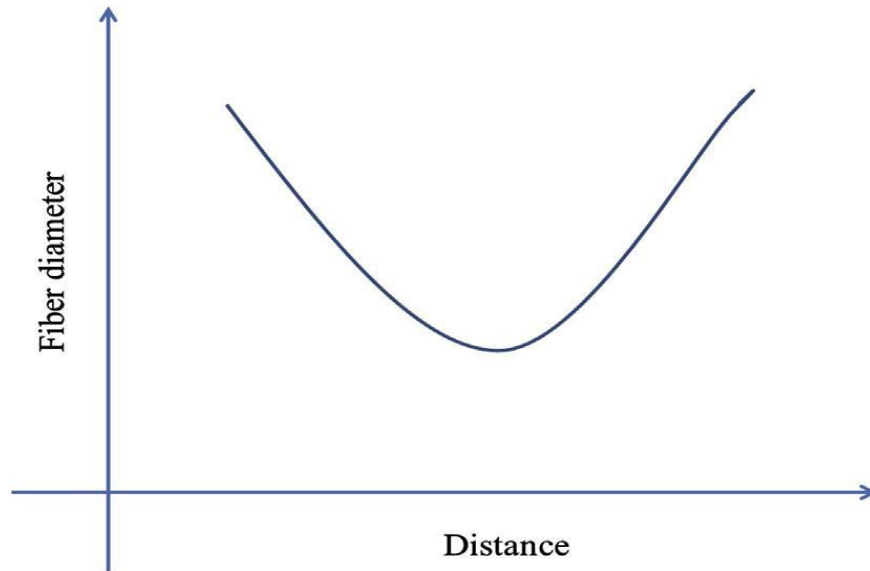
Figure 7Graphical representation of change in the fibre diameter with change in distance (cm) from tip to collector [6].

2.4.3. Ambient parameters

Along with material parameters and process specifications there also lies ambient parameters like humidity and temperature impacting the electrospinning process. In the spinning of the nylon-6, it is found that when the temperature range changes from 20- 60°C give more production by reduced fibre diameter. It happened because of the reduction within the viscosity of the mixture while the increase of the temperature. In the case of humidity, if humidity is more then sample circular pores are formed, and if increase further then pores coalescing takes place on the fibre surface [1]. If there is low humidity, then the solvent is dried completely and the solvent evaporates very quickly and on the other hand, if humidity is high then fibre diameter increases due to the counterpoise of charges on the jets, and stretching forces results low [5]. In another study, it is found that when temperature increases the solution evaporates very quickly and it leads to forming of pores on the surface and further enhancement in temperature leads to an increase in the pore size. Temperature

and humidity are interdependent, when the temperature gets high the humidity goes down and the solvent evaporates faster [2].

2.5 Modifications of the electrospinning process

2.5.1 Modification in collector

Electrospinning setup contains the collector, and it influences the productivity, arrangement, and structure of the collected film. There exists a distinctive variety of collectors utilized for collecting the fibre-like Rotating collector, static collector, and precision deposited collector. According to the requirement and the end-use distinctive variety of collectors has been selected.

A) Rotating collector

A rotating collector is used to collect the most aligned electrospun fibres at a certain rate of speed. There are different types of rotating collectors like Mandrel, wired drum collectors, and rotating discs. In this approach, there is a possibility of getting good alignment of the fibres as an orientation of fibre. In mandrel type of collector, the fibre gets more orientation as speed is increased and the diameter of the fibre gets thinner because of the drawing. In the Rotating wire drum collector, highly aligned fibres are obtained, but it is difficult to get a thick layer of the aligned fibres. In this system, the only drawback is the air turbulence made by the fast rotation of the collector. To remove this the air shield is placed so that it gets easy for getting more aligned fibre with enhanced productivity and less wastage. Figure 8. shows different types of rotating collectors (A) Mandrel, (B&C) Wired drum collector and (D) Rotating disc for the collection of the nanofibers [6].
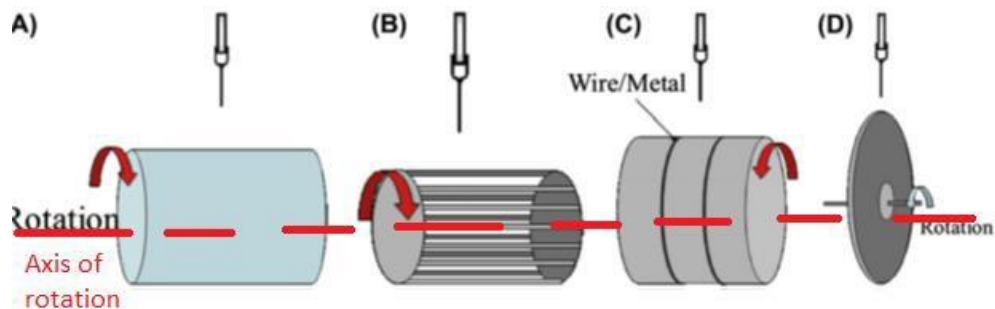
Figure 8.Different types of rotating collectors [6]

B) Static collector

As shown in figure 9.(A) Parallel electrode is used to collect aligned fibres by manipulating the electric field. Air space within two collecting electrodes is used in the parallel electrode collector. This both encourages and compels the electrospinning jet to bounce back and forth in midst of the electrodes, leading into aligned fibres. (B) The collection of crisscrossing nanofibers at the centre of an assembly of parallel electrodes is also demonstrated. (C) The formation of a nanofiber bundle can be aided by two keen edges placed on a line with a space. For better alignment, the electrodes kept inclined.
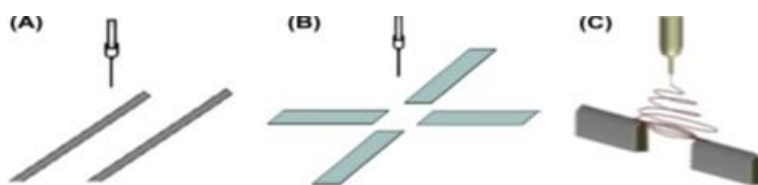
Figure 9.Different types of static collectors are used for the proper alignment of fibre [6]

2.5.2. Modification in the spinneret

The spinneret is another important parameter of the electrospinning assembly from which the charged solution comes out and forms the stable Tylor cone. Generally, the metal nozzle is used with a flat tip, but nowadays different modifications are done, and different spinnerets are used to develop the different fibres in the same process.

A) Coaxial spinneret

This is generally used spinneret to manufacture the core and sheath type of fibre. Different experiments are performed on this spinneret. The main benefit of this spinneret is it may be utilized in two different solutions and obtain the required fibre accountable as per the requirements, e.g. In the outer layer polymer Solution is used and in the inner part, the spinnable solution is used. During the spinning the solution in the outer side is carried into the inner side and when the outer polymer is eradicated, and a considerable inner polymer nanofiber is achieved. Similarly, hollow

24

fibres are produced with help of this spinneret. The schematic representation of coaxial spinneret is explained in figure 10.[6].
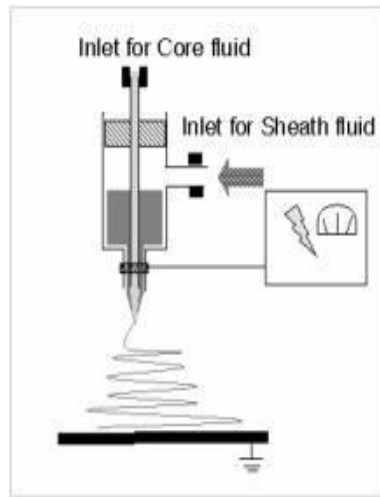
Figure 10.Schematic diagram of the coaxial spinneret [44]

B) **Two component spinnerets**

This is similar to the coaxial spinneret, this variety of two side capillaries is being used. Considering the use of bi-component fibres get manufactured with the use of electrospinning technology. The main application is a switch in the electric circuit, as it is manufactured in such a way that one side may absorb the chemical and the other side is electrically conductive, and due to absorption of chemical in one side it will swell, and nanofibers will bend [6].

C) **Centrifugal electrospinning**

In this method instead of rotating the collector the rotating spinneret is used known as centrifugal electrospinning utilized for creating the aligned fibre. In the process the voltage used required is very less because centrifugal force can master the surface tension of the mixture and electrospinning is initiated. In electrospinning of PAN with higher rotational speed, the centrifugal solution jet followed straight flight towards the collector. The alignment of the fibre in this system generally depends on the direction of the nozzle exit. Given the direction of the centrifugal force, different researchers have observed distinctive alignment of fibres followed by a different arrangement of the machine [6]
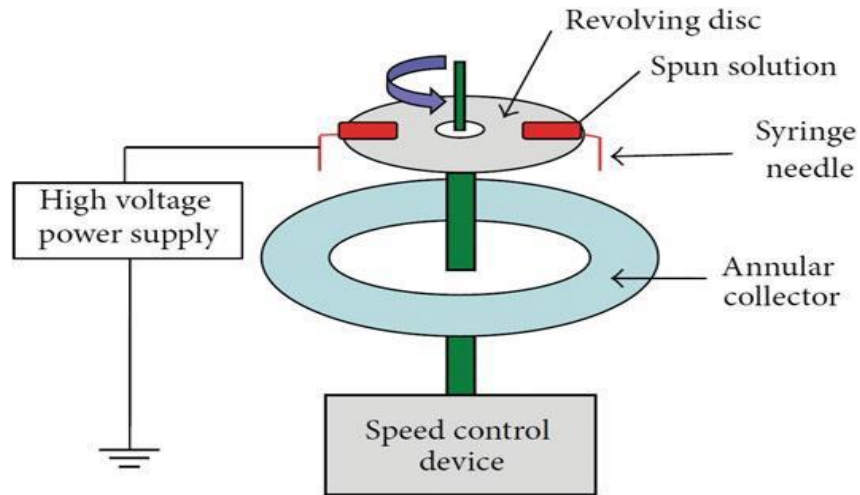
Figure 11.Schematic diagram of centrifugal electrospinning [3]

D) Needless Electrospinning

In conventional electrospinning, the needle is used for generating the Nanofibers but there exist some issues linked to the use of the needle while producing the Nanofibers like needle clogging, low production rate, and very little production. The invention of needleless electrospinning helps in eradicating every problem. In the year 2005 first commercial needless electrospinning was manufactured by the company Elmarco s.r.o. under the brand name nano spider and they used the rotating cylindrical spinneret to produce the nanofibers.

The spinnerets are partially immersed in the spinning solution and when it starts rotating then a layer I of a solution is formed on the surface. After the rotation of spinnerets resulting in form of agitation of the solutions, and also spikes get formed. As soon as the application of high electric field the Tylor cone is formed and with the help of raised spikes of droplets in the last polymeric jets are being stretched for the Nanofibers that gets collected. For the past few years continuous research is going on, but this method has its advantages such as a better production rate compared to needle electrospinning, and the problems related to the needle are avoided. In figure 12, it is shown that cylindrical spinneret is used for the electrospinning process.
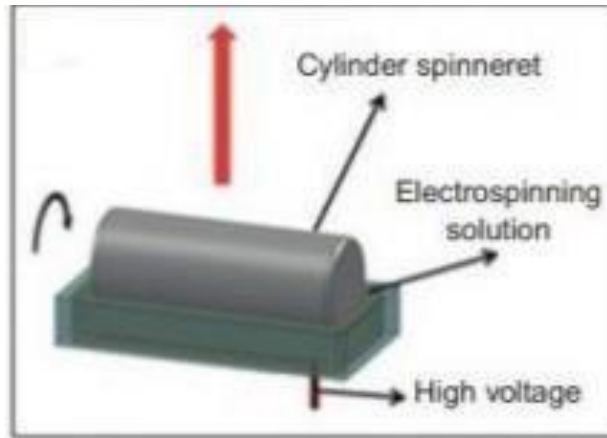
Figure 12.Rotating cylindrical spinneret used for the electrospinning of nanofibers [45]

2.6. Applications of nanomaterials

Products that are made of nanoparticles have several good properties like high surface-to-volume ratio, high porosity, and good mechanical features. The electrospinning procedure provides the desired fibre morphology and mechanical strength because of the ease of distortion of the mixture parameters and procedure parameters. Another advantage is the number of fibres required for the production is less and fibres can manufacture in any shape by using different polymers due to the versatility in the process [1].

Nanofibers can be used for different applications in various fields such as in chemistry for catalyst and cell batteries, in defence for protective clothing, for environmental use to remove toxic waste, and broadly utilized in biomedical uses mainly in tissue engineering for scaffold, wound dressing, and drug delivery. It also can be used in filters and cosmetics [2].

 The focus is mainly on the application of the nanoparticles in the biomedical applications generally concerned with tissue engineering as the thesis is conducted based on the production of material that is considered in the biomedical applications. After a lot of research and analysis, it is found that 2/3 of its applications are in the medical field. Around 50% use is infiltration applications and other applications share the rest. There is a lot of research still going on in this field. Figure 13 is a schematic representation of the application of electrospinning in different sectors. [1]
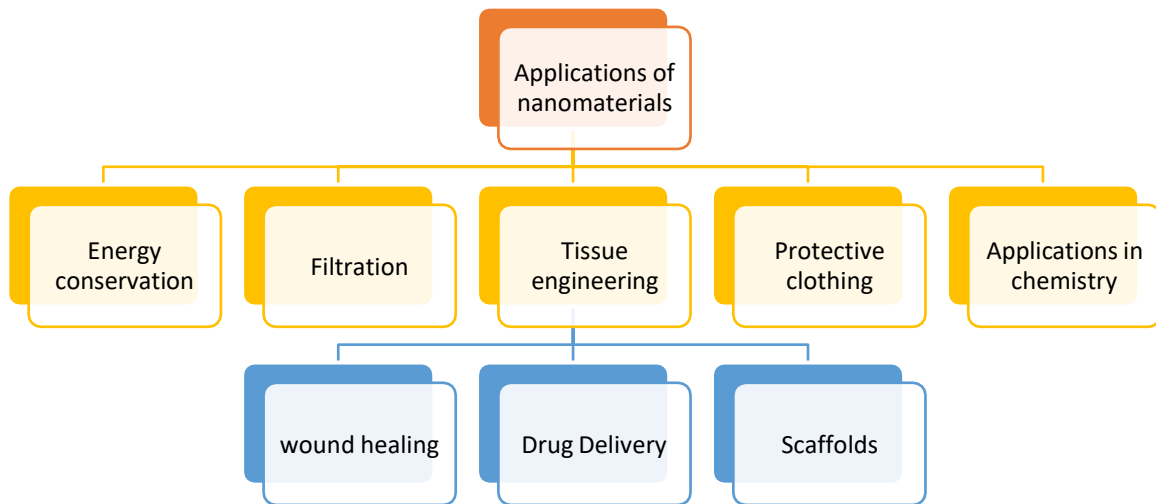
Figure 13.Tree diagram of an example of applications of tissue engineering

(Source: Created by Author)

2.6.1. Tissue engineering applications

A) Wound healing

It is the process of the generation of new tissue in the infected area. To repair the damage to the affected area, a chemical reaction takes place between the skin and applied chemicals. When the body burns deeply there is no possibility of regeneration of cells except at the edges of the wound. Therefore wound dressing helps in protecting the affected area, by emanating the extra fluids from the wound area, protecting the wound from microorganisms, and enhancing the speed of the healing procedure. For this purpose, dressing material protects the wound as well as provides oxygen transmission and moisture.

Considering every needed feature and property, the electrospun material can be considered best for wound healing due to its different properties: it is a highly porous material that helps in transmitting the moisture and oxygen as well as helps in emanating the unwanted portion within the infected area. Its high surface-to-volume ratio provides good control for fluid drainage. In the procedure of manufacturing drugs are added into fibres as per the requirement of further medical treatment and for antibacterial purposes [2]. In earlier days, the materials that were used for wound dressing are mainly made up of hydrocolloids, hydrogel, alginate, and their salts [1].

In one study it is observed that the wound healing of mice with electrospun nanofibrous collagen material is better than that of conventional wound care methods. The study has provided an incredibly good result within the recovery in the initiating phase of the healing process of the wound [1]. In one of the studies, it is found that the nanoparticles of chitosan with PEO of different compositions provided excellent results among the good water transmission rate as it possesses a good water-absorbing and holding capacity. In cross-linked polymers, it is observed that they have more tensile strength along with good flexibility of material [2]. It has also been observed that human epidermal growth factor is enhanced when PCL – PEG block copolymer is used) In vivo wound healing of an ulcer.

B) Drug delivery

The main focus of drug delivery is in releasing the drug at a predetermined rate within time and it is only possible if biodegradable and biocompatible polymers are used because controlling this polymer is not difficult and can be designed according to our needs. A basic principle of a drug delivery system is the rate of dissolution depends on the surface area of the drug and the carrier and it gets enhanced with an increase within the surface area of both parameters. A bigger surface area of the nanoparticles gives quick as well as effective solvent evaporation and this is considered as the main reason following the usage of nano spun fabric concerning the use of drug delivery [1].

Considered as a probable material in the drug release the electrospun nanofibrous material provides several advantages in the procedure of drug delivery. The loading of drugs by this process is very easy and the voltage applied in the electrospinning process throws a little impact upon the drug release action. The drug release through this fabric is easier due to the high specific surface area and the small length for the diffusion passage. The release of the drug also can be controlled by the change in the nanofiber morphology, porosity, and composition [8].

In recent days, for the treatment of cancer patients, new drugs are designed to cure it effectively, but there are a lot of restrictions such as it may damage the cells due to chemotherapy, overdosing, clinical toxicity, etc. In this case, nanofibers play an important role by doing easy drug carrier with proper control of drug release rate. The scientist shoaling should develop the new core-sheath device made up of co-axial electrospinning technology for the treatment of cancer patients which contains PEG and PCL copolymers along with loaded drugs and it had shown very good results

by killing the tumour cells with minimum drug, less frequency of dosages and this has improved the lives of the affected people [2]. A typical example of the drug delivery system is explained in figure 14.
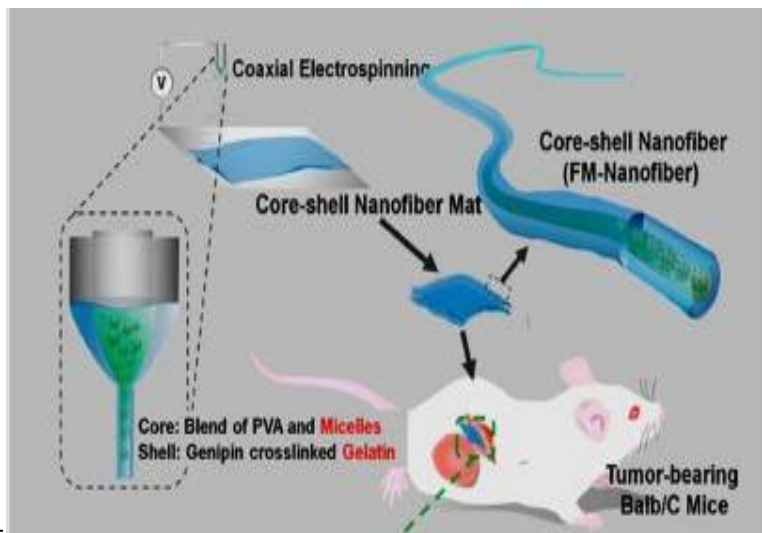
Figure 14.Drug loading technique by tissue-engineered nanofibers [2].

C. Tissue engineering scaffolds

Scaffolds are mainly used for repair, maintaining, or improve tissue function or body parts. The production of the scaffolds depends on the production, design, and characterization of material that makes cell generation along with in vivo and in vitro cell culture. Scaffolds must comprise of the following properties like good pore size, high porosity, distribution, high surface area. It should be biodegradable, when the new tissues grow, provides efficient structural integrity for providing support to the newly generating tissues, be nontoxic to the cells, etc. A lot of research is carried out to find the best suitable material for the scaffolds which has similar properties to the extracellular matrix. As it is seen that the structure of the electrospun nanofibers mat is nearly similar to the extracellular matrix, therefore, it is used as a scaffold material for tissue culture and the 3-D structure made from the nanofibers in electrospinning is suitable to grow all types of cells [8].

The fibre diameter procured through the electrospinning process is like that of the fibrils in the ECM, which gives the same environment and provides the substrate for the growth of the cell. The electrospinning process is the most widely used approach for preparing nanofibrous scaffolds. Many natural polymers are used to produce this scaffold because of their properties such as

30

biocompatibility, Bio functionality, and when some of them are blended with synthetic polymers it can help to improve the cytocompatibility of the scaffolds. Different nanofibers are used in tissue engineering to produce different scaffolds such as bone, skin, cartilage, blood vessels, heart, nerve tissue, etc. [1]. Figure 15 shows the tissue engineering principle of scaffolding. Where the first scaffold is manufactured then it is kept in the culture liquid for the growth of tissues and finally implanted in the human body.
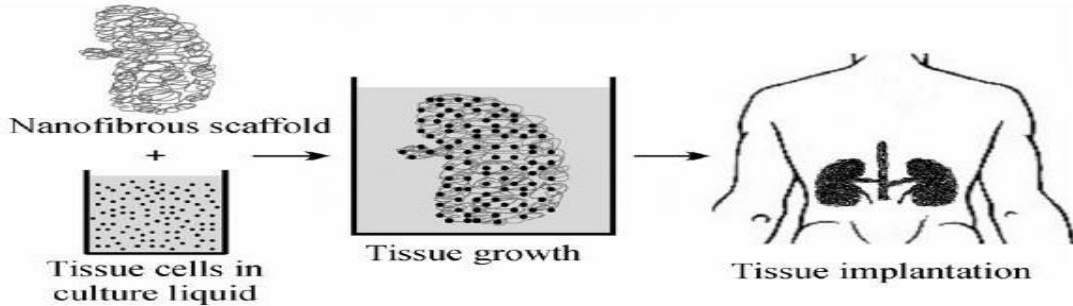
Figure 15.Tissue engineering principle of scaffolds implantation [8].

2.6.2. Filtration application

In doing the procedure filtration the structural properties of the filter fabric should be kept the same as that of the particles or the droplets that need or are going to be filtered. This is the reason for the possibility of using the electrospun nanofibrous fabric that contains the ridiculously small diameter of the fibre. It has been seen that the particles whose size is less than 0.5mm can get trapped easily in the electrospun nanofibers filters that result in improvising the effectiveness of the filtration. The air filters which are designed by the nanofiber's membrane are tested and 100% rejection of airborne particles whose diameter is between 1-5 micrometre is observed. The filtration effectiveness is mainly based on the thickness of the filtration fabric, and it gets increased when the thickness of the fabric increases. This thing is generally achieved through the use of electro-spun nanofibers filters. Also, in one experiment it is found that the efficiency of nylon-6 nanofillers is 99.993% which is more than regular greater efficiency particulate air filter comprising the speed of 5 cm/s and 0.3-micrometre test particle size [1]. Figure 16 shows that nanofiber coating is applied on the nonwoven for the filtration application.
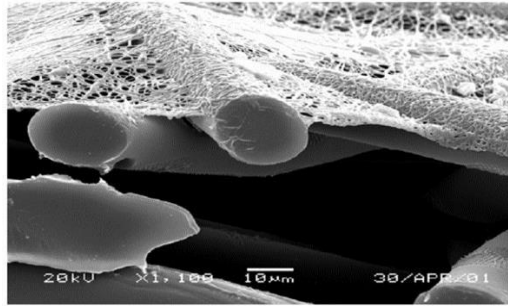
Figure 16.Nanofibrous layer on the Nonwoven for filtration application [8].

2.6.3. Safeguarded clothing applications

Safeguard clothing should contain features like lightweight, water and air porous, breathability, insoluble in every solvent toxic as well as chemical resistance. Electrospun nanofiber membrane is best suitable for this application because it is lightweight, high surface Volume ratio has high porosity which helps to make it breathable, has excellent filtration ability, they help in neutralizing the chemical despite any effect of water vapour and air porousness in the clothing. A different method of modification has been tried to improve surface protection. In one method the surface is treated with reactive chemicals such as oximes and chloramine which helps to protect from the warfare agents [1].

2.7. Information about degradable polymer

2.7.1. Introduction to biodegradable polymers and biocompatibility

For the last two decades, the use of biodegradable polymers in biomedical applications is significant. increased, they are mainly utilized in biomedical use like medical devices, drug delivery, antibacterial, biomaterial, gene delivery, tissue engineering, and diagnosis. It is possible because of the chemical, physical, and physicochemical properties of biopolymers, along with their enhanced functionality, processability, and sensitivity towards the sensation [9].

Biodegradation is revered as biological modification also known as the chemical degradation of material or polymers through the activity of microorganisms like algae, bacteria, and fungi. In most polymers, the primary degradation is carried by the activity of the microorganisms. This activity consists of biological activity. Biodegradation procedure is generally divided into dual roles: 1. aerobic degradation 2. anaerobic degradation. Aerobic reaction in which oxygen is present

and carbon dioxide is produced and in anaerobic reaction oxygen is not present and methane is produced instead of carbon dioxide [10].

When the question comes about the biomedical use like implants and drug delivery techniques which the only function for a limited time in vivo that is generated from the polymers that get eradicated through the human body with the means of hydrolytic degradation after the role of serving is finished. The use of biodegradable biomaterials for tissue engineering has two main advantages:

1. They do not need to remove from the body after its use through secondary surgery as the products that are going to be formed can be carried out through the human body parts by natural means.
2. The utilization of biodegradable material gives a good recovery of the biological system, as the steady loss concerning the mechanical properties of the implant provides a continual simulation in the human tissues.

Different parameters affect the degradation of the polymers like the chemical structure of the polymers affects the degradation of the polymers the chemical containing ester binds degrades faster. Another thing is molecular weight the polymer with less molecular weight degrades rapidly when compared to a higher molecular weight. The morphology of the polymer is also one of the important things crystalline regains degrades slowly than the amorphous region because of difficulties into the penetration of the water molecules. Polymers having glass transition temperatures less than that of body temperature are in a rubbery state and gets degrades faster than the polymers in the glassy state. Along with this high surface-to-volume ratio it also provides high water penetration. This framework can be attained through electrospinning technology that provides a high degradation rate and lastly, environmental factors such as relative humidity and temperature, ph., ionic strength, site of implantation affect both rate and extent of hydrolytic degradation [11].

Biocompatibility is another main part of biomaterial engineering that is related to the engagement of any material utilized for placing inside the human body or transplanting within the body. In short, biocompatibility is any material other than a drug or mixture of substances, that is a combination of natural and synthetic material utilized at any time for replacing or curing the organs

and tissues as well as the function of the body that comprise the whole form of an organ or the body system. To determine the biocompatibility, it is important to consider the applications, and the cell types that may encounter the material, and the time required for the material to go into in-vivo, based on the standard testing methods that are formed. This test method provides an idea concerned by the way a material may behave before any clinical trial. The selection is based on biocompatibility as well as other properties like cost-effectiveness, easy handle, and easy accessibility [12]. While selecting the biomaterial for the biomedical applications following properties should be present in the polymer such as:

1. They should be in pure form and easily sterilized by simple and old methods.
2. They must be free from any impurities.
3. They should contain desired biomechanical properties such as tension, compression, and shear.
4. They contain other properties such as hydrophobicity, hydrophilicity, wettability, surface charges, etc.
5. They should maintain their main properties and functions the entire time in-vivo.

In short, biocompatibility is a broad notion that contains the very type of responses comprise in the biological system of the biomaterial, and the use of non-biocompatible material causes problems such as blood clotting, death of the cells, injury, and cancer [11].

In a major portion of biomedical applications electrospinning technology is used, as an enhanced interest in nanotechnology, and its unique material properties. It is considered cost-effective and quite simple technology of nanofibers manufacturing. The main advantage to use electrospinning technology is it gives nanofibers, and that is considered appropriate in creating the necessary biological environment due to the physical characteristics that are the same as biological molecules. The large surface area to the volume ratio of this gives high porosity which helps to load the bioactive molecules and transfer nutrients and waste. Till now hundreds of polymers were used to produce the nanofibers such as collagen, gelatine, alginate, polyester, polyurethane, etc. In biomedical applications, biodegradable and bio comparable polymers are used because they digest into biocompatible degradation products in human structure so that second surgery to remove the

implant from the human body is not mandatory. Another advance is they degrade as the new tissues start growing [13].

2.7.2. Categorization of biodegradable polymers

Biodegradable polymers are generally classified into three different types

1. Natural polymers: polymers that are directly derived with the help of natural sources.
2. Semi-synthetic: In this process, the raw material is acquired through nature and for polymerization done after chemical modification.
3. Synthetic polymers: These are considered the polymers which are directly taken from the synthesis of chemicals [9].

Following is information about some biodegradable polymers which are mostly used in biomedical applications.

I. Polyglycolic acid (PGA)

It is revered as the most broadly utilized polymer in biomedical applications. This is hydrophilic and acquired through glycolic acid monomer by ring-opening polymerization or polycondensation. PGA acquired by the condensation polymerization provides low molecular weight with poor mechanical properties on the other side PGA acquired from ring-opening polymerization provides a greater molecular weight. When degradation happens due to the hydrolysis that initiates at the amorphous region and then continues into crystalline regains [9], [11]. PGA is biocompatible and biodegradable polyester and this is the reason it can be used widely within the medical application. Many studies it found that PGA is best suitable for the regeneration of cartilage and blood vessels [14]. Another main application is in suture material for surgery, this suture is a replacement for the collagen-based biodegradable suture because there is better matching with tissue (tissue compatibility) and good mechanical strength along with expected biodegradation. Also, it can be used for other medical applications such as burn treatment, skin damage, and packing for surface bleeding organs [11]. In studies of preparation of blend of PCL and PGA, it is observed that PGA has more average diameter than PCL with the same solution concentration but because of difference in viscoelastic properties the PGA that affect the solution. PGA is more porous and hydrophilic than PCL.

II.   Poly Lactic Acid (PLA)

PLA is aliphatic polyester that is derived through lactic acid by ring-opening polymerization. From many experiments, it is proven that PLA is biodegradable, biocompatible, and non-toxic after being destroyed in the body. In one experiment PLA is used as a semipermeable membrane for microcapsule having different contents, because of its biodegradability and nontoxicity after being destroyed. In another research where PLA is used with coated chitosan for bone regeneration shown exactly similar properties like natural bone along with good reproduction which gives the example of biocompatibility [13]. The presence of polar oxygen linkages and methyl group side chain is responsible for the hydrophilic nature of PLA and therefore it can go under decomposition in humid conditions. Contempt having all good properties, the use of PLA is restricted because of its low heat distortion temperature, poor crystallinity, and considerably high cost [9]. Mostly PLA is soluble in chloroform and methyl chloride and due presence of the methyl group it is more hydrophobic than PGA so the degradation rate of PLA is slower than PLA. After degradation, it forms L- lactic acid, which is carbohydrate metabolism in humans, so it grabs more attention as biodegradable biomaterials [11].

III.   Polylactic co-glycolic acid (PLGA)

It is acquired from the process of ring-opening polymerization of PLA and PLGA. It is the most common material used for biomedical applications. In the copolymerization process by changing the ratios of PLA and PGA monomers it is possible to get good chemical and mechanical characteristics properties without any changing the biodegradability and biocompatibility [9], [13]. The combination of PLGA with hydroxyapatite used to manufacture scaffolds is useful for bone tissue engineering which improves the modules of the scaffold and improves the cells that help to form new bone [13]. The main aim behind the combination of PLA and PGA is to control biodegradation. The most interesting thing is the sutures made from PLGA have shown good mechanical strength Along with a faster rate of absorption in-vivo [11].

IV.   Chitosan

Chitin is usually obtained from natural materials for example the shells of crabs and other insects and deacetylated derivative of chitin is chitosan. This natural derivative is biodegradable, it avoids

or stops blood clotting and insurers proper cell growth because of this property it may be utilized for different applications like wound dressing, drug delivery, and contact lenses [11]. Along with biodegradability and biocompatibility, it is nontoxic, and it also contains remarkably high blood compatibility. The material in form of a porous scaffold or hydrogel is used for injection in tissue engineering [13].

V.   Gelatine

It is acquired through partial hydrolysis of collagen tissues that transfer the fibrous protein into undisposed water-soluble protein [11]. Gelatine is mainly obtained inside animal skin and bones which is the product of hydrolysis of collagen. This is a solid, semitransparent, and colourless material. The chemical properties of gelatine are almost the same as that of collagen, but the physical properties of gelatine are chemical-dependent. This is used to manufacture the shell of capsules, in drug delivery systems and scaffolds [13]. Material made from a composite of cellulose acetate and gelatine is like the Extracellular matrix, the proportion of 25:75 shown high adherence and rapid increase in human dermal fibroblast. When it is combined with PCL it gave particularly good results for skin tissue engineering [12]. This polymer can be used for much medical utilization like surgical sutures, heart valves, vascular grafts, nerve regeneration, and surgery, etc. Because of their biocompatibility and non-toxicity [11].

Above is the general information about the polymers which are widely used nowadays, the next part is continuing with detailed information about polycarbonate which is used in the project. So, the next part is focused on information from types of polymerizations to degradation along with all properties of the polycarbonate:

VI.   Polymerization of PCL

For the last few years polycarbonate has proved its importance in biomedical and tissue engineering applications in all synthetic polymers because of the following reasons:

1. Ease of the process and can be converted into a large range of shapes and sizes because of its low melting temperature.
2. It contains exceptionally good viscoelastic properties.

Polycarbonate is aliphatic polyester, hydrophobic, semicrystalline, relatively slow degradation rate, and biocompatible along with this it shows good thermoplastic properties, and this property mainly depends on the synthesis of the polymers.

Polycarbonate polymer can be produced from two different types of polymerizations.

1. Condensation polymerization
2. Ring-opening polymerization [15] [16].

Monomer preparation

The monomers which are used to produce polycarbonate are E- caprolactone for ring-opening polymerization and 6- hydroxy caproic acid for polycondensation polymerization. Both monomers are the intermediate products of the oxidation of cyclohexanol into adipic acid using microorganisms. For commercial use, PCL is produced by the oxidation of cyclohexanone by peracetic acid [14].

Polymerization

a. Polycondensation

In this method, caprolactone polymer is synthesized by the polycondensation of 6- hydroxy caproic acid in the vacuum by removing the water. This reaction is carried out for 6 hours at gradually increasing temperature from 80°C to 150°C in absence of the catalyst. The different scientists did the polymerization of 6- hydroxy caproic acid by using different enzymes and they obtained polycarbonate with lower molecular weight and high polydispersity index, which is the reason that polycondensation is not mostly used to produce polycarbonate [15].

b. Ring-opening polymerization

Mainly polycaprolactone is integrated through the ring-opening polymerization of the E- caprolactone with the different catalysts in the polymerization process. Ring-opening polymerization of E- caprolactone is done by the distinctive mechanisms such as:

> ➤   Cationic ring-opening polymerization.
> ➤   Anionic ring-opening polymerization.

> ➢ Monomer activated ring-opening polymerization.
>
> ➢ Coordination- insertion ring-opening polymerization.

The main reason behind using ring-opening polymerization is to obtain high molecular weight and low polydispersity [14] [15] [17].

2.8. Properties of polycaprolactone.

Table 1. Properties of the PCL [14][15].

Properties	Range
Density (G/Cc)	1.07-1.200
Glass Transition temperature (°C)	-65 to -60
Melting temperature (°C)	56-65
Tensile stress (MPa)	4-785
Youngs Modulus (GPa)	0.21-0.44
Water Permeability (at 25°C in $g/m^2/day$)	177
Surface Tension (Mn/m)	51

2.9. Applications of electrospun polycaprolactone in the medical field

2.9.1. Electrospun mats for delivery system

PCL and PCL embedded types of equipment are generally electro-spun to produce mats and these mats are used in different medical applications. Electrospun mats contain a bigger surface area in comparison to solvent films due to the fibrous design and this results in a higher number of drugs that can be provided to the sites where mats are placed. Drugs containing nanofibers can be formed in two different ways either by dissolving both drugs and polymer collaboratively and after this solution is used for electrospinning or using a coaxial system. In the study of the formation of PCL mats with dexamethasone drug, two different solvents are used dichlorodiphenyltrichloroethane and acetone and from that fibre with 300 nm to 670nm were obtained. This is mainly used for inner ear disease and this combination of the modified system provides a stretched release of dexamethasone for one month. In a different case, it is necessary to change the surface of the fibre to increase the attachment of the drug to the nanofibers. In the case of doxorubicin for attachment,

the electrospun mats are partially hydrolysed. And the binding of nanofibers is achieved by putting hydrolysed mats in different PH drug solutions and drug release is more in acidic form and has shown exceptionally good attraction towards tumour area. From different studies, it is found that the composition of mats and morphology of mats can be changed and because of porosity, surface morphology and method of preparing mats are preferred than the films [19].

2.9.2. Electrospun membrane as a skin substitute

PCL electrospun membrane can be either used for skin substitute or as a wound dressing. The advantage of using PCL for this application is it provides flexibility in adding different bioactive materials like nanoparticles, wound healing drugs, development factors, etc. into the nanofibers. Another thing is they will not allow the microbes so that it helps to avoid the infection also in addition they have small pores (1-10 micrometre) they avoid bacterial effects on the sites. When PCL membrane is applied to the wound then cells migrate from the outer edge to the centre, which gives faster wound healing. Cell growth and proliferation are depending on the fibre orientation and packing of the fibre. The polyester family is an important class of biodegradable polymer. From that polycaprolactone gained much more attention towards this application because of the biocompatibility and the semi-crystalline structure is obtained from ring-opening polymerization of E- caprolactone. Wound healing or skin substitute material is capable of maintaining a moist atmosphere into would interface, allow the exchange of gases, and avoids the microbial attack so this consideration should be kept in the account as PCL is hydrophobic. But in the earlier study, it is found that PCL goes under surface hydrolysis and exposes hydrophilic surface functional group that would the wound healing and cell proliferation [19].
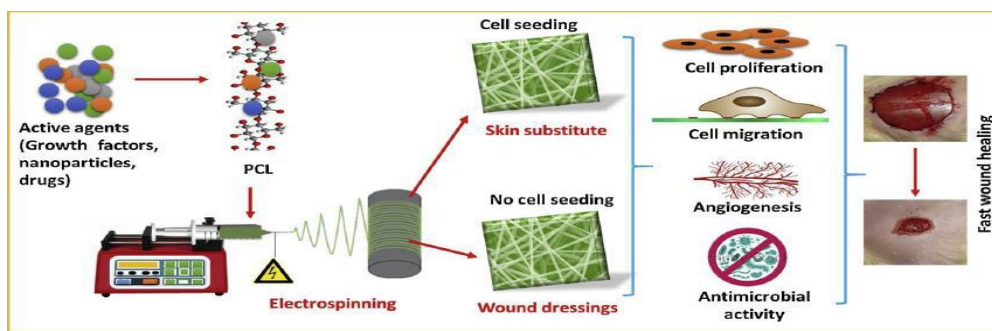
Figure 17.Application of electro-spun PCL for wound dressing and skin substitute [19]

2.9.3. Wound dressing

It is mainly used for the healing process that is the regeneration of new tissues after injury. Generally, the thicker and deeper wounds are not able to heal by themselves that is why there is a need for wound dressing element and electrospun wound-healing material to provide necessary properties such as exchange of oxygen, remove extra fluid from the contaminated area, protect from microorganisms due to an individualistic porous structure and high surface area. For wound dressing application PCL is combined with different polymers to improve the required properties. When ascorbic palpitate with silver nanoparticles is applied on the PCL it retained antioxidant and antibacterial properties. When PCL mat is attached with tetracycline it enhances cell attachment, blood clotting ability, and cell proliferation. When PCL/PLA fibre mats were treated with keratinocytes they have shown better results in wound healing by re-epithelialization, keratinocyte proliferation in the mice body. When PCL is used with chitosan it showed best wound healing [21]. The application of PCL for wound dressing is explained in figure17.

2.9.4. Scaffold for tissue engineering

Tissue engineering is the combination of medicine, biology, and bioengineering, to regenerate and modify body organs and tissues. In tissue engineering scaffold plays an especially important role in tissue repair and regrowth. The ideal scaffold which is used for tissue engineering is biocompatible, biodegradable, contains good mechanical properties, which provides a proper atmosphere concerning proliferation and cell growth. Through this aspect, polycaprolactone gained importance because of its adequate mechanical features, biodegradable, and low manufacturing cost. PCL can be used for different aspects of tissue engineering as follows:

- Bone tissue engineering

Scaffold shows a particularly important role in bone tissue engineering to repair bone defects and damage. PCL has shown incredibly good potential because of good mechanical features and possesses the capability of adjusting the degradation rate according to the growth of the bones. For bone tissue engineering the PCL is combined with different polymers to use it for different bone tissue engineering applications and got achievable results. The study of PCL with Panax ginseng release it shown excellent mechanical properties along with cell growth and adhesion. PCL is

widely used because of the ease of electrospinning but the drawback of low biodegradable rate can be overcome by combining it with different degradable polymers [22].

- Muscle tissue engineering

When PCL is used in muscle tissue engineering it given smooth muscle cells with gods adhesion and good proliferation. While the combination of collagen with nanofibers gives good elasticity and strength with improvement in the cell adhesion. To reduce the fibre surface hydrophobicity PS nanofibers are used with argon plasma which gives better results in cell attachment. Cell culture is mainly depending on the alignment of fibres, so aligned fibres shown more cell culture than randomly oriented fibres [23].

- Scaffolds for cardiovascular tissue engineering

This sphere of tissue engineering is concerned with the regrow and repair of the vessel, heart valves as well as entire heart. The scaffold is mainly used here to support the tissues and improve their mechanical properties. Here also PCL is combined with naturally different as well as synthetic polymers that show distinctive results. An interesting study of the application of core-shell fibre by coaxial electrospinning with gelatine, where the PCL is placed in core and gelatine at the shell and normal fibroblast were imparted in the scaffold. Due to the presence of gelatine, the hydrophobicity is improved, and blood interaction activity is also increased. This also results in reducing other drawbacks like thermogenic and haemolytic and gives good biocompatibility, non-toxicity, and mechanical properties [21].

- Skin tissue engineering

Skin is the largest body tissue whose function is to give protection from outside the intrusion, heat losses, regulating water retention. PCL electrospun mats give a good ability to support cell attachment and cell growth. Collagen- PCL core-sheath produced by coaxial electrospinning gives good support to cell proliferation and enhances the cell migration inside the scaffold, the cell proliferation is higher in coaxial electrospinning than normal solution coating method. Also, the

combination of PCL with collagen gives increased hydrophobicity and this leads to an increase in bio reactivity of fibroblast cells [21].

2.10. Polycaprolactone degradation

In the last few years, a lot of researchers studied the use of polycaprolactone for biomaterials and tissue engineering scaffolds but only a few of them studied the degradation of PCL scaffolds. PCL can be degraded by bacteria or fungi (by use of enzymes) but in the human body, they are not degradable because of less quantity of suitable enzymes. The degradation of the polymer is generally observed seen through distinctive approaches like the weight-loss method, calorimetry, microscopy, and by observation of the mechanical degradation [17],[24]. The hydrolytic degradation takes place by surface and bulk degradation. In hydrolytic surface degradation, the poker separation takes place only at the surface. This situation takes place when the water insertion inside the polymer is less than cutting and the formation of a new monomer or oligomer takes place. This changes the polymer structure which decreases the polymer thickness without a change in the molecular weight. The molecular weight remains unchanged in this degradation process. Generally, this gives the forecasting of process, which is useful in drug release application [16].
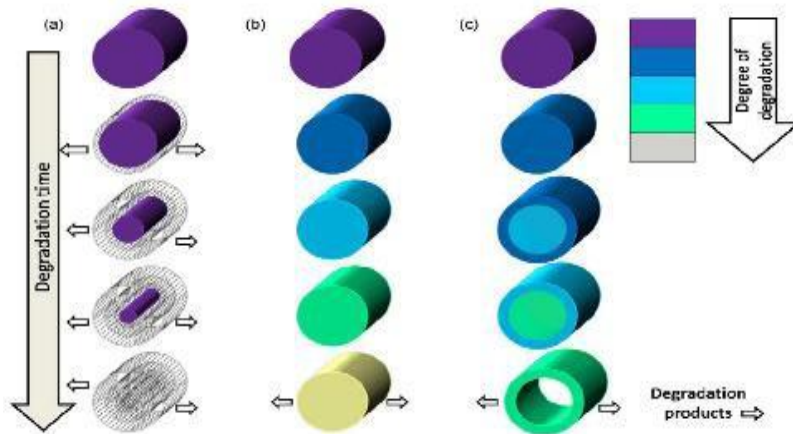
Figure 18.Different mechanisms of degradation a. Surface erosion b. Bulk degradation c. Catalytic bulk degradation [16].

When water goes fully inside the polymer then bulk degradation happens. Random chain separation happens in the pace place and decreases the molecular weight of the polymer. When water penetrates inside the polymer then the polymer chain initiates to be separated due to hydrolysis, and equilibrium occurs. If the equilibrium is distorted then the degradation mechanism

43

enhances the internal autocatalysis by distinctive side products like hydroxyl and carboxyl group. In case the internal catalyst changes acid concentration according to the time it provides new carboxyl groups in the separation stage of the ester bond. This accelerates the internal degradation rate in comparison to surface degradation. Therefore, it is defined as bimodal molecular weight degradation and leads to a hollow structure of polymer [16]. In the latest studies, it is found that for low molecular weight PCL pullularia pullulans are more effective and for PCL with higher molecular weight around 25000 to 35000 penicillium and yeast are effective. Another important factor that affects the PCL biodegradation is molecular weight and it is proved that when the molecular weight of PCL increases the degradation rate also decreases and during degradation the amorphous part degrades before crystalline regain [10]. In vivo study of PCL in rats for 3 years given results that, the PCL capsule with molecular weight 66000g/mol remained as it is for 2 years and after that, it broke down into small pieces of 8000g/mol, so it shows that molecular weight of PCL reduces by the duration. In the study of incredibly low molecular weight PCL 3000 g/mol powder, it is seen that this powder is degraded within 13 days which again proves that degradation depends on the polymer's molecular weight. In the comparison of degradation of random copolymer and homopolymer (PCL and PLA) under the same condition, it is found that as the amount of PLA is increasing the degradation rate also increased. So, it is proof of the degradation of homopolymer is more than copolymer [16]. Figure 18 shows the steps in the degradation of the PCL Polymer.
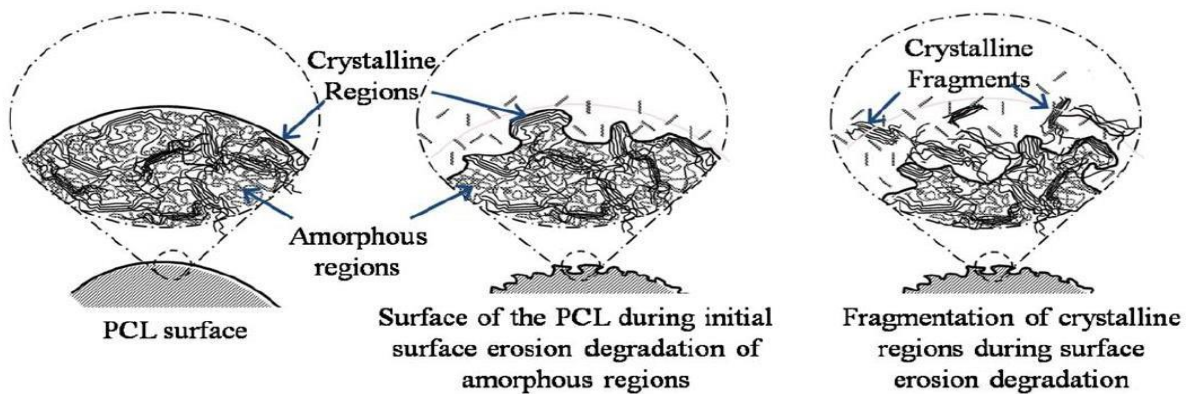
Figure 19.Degradation mechanism of PCL [16].

In the study of enzymatic degradation of PCL by Lipase, it is observed that the degradation generally occurs from surface to fibre centre, resulted by the weight loss as seen by the surface degradation and it is also confirmed from SEM. It is also seen that the rate of loss of strength is more than that of degradation is quite different from other degradable fibres. This mechanism of degradation is quite like the environmental degradation of PCL [25].

In another study of hydrolytic degradation and enzymatic degradation of PCL following results were obtained: in hydrolytic mechanism, PCL is treated in HPLC water and PBS solution, and both they got similar results on the other hand it is seen that enzymatic degradation is rapid compared to hydrolytic degradation and in the procedure is distinctive. In enzymatic degradation, it follows surface erosion mechanism while hydrolytic follows bulk erosion mechanism. So finally, while comparison in the change of properties, the enzymatically degraded sample shown some change in properties compared to the hydrolytic degraded sample on a similar weight loss percentage [23].From all the above study it is seen that 1. degradation rate concerning the PCL is very slow compared to other polymers so that it is possible to use this polymer for long-term degradable applications such as drug delivery. 2. The degradation rate of the PCL is based on the molecular weight, and it is inversely proportional to the molecular weight this happens because the molecular weight is more than the chain length also a greater number of ester binds need to be separated to produce the water-soluble monomers. 3. The enzymatic degradation is much rapid compared to hydrolytic degradation but the property in enzymatic degradation affects in the same condition [10],[16],[23],[24].

2.11. Wettability and measurements

2.11.1. Wettability

The success of the biomaterial when they come in the contact with the human body mainly depends on how they avoid the problems like thrombosis, inflammatory reactions, infection when they are used as materials in biomedical applications. Several factors affect the performance of the polymeric biomaterials, but wettability is an essential property that affects the response of interaction of cell, tissue with the substrate. In simple terms, wettability is defined as how easily liquid is spreading on the surface of the solid, and this is mainly measured by the contact angle method. The contact angle is revered as an angle created at the contact point of liquid-vapour and

solid-liquid surface which is determined by putting line perpendicular to liquid-vapour interface [26]. Wetting is defined as wetting of the solid surface with a specific liquid as per particular conditions. Wettability is the ability of an interface for engaging with liquid with determined features. Also, it is explained as engagement within the liquid and the material before the wicking condition [27]. The wettability of biopolymers is an especially important property when it is used in medical devices, implants, drug delivery systems, for diagnosis and bioreactors. In the study of PLGA based scaffolds firstly it is found that accumulation of protein on the surface limits the use of PLGA, so scientists modified the surface by use of the hyaluronic acid and chitosan for tissue engineering application and to avoid the adsorption of the protein wettability is modified with protein repulsive substance like polysaccharides. Also, it is an important parameter while growing endothelial cells because proper cell adhesion is needed on the substrate and if polymers are unprocessed then gives wettability and poor cell adhesion. So, migration, proliferation, cell adhesion, may be improvised by wettability and changed surface features without a change in huge features in biomaterials [26]. In the study of protein adhesion to biomaterial surfaces, surface wettability is one of the most important factors. In this experiment, it is found that protein adhesion depends on the wettability of the surfaces which is dependent on the change in the contact angle region, but it is not always constant. In general, hydrophobic surfaces are more protein absorbent than hydrophilic [28]. Cell adhesion in the artificial resources is impacted by the surface features like roughness, surface charge, wettability, and functional groups. Cells adhere to the polymeric surface on moderate wettability with a contact angle between 40 °-70°. From the study, it is proved that water contact angle 70° gives the most suitable surface for the cell adhesion, where the surfaces are stylized for the study is made ready with the use of commercially accessible polymers under the normal condition without any treatment. The cell adhesion not only depends on the wettability but also depends on the surface and the polymer used, for polyethene, the maximum cell adhesion is observed at 55° and for the chloromethyl silane, it is around 50° [29]. Another good application of wettability is use in electrospun mats by changing their surface properties for the different applications. Hydrophilic mats are used for cell proliferation and attachment. Hydrophobic or superhydrophobic mats are used for the applications such as block water, oil-water separation [30]. In the study of the impact of contact angle on biocompatibility, it is assumed that if the contact angle is less that means if it is hydrophilic then it is biocompatible, but it is not

mandatory. In the study, it is observed that if the contact angle is less then it is not necessary to give the enhanced biocompatibility. Blood containing devices and tissue engineering application need a balance of hydrophilic and hydrophobic surface, because if the surface is more hydrophobic then it gives good cell affinity, on the other hand, the biocompatibility is less, and more hydrophilic surface prevents cell to cell interaction, that is considered an important things in the field of tissue engineering [31].

2.11.2. Surface tension and surface energy

Surface tension is revered as a property that is provided through the molecules which are present on the surface of the liquid or the boundary of liquid. These surface molecules possess more energy than the molecules which are present inside the liquid because on the surface there are a smaller number of side molecules than bulk phase molecules and this excess energy is known as surface energy and resulting inward tension called surface tension. Both plays important role in the wetting of material [26].

2.11.3. Interfacial tension

When two different surfaces come in contact, and with the interaction of molecules a common interface is formed that produces interfacial tension, based on the contributing surfaces. When liquid droplets are present on the surface of the solid then it is needed to consider three different tension 1. Solid-liquid 2. liquid-vapour. 3. Solid- vapour. And depending on these 3 forces the wettability is determined. For determining the wettability, it is considered vital in knowing 3 different forces and based it makes the possibility of calculating equilibrium surface coefficient that imparts ideas about wetting. However, searching equilibrium surface coefficient is considered a difficult task and does not provide the exact measurement. That is why contact angle is the best method for measuring wetting on the solid substrate [26].
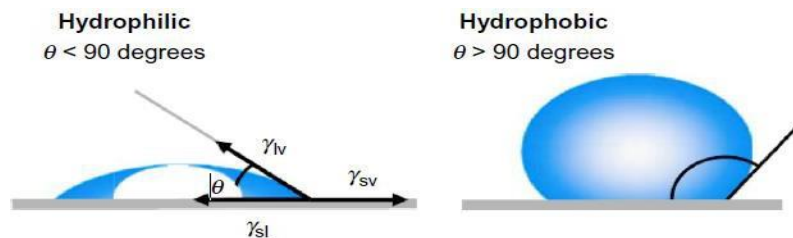
Figure 20.Hydrophilic and hydrophobic nature of solid [26].

2.11.4. Telescopic goniometry

It is considered one of the generally used methods used for determining the contact angle of liquid. In this method, the horizontally mounted telescope is utilized for measuring the angle formed by liquid on the solid surface. In this instrument, the horizontal stage is present on which sample is placed. To put a drop of water on the surface small syringe is used and for observation telescope with an eyepiece is used which gives the image. This method provides a droplet image, as well as provides the information on the contact angle along with time. The main advantage is of this method it is applicable for a small amount of liquid with small samples. On the contrary, it is considered extremely sensitive for vibration and impurities for this purpose the goniometer is mounted on the stable table for minimizing the change in the droplet shape according to the vibration. Another thing is humidification chamber is also utilized for preventing the evaporation of liquid. In short, considering the continual measurements, every parameter must be kept constant [26]. The experimental setup of the telescopic goniometry is shown the figure 21.
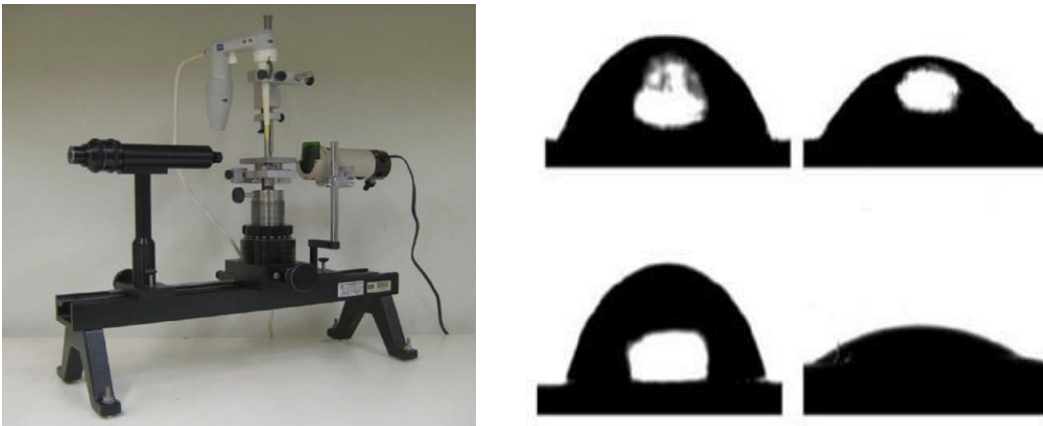
Figure 21.Experimental setup of telescopic goniometry and calculated contact of the surface with help of goniometric method [26]

2.11.5. Wilhelmy balance method

This method of determining the contact angle is related to the indirect measurement of contact angle. In this method, a very thin sample is used which is attached to the weighing balance. Then the sample meets the desired liquid by adjusting the stage of the sample and then force applied on the arm is measured and the force is known as wetting force.

$$f = \gamma lvP\cos\theta$$
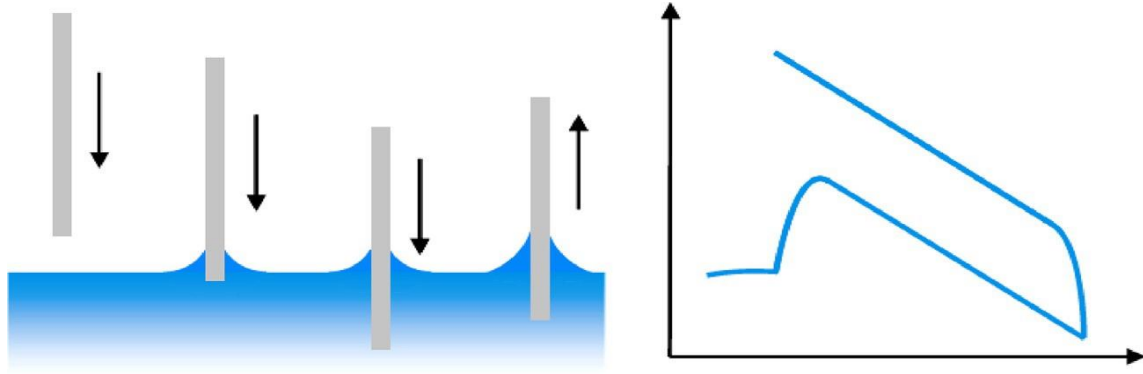
Figure 22.Wilhelmy balance method [26].

This method of providing a meaning of contact angle is considered more accurate than the optical method and provides more average value. This gives the surface properties for the whole sample as well as provides the hysteresis that makes it easy in determining the contact angle with different wetting speeds. For this method, the sample should be on uniform cross-section, composition from all the direction and if excess of liquid is used then swelling of the sample takes place [26].
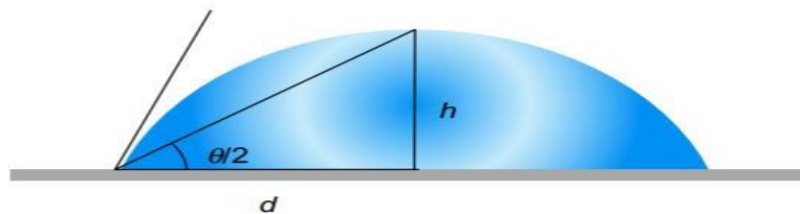
2.11.6. Drop shape analysis method

It is another method for determining the contact angle by finding the Shape of the drop. The shape of the drop depends on the surface tension of liquid that contains a property intended always in minimizing the surface and turning into a spherical shape. Therefore, the Laplace equation is used to determine the surface tension of the liquid. By considering a spherical liquid drop one contact angle is determined by measuring the diameter of the drop and height of the drop [27]. In this method, the shape of the drop is calculated by the equation, $\frac{\theta}{2} = \tan^{-1}\frac{h}{d}$

Figure 23.Drop shape analysis method [27]

49

2.12. Electrospinning of PCL: Parameters and effect on fibre morphology

In the Electrospinning procedure, several criteria govern the fibre morphology like solution parameters. Electrospinning parameters and ambient parameters are revered for the electrospinning of PCL. There are distinctive permutations and combinations executed by the different researchers in different publications are listed in table 2.

Table 2. Parameters for electrospinning of PCL [31].

Solution Parameters			E-spin Parameters			Fibre morphology
Sr.No.	Polymer concentration (w/v%)	Solvent	Voltage (kV)	Flow rate (m/min)	Distance (cm)	Avg. Fibre diameter (nm)
1	5-15	Chloroform/Ethanol (7:3)	15-20	12-24	15-20	200-1800
2	15	Chloroform/Ethanol (7:3)	20	12	20	1900
3	10	Chloroform	13	6	20	400
4	14	DMF/Tetrahydrofuran (THF) (1:1)	12	0.4	20	700
5	17	DMF/Chloroform (3:7)	10	1	20	750
6	10	DMF/Chloroform (7:10)	18	.04	15	300-500
7	10	Chloroform/Methanol (1:1)	12	6	30	250
8	8-15	Chloroform/Methanol (5:1-5:7)	19-27	3.5-18	18-33	Alternate layers of micro

2.13. PCL and Contact angels

PCL is hydrophobic when it is freshly electro spun but it changes its wetting properties after a few months or years, from the following table hydrophobic nature of PCL is seen but when it is combined with other polymers it changes its wetting property, wetting behaviour of PCL is given in Table 3. PCL with different contact angles

Table 3. PCL with different contact angles

SR.NO.	PCL With a different molecular weight	PCL Water contact angel
1	PCL-80000	118±5° [38]
1.1	(PCL+PGA) (80/20)	103° [38]
1.2	(PCL+PGA) (65/35)	76°[38]
1.3	(PCL+PGA) (50/50)	54°[38]
2	PCL+ Gelatin	53.95° [39]
3	PCL-95000	102±2° [40]
4	PCL-80000	126.5° [41]
5	PCL-40000	122° [42]
6	PCL-80000	130° [37]
7	PCL-80000	113±5° [35]
8	PCL-80000	110° [31]
9	PCL-80000	132.1±4.9° [36]
10	PCL-80000	122° [43]

3. Experimental part

The experimental part of the thesis is devoted to the testing of wetting properties of the electrospun materials which are stored at different storage conditions (Deep freeze, room temperature, and incubator) and the material is tested after every week for two months to see exactly how PCL changes its wetting properties from hydrophobic to hydrophilic. For wetting testing, it is been decided to test the dynamic wetting of material because of the drawbacks among other testing methods such as difficulty in bringing it in equilibrium position. Also in some techniques, there exists a problem in getting the accurate contact angles as water penetrates inside the sample and the contact fluctuates continually. For testing electrospun materials were PCL and media is Water.

3.1 Material selection and method of production

Selection of the material is the most important thing to decide, after research PCL polymer is selected for the following reasons: In recent years it is mostly used polymer in the biomedical and drug delivery application. It has hydrophobic and semicrystalline nature as well as possesses a low melting point (59°-64°C) and very good solubility among the solvents. Due to such characteristics, it supports stimulated and extensive research in the biomedical field. Unlike other polymers it gives another big advantage that its tailorable degradation properties and good mechanical properties because of this it is possible to use for controllable drug delivery applications and other long-term degradable devices such as sutures etc. Outside of the human body it is degradable but inside of the human body, it is not easily degradable because of lack of enzyme groups, while degradation hydrolytic degradation takes place where it starts from the surface, because of the presence of the α- Hydroxy easter groups and the bulk degradation takes. In surface degradation, thinning of the polymer takes place without affecting the molecular weight which remains constant over time from that it is easy to predict the erosion process which is useful for the desired drug delivery with a predetermined rate. Although PCL being slow biodegradable it is still being used in tissue engineering applications. Its increasing scope in tissue engineering makes it a perfect fibber for testing because the application includes liquid(blood) contact.

As a liquid for the wicking test water were selected because:

-Water occupies 90% of blood and its role in body and tissue development cannot be ignored. Hence it can be considered as an adequate liquid for the study.

- Water is the main component in the biomaterials as water is available in the interstitial liquid in cells and also helps in carrying out biochemical processes.

So, for this experiment, the polymer used PCL (polycaprolactone) purchased from Merck (Sigma Aldrich) company. Where chemicals ethanol and chloroform are provided by the PENTA chemicals. Distilled water was taken from distillation equipment at the Department of Nonwovens and Nanofibrous Materials, TUL. Values and parameters of ethanol, chloroform, and water are listed in the table.

Table 4. Properties of liquid used for the experiment

Liquid	Density	Surface tension	Viscosity
Ethanol	789 kg/m³	22.10mN/m	0.98mPa
Chloroform	1478.8 kg/m3	27.50mN/m	0.542mPa
Water	998.20 kg/m³	73.5mN/m	1.002mPa

3.2 Solution Preparation for the electrospinning

Solution of 16wt% is prepared with solvents (Chloroform and ethanol as 8:2 by weight). For the proper homogeneous solution, polymer PCL M_n 45000 (PCL-45) molecular weight is mixed with solvents and the solution for the electrospinning is prepared and stirred on the magnetic stirrer.

Figure 24.Solution preparation on the magnetic stirrer

3.3 Electrospinning of PCL

Electrospinning of PCL-45 is carried out on the Nanospider™ machine manufactured by Elmarco company which is present in the lab in CXI of TUL. With the following Parameters: collector to tip distance: 180mm; Rewinding speed: 10mm/min; SE supply voltage: 40kV; CE supply voltage: -10kV; Relative humidity: 50%; Temperature: 22°C.

Figure 25.Electrospinning of PCL and sample preparation for storage

Sample Preparation and storage of sample at different Temperature conditions:
After electrospinning, the sample is stored in the distilled airtight envelope and the samples are stored in the deep freeze, room temperature, and incubator for further testing.

3.4 Testing of the samples

A) SEM Image analysis

It is necessary to test the sample fibre diameter to observe the fibre diameter and surface characteristics, Following is the procedure followed to perform SEM image analysis and to calculate fibre diameter:

1. samples prepared for the SEM Image analysis
2. Then observe the prepared samples under SEM
3. Save the images of different magnification and finally calculate the diameter of the fibre by Fiji ImageJ software with help of the Saved images.

B) Surface density calculations

1. Cutting of samples for wetting testing (3cm×4cm).
2. Weighing the samples before testing.
3. Note down the weight of the samples and calculate the average weight of samples.

4. And finally, divide it by the 12cm² because sample dimensions are 3cm×4cm.

C) Wetting Testing

The final wicking testing was done by Force Micro tensiometer Krüss K1000, (Krüss, Germany) There is a very simple procedure for testing of material on this machine, but it needs very precise sample preparation. The samples should be cut precisely without deforming the edges. Following are the steps that followed while testing on Kruss.

1. Samples which are prepared in (4cm ×3cm) are taken and claimed in the machine on the upper jaw in such a way that the other is must be parallel to the water level.
2. Set the other apparatus of the machines.
3. Set the parameters of the machine Such as testing time, fastest acquisition time, etc.
4. Then start the test and wait until it stops automatically.
5. Then after that software gives the results in terms of the graph so save that graph and table on Microsoft excel.
6. Repeat the process.
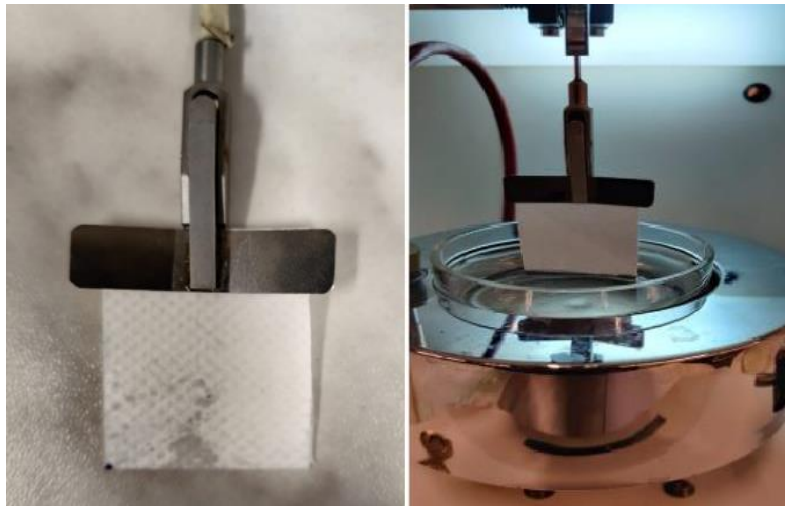
Figure 26.Clamped sample and sample during exact testing above the water
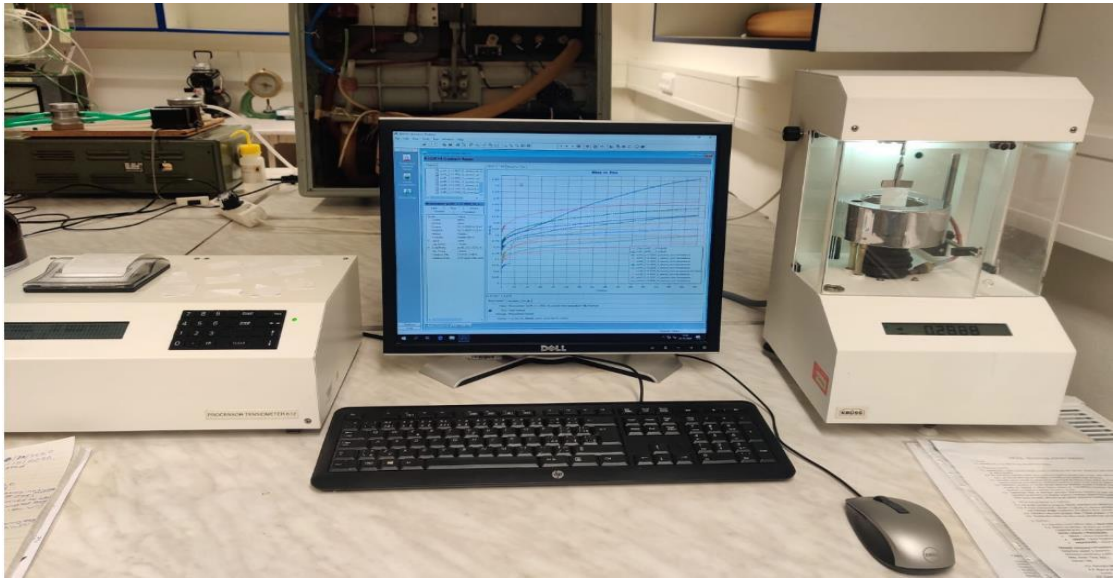
Figure 27.Whole setup of the KRUSS measurement Testing machine

1. Results and Discussion

4.1.1. SEM Results and graph of fiber diameter

SEM images of the samples which are produced with different magnification on the different time presented in this section. In figure 28, figure 29 and figure 30 Shows that SEM images of the material produced on April 2020, December 2020 and April 2021 and fiber diameter graph respectively.
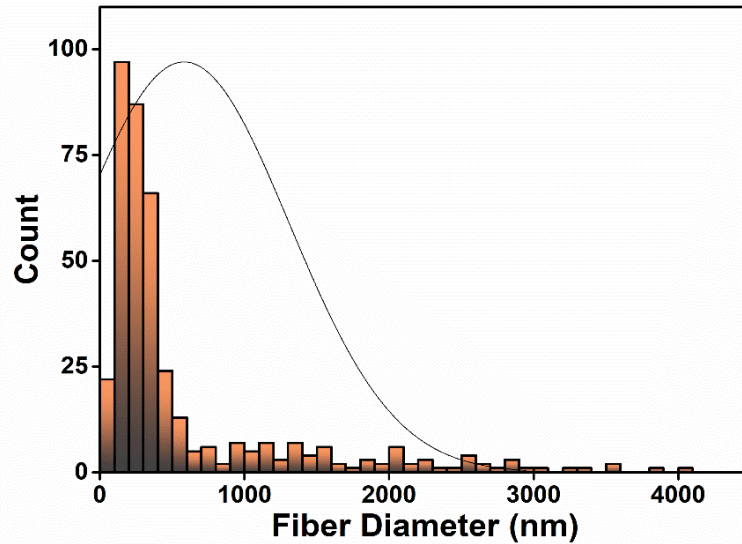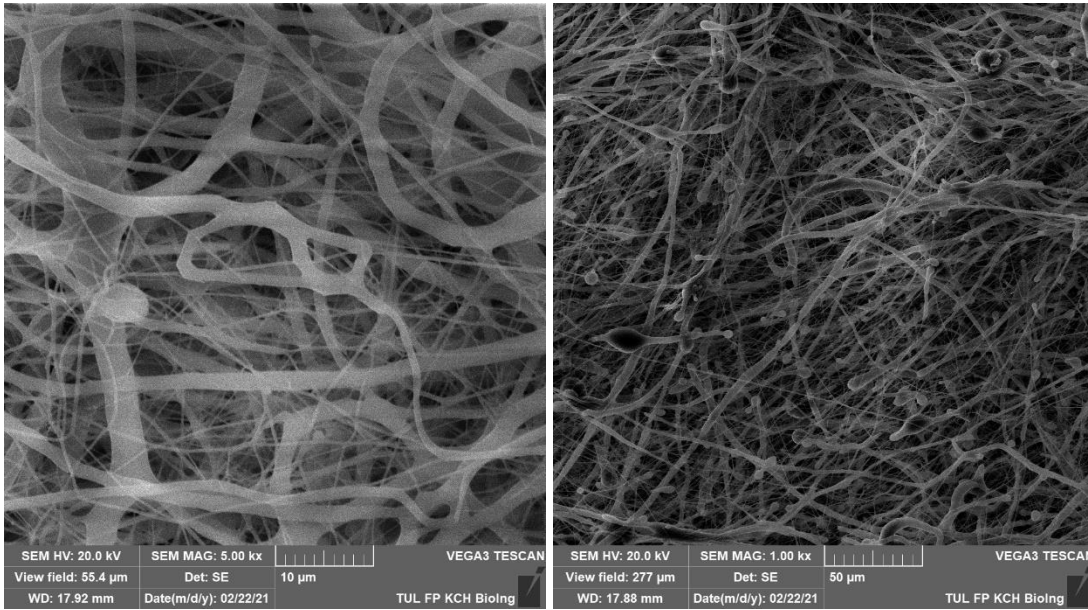
Figure 28.SEM image of PCL-45 electrspun in July 2020 with scale bar 10micrometer on left side and 50micrometer on right side and graphical representation of fiber diameter
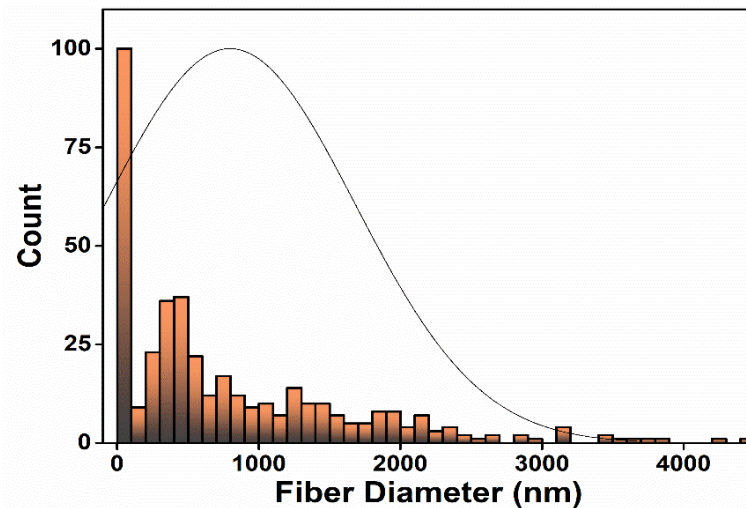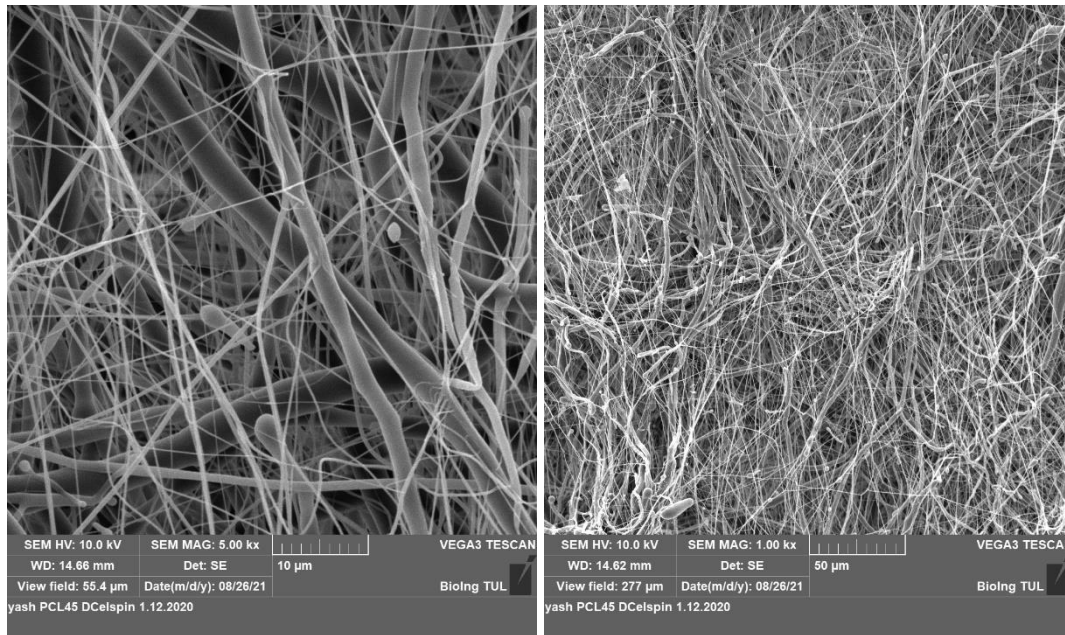
Figure 29.SEM image of PCL-45 electrospun in December 2020 with scale bar 10micrometer on left side and 50micrometer on right side and graphical representation of fiber diameter
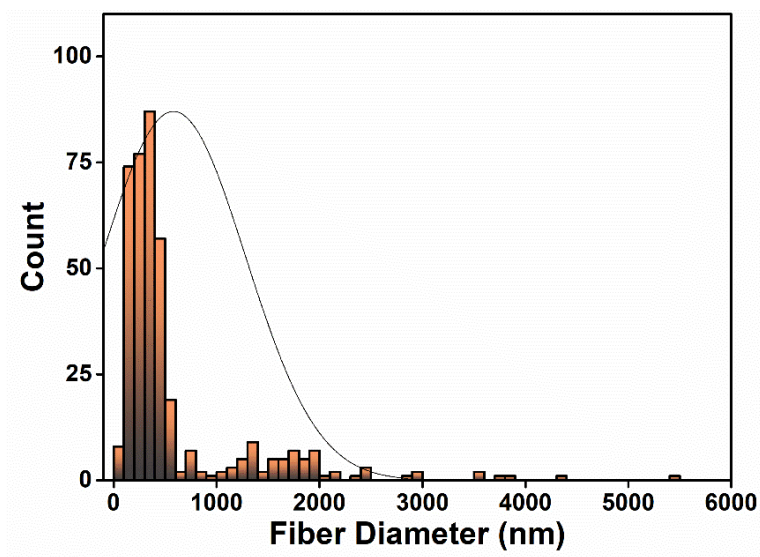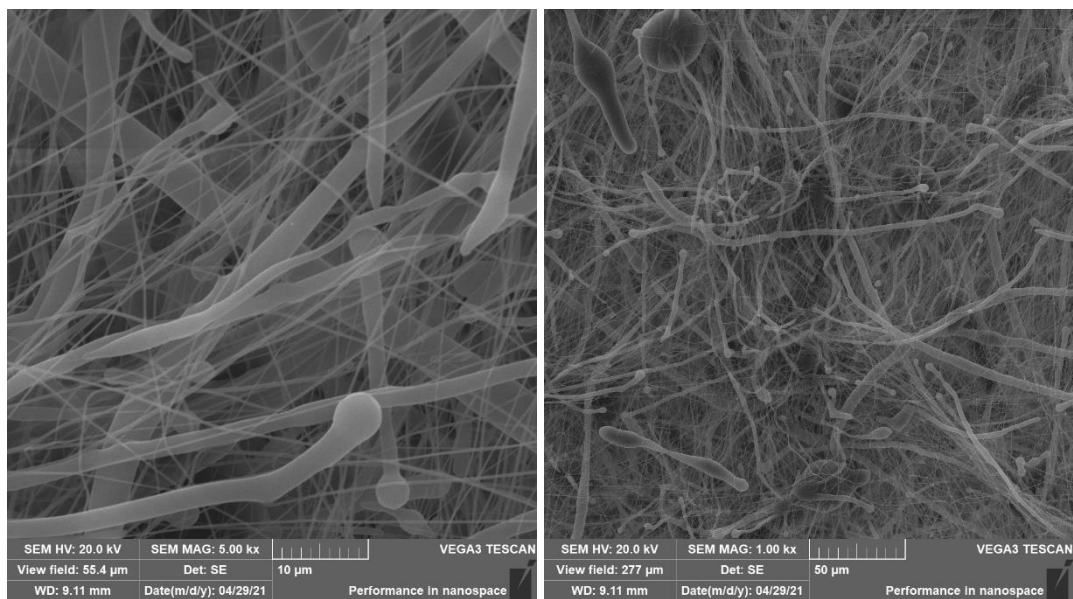
Figure 30.SEM image of PCL-45 electrospun in April 2021 with scale bar 10micrometer on left side and 50micrometer on right side and graphical representation of fiber diameter

4.1.2. Fiber diameter calculation

After collecting SEM images the fiber diameter is calculated with the help of FIJI-ImageJ. The Average of 400 readings taken from the different samples. Calculated results of SEM images listed in the table 5 and figure 31 is graphical representation of fibre diameter electrospun PCL-45 Produced at July 2020 , December 2020 and April 2021.

Table 5. Diameter calculations of SEM images .

Sample	Mean (nm)	Standard deviation (nm)	95%confidence interval (nm)
July 2020	583.52	726.76	71.43
December 2020	798.41	881.57	86.65
April 2021	582.17	699.69	68.77
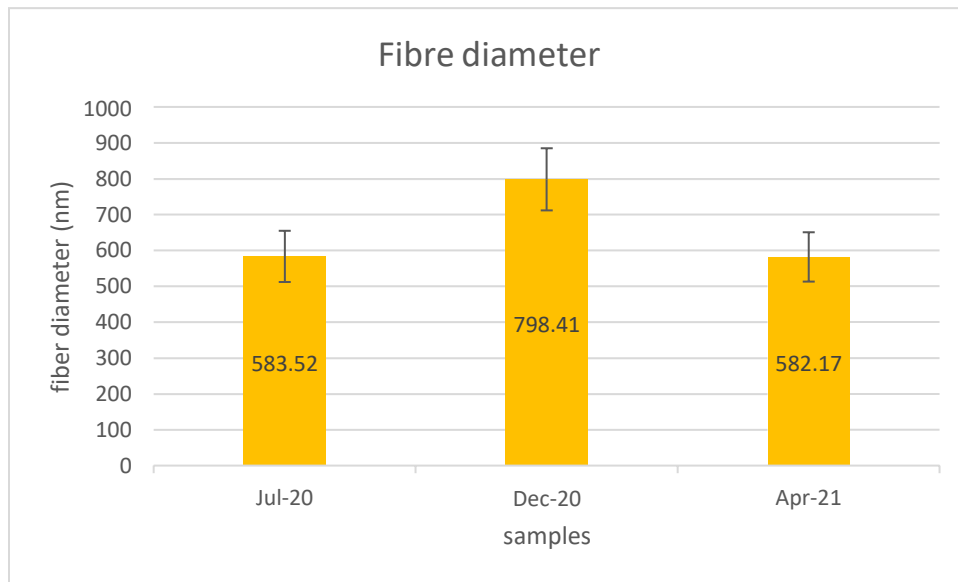
Figure 31. Graphical representation of surface density with 95% CI error bar (July2020) Samples with exact value of surface density in the center

4.1.3. Surface density of the fiber

Before wettability testing of each sample the weight of the sample is calculated and from that the surface density of the sample is calculated. It is interesting to see that how the sample behaves as storage time increases. Also it is necessary to check whether each sample have same density or not. And it helps to explain the trends in the result of wetting of sample.

Table 6. Calculation of the Surface density of sample from December 2020

Samples from December 2020	Weight (gm) (Average)	Surface density (g/m²)	Standard Deviation (g/m²)	95% Confidence Interval
DAY 1	0.043	36.33	15.25	10.24
DAY 3	0.057	47.75	16.91	11.36
DAY 7	0.06	55	19.24	12.93
DAY 14	0.081	67.67	18.06	12.13

Figure 32.Graphical representation of surface density with 95% CI error bar (December 2020) Samples with exact value of surface density in the center

Table 7. Calculation of the Surface density of sample from April 2021

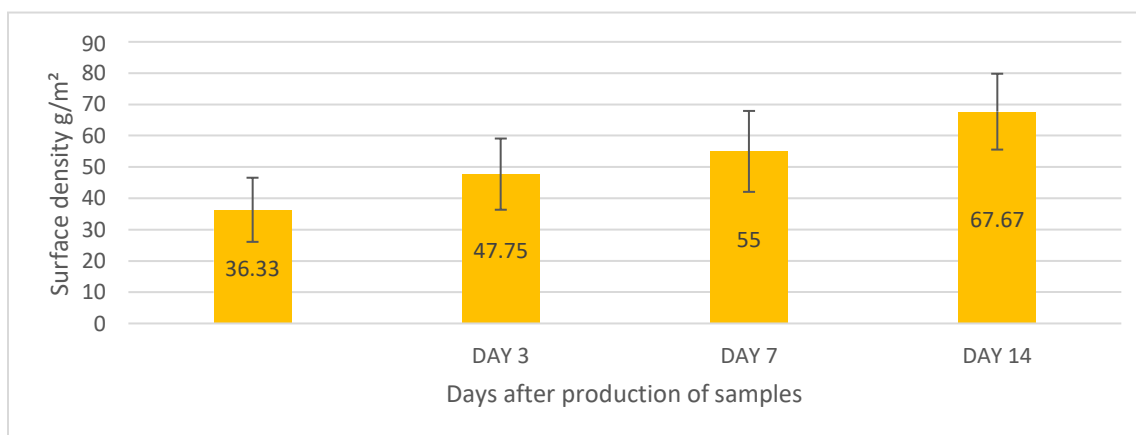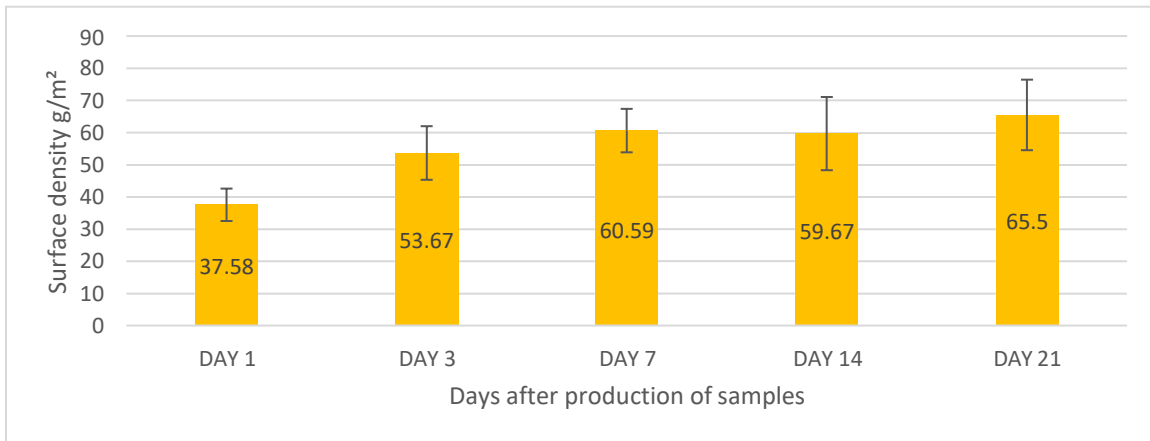| Samples from April 2021	Weight (gm) (Average)	Surface density (g/m²)	Standard Deviation (g/m²)	95% Confidence Interval (g/m²)
DAY 1	0.045	37.58	7.52	5.05
DAY 3	0.064	53.67	12.39	8.32
DAY 7	0.072	60.59	10.06	6.76
DAY 14	0.071	59.67	16.95	11.39
DAY 21	0.078	65.5	16.34	10.98

Figure 33.Graphical representation of surface density with 95% CI error bar (April 2020) Samples with exact value of surface density in the center

Table 8.Calculation of the Surface density of sample stored at -80°C from July 2020 and tested at December 2020 and April 2021

Samples stored at -80°C from 9/7/2020 Measured at	Weight (g) (Average)	Surface density (g/m²)	Standard Deviation (g/m²)	95% Confidence Interval (g/m²)
9/7/2020	0.057	47.82	3.71	2.65
10/12/2020	0.05	41.66	1.36	0.97
15/4/2021	0.052	43.25	2.20	1.57
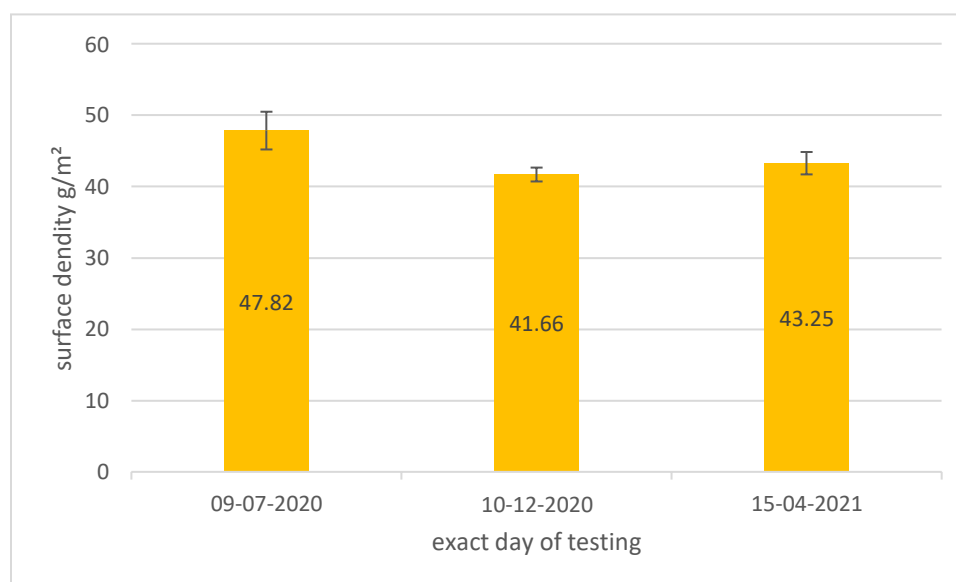
Figure 34.graphical representation of the Surface density of sample stored at -80°C from July 2020 and tested at December 2020 and April 2021with error bar of 95% CI and exact values of surface density at center

4.1.4. Wetting calculations

Here, the presentation of graphs from the dynamic wetting test performed on Force Micro tensiometer Krüss K-100 are shown in figures. All the graphs displayed here are average of 10 individual measurements.
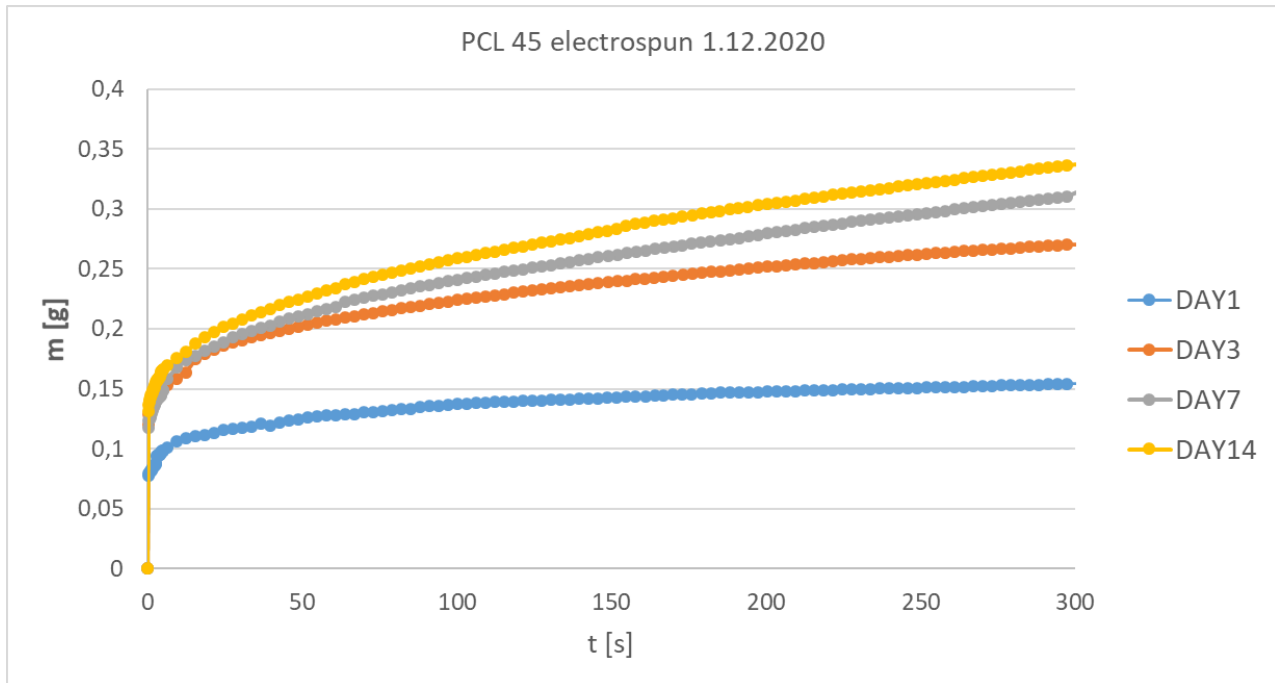
Figure 35.Graph of average data from water wicking measurement by force microtensiometer Krüss K100 for PCL-45 electrospun in December 2020 (each single line is the average of 10 reading*)*
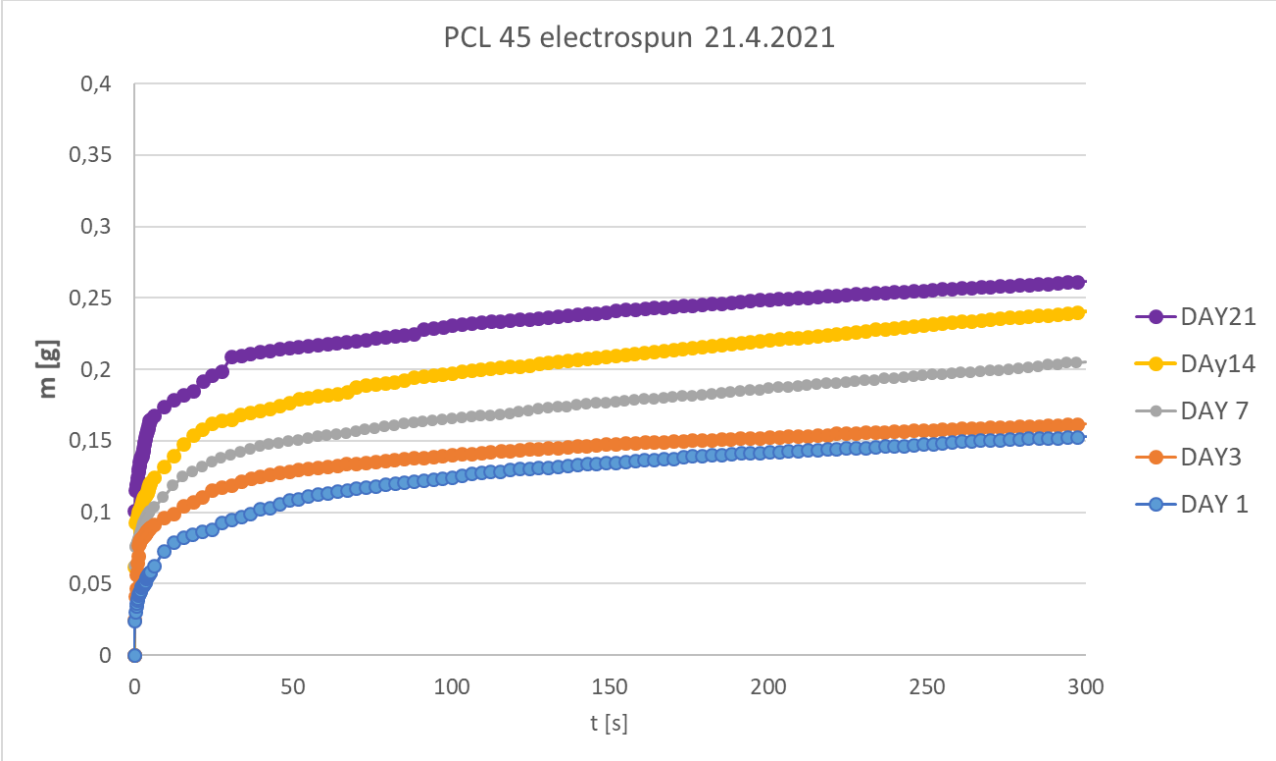
Figure 36.Graph of average data from water wicking measurement by force micro tensiometer Krüss K100 for PCL-45 electrospun in April 2021(each single line is the average of 10 reading)

***Figure 37.Graph of average data from water wicking measurement by force micro tensiometer
Krüss K100 for PCL-45 electrospun in April 2021 with normal sample and Ethelene oxide
treated for sterilization (each single line is the average of 10 reading)***

The initial phase of water wicking into the nanofibrous materials is leaded by the capillary force
and the dynamic can be described according Lucas-Washburn equation. There is known that for
small time the gravitational part of equation can be neglected and the linear dependence should
exist according equation $m^2=At$, where m is mass of water wicked into the nanofibrous material; t
is time and A is constant including material and liquid parameters. Graphs in figure 38 and figure
39 shows the initial part of water wicking into PCL45 electrospun layers produced in December
2020 and April 2021.
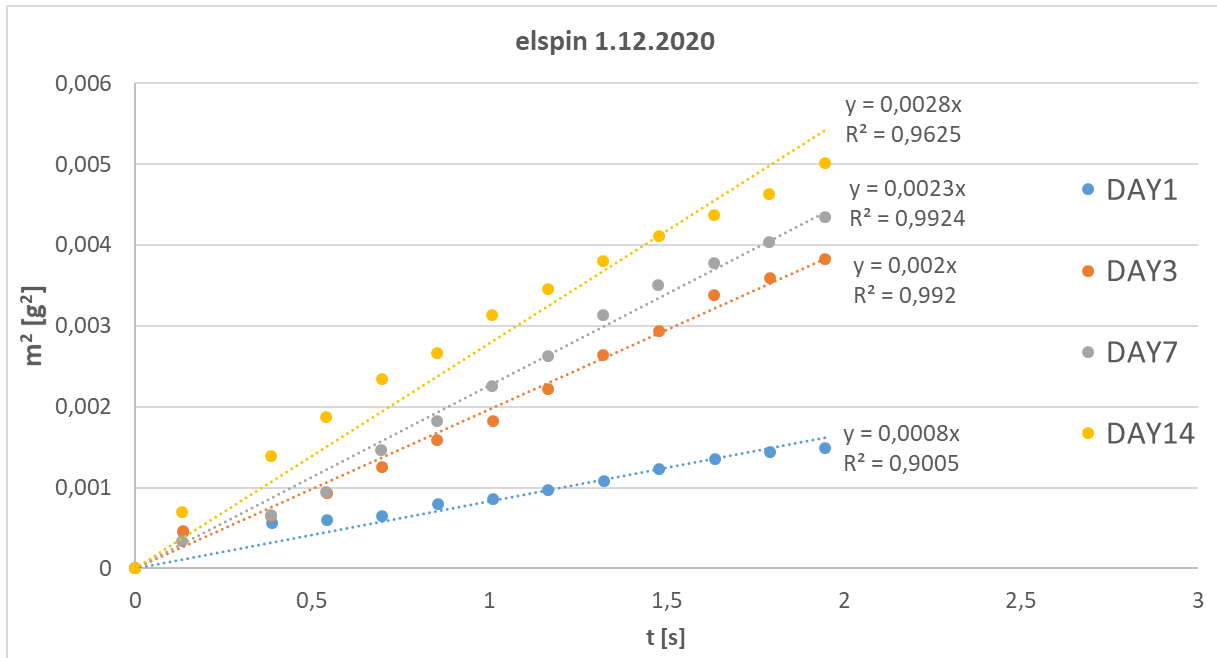
Figure 38.Graph representing initial part of water wicking into electrospun PCL45 nanofibrous material in different days after electrospinning in December 2020. Each curve is representing by linear regression equation and squared regression coefficient.
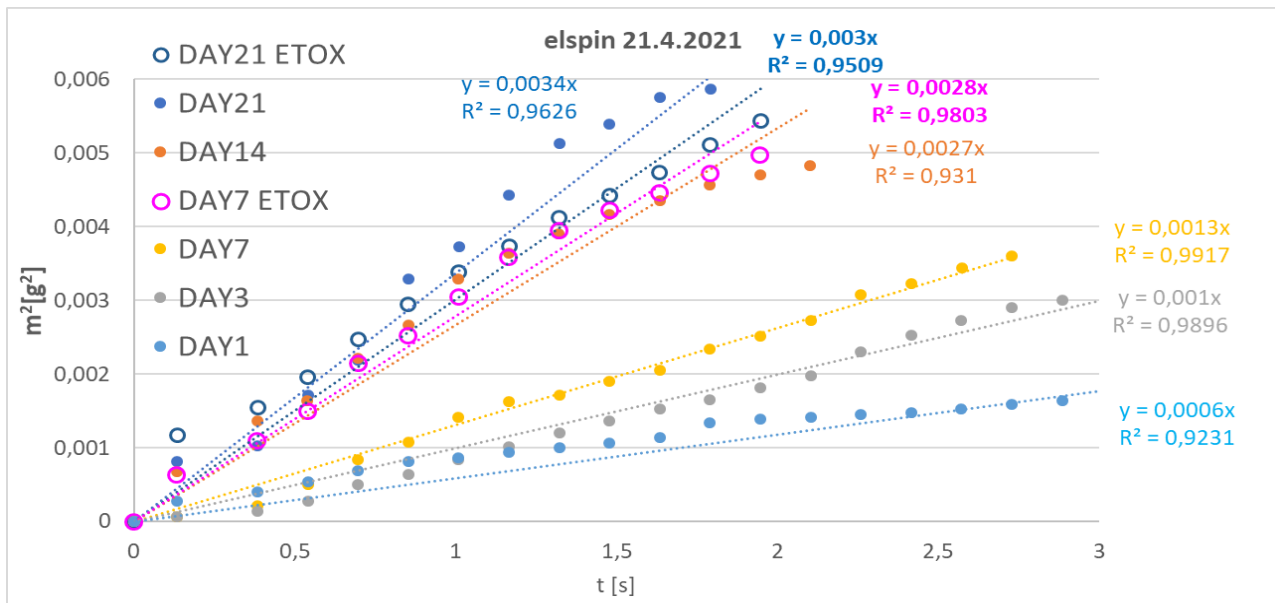
Figure 39.Graph representing initial part of water wicking into electrospun PCL45 nanofibrous material in different days after electrospinning in April 2021 and with sterilization by ethylenoxide. Each curve is representing by linear regression equation and square.

DAY	A [g²/s]					
	elspin 1.12.2020	R	elspin 21.4.2021	R	Ethylenoxide sterilization 21.4.2021	R
1	0.0008	0.949	0.0006	0.961		
3	0.002	0.996	0.001	0.995		
7	0.0023	0.996	0.0013	0.996	0.0028	0.990
14	0.0028	0.981	0.0027	0.965		
21			0.0034	0.981	0.003	0.975
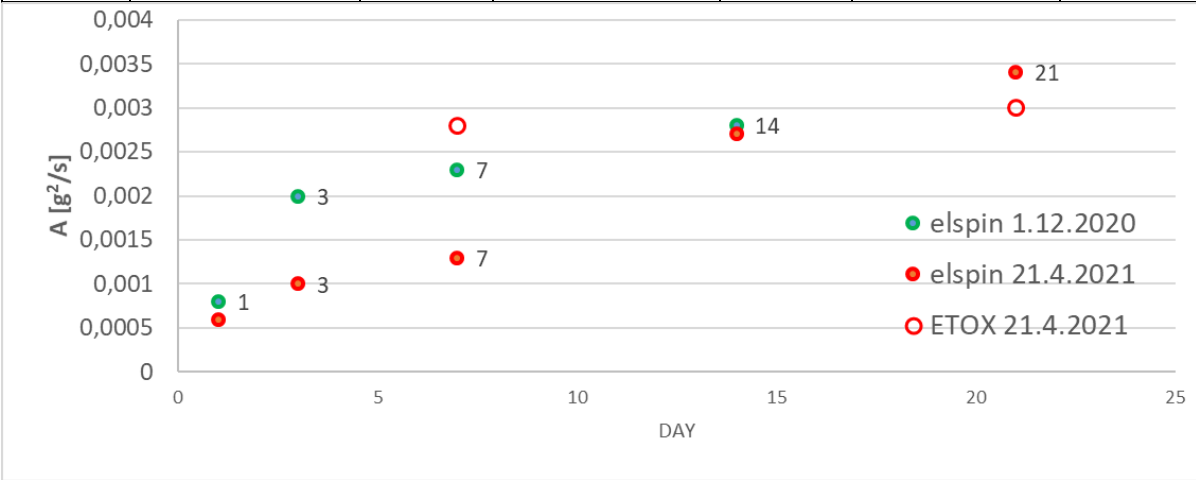
Figure 40.Table and graph representing coefficient A calculated from graphs of dependence of squared wicked mass of water into electrospun PCL nanofibrous layer (i: produced in December 2020; ii: produced in April 2021; iii: produced in April 2021 and sterilized

4.2 Discussion

The electrospinning of PCL45 was successful at Nanospider™ and the layer of produced material is uniform, no major defects as droplets or holes are observed on the samples. From table 5 and figure 31, there is seen that the diameter of the electrospun fibers produced at different time is not same which was not predicted, even all process and material parameters of electrospinning were kept same. This may have occurred because of the difference in original raw material used for electrospinning, which is taken from different bottles and which may contain the variation in the molecular weight or variation in distribution of molecular weight. To avoid this it was possible to test the granules with gel chromatography but it is expensive and time spending so it was not done.

Surface density of the sample was measured before each wettability testing, and it was observed that as time changes the surface density values are increased which is calculated in table 6 and table 7 but sample which are kept at -80°C on July 2020 and tested at December 2020 and April 2021 shown almost similar values as explained in table 8 and figure 34 From this results of surface density changes it seems when sample stored in the room temperature or even in more temperature like incubator the surface density of the fabric is changed but the surface density of the samples which are stores at -80°C is not changed because below glass transition temperature (T_g for PCL is -60°C) sample restricts all the moments, so there will be no or not observed absorption of water into fibers neither on the surface of material from air humidity. The surface density values for the samples which produced at July 2020 and stored at -80°C which tested on December 2020 and April 2021 shown nearly the same value which shown exactly similar trend in wetting measurement of the sample which is seen in table 8 and figure 34.

For the wetting testing samples produced in the December 2020 shown that change in the wettability of the sample as the storage time is increases. From the figure 35 there is clearly seen that as storing time increases the water wettability of the material also increases because some structural changes are happening inside or the surface of the material and the change in the water wettability is relatively high.

For the samples which are produced in the April 2021 also shown the change in the wettability of the samples as storage time increases. From the figure 36 there is seen that as time increases the water wettability of the material increases but the change in the wettability is not so high as it shown for the samples from the December 2020.

The difference in the change in the wettability of the samples from December 2020 and April 2021 is maybe because of the change in the room temperature of the samples or the variation in the diameter of fibre because this the trend in the wetting of this samples is different from each other (see Table 5). When there is compared the DAY-1 testing of both samples prepared on December 2020 and April 2021 is almost same but after that the sample from April 2021 shows different trend.

Figure 37 shows the wetting of material after sterilization with ethylene oxide and it is seen that the water wettability of the sample is changed drastically as compared with untreated sample at DAY-7, this could be because of the oxidation the sample become the more hydrophilic. This samples observed after day 7 and day 21 which shows nearly same results of the wetting, so from the figure 37 it shows that once the wettability changes after sterilization then it remains same but it is not final statement because it may change after that also but here it is not tested further.

There is obvious from figure 38 and figure 39, the rate of water wicking (penetration) into the electrospun PCL45 layers representing by constant A increased with aging of the electrospun material respectively with the increasing number of days after the initial electrospinning. The claim can be explained by changing of the electrospun PCL45 nanofibrous surface depending of the storing time. The figure 40 show a little bit different development of constant A in time for materials electrospun in December 2020 and April 2021, what can be explained by different fibre diameter in these materials and different ambient conditions connected for example with heating in laboratories during Winter or higher humidity in Spring. The similar experiment with controlled ambient conditions at room temperature for example 20°C and 60%RH is suggested as the future research.

Ethylene oxide sterilization yielded interesting results. From the wetting measurements on the seventh day after spinning, the increase in wettability with water is evident compared to the material without sterilization. However, on the twenty-first day after spinning, the water

70

wettability measurement already showed almost comparable, if not lower, results. Therefore, a detailed analysis is recommended focusing on the effect of ethylene oxide sterilization on the wettability of PCL electrospun materials with water and its development over time after individual days from sterilization. This is also in connection with the fact that there is no complete agreement among the professional public on how long to let the materials sterilized by ethylene oxide so-called aeration before their further application in medicine or for biological tests (7 or 14 days). even if standard exists (ISO 10993-7:2008).

In the future work , it is suggestion that to check the wetting property of the material with different storing conditions such as -20°C normal freezer, 8°C fridge, in incubator with 97% relative humidity and so on, to see the exact wetting behaviour of the electrospun PCL-45 or also to the second widely used PCL with higher molecular weight M_n 80000.

5. Conclusion

This diploma thesis proves that electrospun PCL-45 material change its water wetting properties in wicking testing as storing time increases. The exact reason behind changing of wetting properties is clearly unknown and has to be studied. Humidity and temperature as a storing conditions changes the water wetting property of the electrospun PCL-45 it means that material start changes from hydrophobic to hydrophilic, so for the storage of the material it must be considered. Electrospun PCL-45 changes the wetting property in the room temperature as well as in the incubator but the exact trend of the change in wettability is not observed and it is suggested to study the topic in detail with exact controlled conditions. For the storage of electrospun PCL-45 at -80°C is the condition, where no changes in surface density and water wettability was found, apparently because at this temperature PCL is bellow glass transition temperature.

The phenomena studied in the diploma thesis are very interesting and showed not easy explained results. There is necessary to deeply study the interesting water wettability of PCL electrospun materials with precise controlled conditions and explain the internal of surface changes which happen over time.

6.Reference List

[1] N. Bhardwaj and S. C. Kundu, 'Electrospinning: A fascinating fibre fabrication technique', *Biotechnology Advances*, vol. 28, no. 3, pp. 325–347, May 2010, doi: 10.1016/j.biotechadv.2010.01.004.

[2] S. Thenmozhi, N. Dharmaraj, K. Kadirvelu, and H. Y. Kim, 'Electrospun nanofibers: New generation materials for advanced applications', *Materials Science and Engineering: B*, vol. 217, pp. 36–48, Mar. 2017, doi: 10.1016/j.mseb.2017.01.001.

[3] S. Mohammadzadehmoghadam *et al.*, 'Electrospinning: Current Status and Future Trends', in *Nano-size Polymers*, S. Fakirov, Ed. Cham: Springer International Publishing, 2016, pp. 89–154. doi: 10.1007/978-3-319-39715-3_4.

[4] Z. Li and C. Wang, 'Effects of Working Parameters on Electrospinning', in *One-Dimensional nanostructures*, Berlin, Heidelberg: Springer Berlin Heidelberg, 2013, pp. 15–28. doi: 10.1007/978-3-642-36427-3_2.

[5] A. Haider, S. Haider, and I.-K. Kang, 'A comprehensive review summarizing the effect of electrospinning parameters and potential applications of nanofibers in biomedical and biotechnology', *Arabian Journal of Chemistry*, vol. 11, no. 8, pp. 1165–1188, Dec. 2018, doi: 10.1016/j.arabjc.2015.11.015.

[6] Y.-Z. Long, X. Yan, X.-X. Wang, J. Zhang, and M. Yu, 'Chapter 2 - Electrospinning: The Setup and Procedure', in *Electrospinning: Nanofabrication and Applications*, B. Ding, X. Wang, and J. Yu, Eds. William Andrew Publishing, 2019, pp. 21–52. doi: 10.1016/B978-0-323-51270-1.00002-9

[7] Maheshwari, S., & Chang, H. (2009). Assembly of Multi-Stranded Nanofiber Threads through AC Electrospinning. *Advanced Materials*, *21*(3), 349-354. doi: 10.1002/adma.200800722

[8] J. Fang, H. Niu, T. Lin, and X. Wang, 'Applications of electrospun nanofibers', *Chinese Science Bulletin*, vol. 53, pp. 2265–2286, Aug. 2008, doi: 10.1007/s11434-008-0319-0.

[9] T. V. Shah and D. V. Vasava, 'A glimpse of biodegradable polymers and their biomedical applications', *e-Polymers*, vol. 19, no. 1, pp. 385–410, Jun. 2019, doi: 10.1515/epoly-2019-0041.

[10] Katarzyna Serwańska-Leja and Grażyna Lewandowicz, 'Polymer Biodegradation and Biodegradable Polymers - a Review', *Polish Journal of Environmental Studies*, vol. 19(2), pp. 255–266, Nov. 2009.

[11] E. Piskin, 'Biodegradable polymers as biomaterials', *Journal of Biomaterials Science, Polymer Edition*, vol. 6, no. 9, pp. 775–795, Jan. 1995, doi: 10.1163/156856295X00175.

[12] F. Asghari, M. Samiei, K. Adibkia, A. Akbarzadeh, and S. Davaran, 'Biodegradable and biocompatible polymers for tissue engineering application: a review', *Artificial Cells, Nanomedicine, and Biotechnology*, vol. 45, no. 2, pp. 185–192, Feb. 2017, doi: 10.3109/21691401.2016.1146731.

[13] D. Kai, S. S. Liow, and X. J. Loh, 'Biodegradable polymers for electrospinning: Towards biomedical applications', *Materials Science and Engineering: C*, vol. 45, pp. 659–670, Dec. 2014, doi: 10.1016/j.msec.2014.04.051.

[14] D. Mondal, M. Griffith, and S. S. Venkatraman, 'Polycaprolactone-based biomaterials for tissue engineering and drug delivery: Current scenario and challenges', *International Journal of Polymeric Materials and Polymeric Biomaterials*, vol. 65, no. 5, pp. 255–265, Mar. 2016, doi: 10.1080/00914037.2015.1103241.

[15] O. Suwantong, 'Biomedical applications of electrospun polycaprolactone fiber mats: Biomedical Applications of Electrospun Polycaprolactone Fiber Mats', *Polym. Adv. Technol.*, vol. 27, no. 10, pp. 1264–1273, Oct. 2016, doi: 10.1002/pat.3876.

[16] M. Labet and W. Thielemans, 'Synthesis of polycaprolactone: a review', *Chem. Soc. Rev.*, vol. 38, no. 12, p. 3484, 2009, doi: 10.1039/b820162p.

[17] M. A. Woodruff and D. W. Hutmacher, 'The return of a forgotten polymer—Polycaprolactone in the 21st century', *Progress in Polymer Science*, vol. 35, no. 10, pp. 1217–1256, Oct. 2010, doi: 10.1016/j.progpolymsci.2010.04.002.

[18] D. Kai, S. S. Liow, and X. J. Loh, 'Biodegradable polymers for electrospinning: Towards biomedical applications', *Materials Science and Engineering: C*, vol. 45, pp. 659–670, Dec. 2014, doi: 10.1016/j.msec.2014.04.051.

[19] E. Malikmammadov, T. E. Tanir, A. Kiziltay, V. Hasirci, and N. Hasirci, 'PCL and PCL-based materials in biomedical applications', *Journal of Biomaterials Science, Polymer Edition*, vol. 29, no. 7–9, pp. 863–893, Jun. 2018, doi: 10.1080/09205063.2017.1394711.

[20] B. Joseph, R. Augustine, N. Kalarikkal, S. Thomas, B. Seantier, and Y. Grohens, 'Recent advances in electrospun polycaprolactone based scaffolds for wound healing and skin bioengineering applications', *Materials Today Communications*, vol. 19, pp. 319–335, Jun. 2019, doi: 10.1016/j.mtcomm.2019.02.009.

[21] O. Suwantong, 'Biomedical applications of electrospun polycaprolactone fiber mats: Biomedical Applications of Electrospun Polycaprolactone Fiber Mats', *Polym. Adv. Technol.*, vol. 27, no. 10, pp. 1264–1273, Oct. 2016, doi: 10.1002/pat.3876.

[22] M. Janmohammadi and M. S. Nourbakhsh, 'Electrospun polycaprolactone scaffolds for tissue engineering: a review', *International Journal of Polymeric Materials and Polymeric Biomaterials*, vol. 68, no. 9, pp. 527–539, Jun. 2019, doi: 10.1080/00914037.2018.1466139.

[23] J. Fang, H. Niu, T. Lin, and X. Wang, 'Applications of electrospun nanofibers', *Sci. Bull.*, vol. 53, no. 15, pp. 2265–2286, Aug. 2008, doi: 10.1007/s11434-008-0319-0.

[24] I. Castilla-Cortázar, J. Más-Estellés, J. M. Meseguer-Dueñas, J. L. Escobar Ivirico, B. Marí, and A. Vidaurre, 'Hydrolytic and enzymatic degradation of a poly(ε-caprolactone) network', *Polymer Degradation and Stability*, vol. 97, no. 8, pp. 1241–1248, Aug. 2012, doi: 10.1016/j.polymdegradstab.2012.05.038.

[25] Takanori HAYASHI and Hiroyoshi KANAI, 'Enzymatic Degradation of Poly(e-caprolactone) Fibers in vitro', *Polymer Journal,* vol. Vol. 33, No. I, pp. pp38-41, Sep. 2000.

[26] G. Agrawal, Y. S. Negi, S. Pradhan, M. Dash, and S. K. Samal, 'Wettability and contact angle of polymeric biomaterials', in *Characterization of Polymeric Biomaterials*, Elsevier, 2017, pp. 57–81. doi: 10.1016/B978-0-08-100737-2.00003-0.

[27] A. Patnaik, R. S. Rengasamy, V. K. Kothari, and A. Ghosh, 'Wetting and Wicking in Fibrous Materials', *Textile Progress*, vol. 38, no. 1, pp. 1–105, Jan. 2006, doi: 10.1533/jotp.2006.38.1.1.

[28] L.-C. Xu and C. A. Siedlecki, 'Effects of surface wettability and contact time on protein adhesion to biomaterial surfaces', *Biomaterials*, vol. 28, no. 22, pp. 3273–3283, Aug. 2007, doi: 10.1016/j.biomaterials.2007.03.032.

[29] Y. Arima and H. Iwata, 'Effect of wettability and surface functional groups on protein adsorption and cell adhesion using well-defined mixed self-assembled monolayers', *Biomaterials*, vol. 28, no. 20, pp. 3074–3082, Jul. 2007, doi: 10.1016/j.biomaterials.2007.03.013.

[30] R. S. Kurusu and N. R. Demarquette, 'Surface modification to control the water wettability of electrospun mats', *International Materials Reviews*, vol. 64, no. 5, pp. 249–287, Jul. 2019, doi: 10.1080/09506608.2018.1484577.

[31] A. Cipitria, A. Skelton, T. R. Dargaville, P. D. Dalton, and D. W. Hutmacher, 'Design, fabrication and characterization of PCL electrospun scaffolds—a review', *J. Mater. Chem.*, vol. 21, no. 26, p. 9419, 2011, doi: 10.1039/c0jm04502k.

[32] P. Ginestra, E. Ceretti, and A. Fiorentino, 'Electrospinning of Poly-caprolactone for Scaffold Manufacturing: Experimental Investigation on the Process Parameters Influence', *Procedia CIRP*, vol. 49, pp. 8–13, 2016, doi: 10.1016/j.procir.2015.07.020.

[33] M. J. Mochane, T. S. Motsoeneng, E. R. Sadiku, T. C. Mokhena, and J. S. Sefadi, 'Morphology and Properties of Electrospun PCL and Its Composites for Medical Applications: A Mini Review', *Applied Sciences*, vol. 9, no. 11, p. 2205, May 2019, doi: 10.3390/app9112205.

[34] L. Van der Schueren, B. De Schoenmaker, Ö. I. Kalaoglu, and K. De Clerck, 'An alternative solvent system for the steady state electrospinning of polycaprolactone', *European Polymer Journal*, vol. 47, no. 6, pp. 1256–1263, Jun. 2011, doi: 10.1016/j.eurpolymj.2011.02.025.

[35] A. Martins *et al.*, 'Surface Modification of Electrospun Polycaprolactone Nanofiber Meshes by Plasma Treatment to Enhance Biological Performance', *Small*, p. NA-NA, Feb. 2009, doi: 10.1002/smll.200801648.

[36] F. Yang, J. G. C. Wolke, and J. A. Jansen, 'Biomimetic calcium phosphate coating on electrospun poly(ε-caprolactone) scaffolds for bone tissue engineering', *Chemical Engineering Journal*, vol. 137, no. 1, pp. 154–161, Mar. 2008, doi: 10.1016/j.cej.2007.07.076.

[37] E. V. Melnik *et al.*, 'In vitro degradation behaviour of hybrid electrospun scaffolds of polycaprolactone and strontium-containing hydroxyapatite microparticles', *Polymer*

Degradation and Stability, vol. 167, pp. 21–32, Sep. 2019, doi: 10.1016/j.polymdegradstab.2019.06.017.

[38] Š. Zupančič *et al.*, 'Impact of PCL nanofiber mat structural properties on hydrophilic drug release and antibacterial activity on periodontal pathogens', *European Journal of Pharmaceutical Sciences*, vol. 122, pp. 347–358, Sep. 2018, doi: 10.1016/j.ejps.2018.07.024.

[39] R. M. Aghdam, S. Najarian, S. Shakhesi, S. Khanlari, K. Shaabani, and S. Sharifi, 'Investigating the effect of PGA on physical and mechanical properties of electrospun PCL/PGA blend nanofibers', *J. Appl. Polym. Sci.*, vol. 124, no. 1, pp. 123–131, Apr. 2012, doi: 10.1002/app.35071.

[40] M. I. Hassan1, L. H. Chong1 and N. Sultana1, 2, 'WETTABILITY AND WATER UPTAKE PROPERTIES OF PLA AND PCL/GELATIN-BASED ELECTROSPUN SCAFFOLDS', *ARPN Journal of Engineering and Applied Sciences*, vol. 11, no. 2016.

[41] H. Jeon and G. Kim, 'Preparation and characterization of an electrospun polycaprolactone (PCL) fibrous mat and multi-layered PCL scaffolds having a nanosized pattern-surface for tissue regeneration', *J. Mater. Chem. B*, vol. 2, no. 2, pp. 171–180, 2014, doi: 10.1039/C3TB21230K.

[42] S. Surucu, K. Masur, H. Turkoglu Sasmazel, T. Von Woedtke, and K. D. Weltmann, 'Atmospheric plasma surface modifications of electrospun PCL/chitosan/PCL hybrid scaffolds by nozzle type plasma jets for usage of cell cultivation', *Applied Surface Science*, vol. 385, pp. 400–409, Nov. 2016, doi: 10.1016/j.apsusc.2016.05.123.

[43] N. E. Zander, J. A. Orlicki, A. M. Rawlett, and T. P. Beebe, 'Quantification of Protein Incorporated into Electrospun Polycaprolactone Tissue Engineering Scaffolds', *ACS Appl. Mater. Interfaces*, vol. 4, no. 4, pp. 2074–2081, Apr. 2012, doi: 10.1021/am300045y.

[44] ElectrospinTech. 2021. *Coaxial Electrospinning: The Spinneret*. [online] Available at:

https://electrospintech.wordpress.com/2014/04/26/coaxial-electrospinning-the-spinneret/ [Accessed 18 August 2021].

[45] Maheshwari, S., & Chang, H. (2009). Assembly of Multi-Stranded Nanofiber Threads through AC Electrospinning. *Advanced Materials*, *21*(3), 349-354. doi: 10.1002/adma.200800722