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EFFECT OF POLYMER CONCENTRATION AND BLEND RATIO ON THE MORPHOLOGY AND WOUND HEALING EFFICIENCY OF ELECTROSPUN PULLULAN/PVA NANOFIBERS

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ABSTRACT

Electrospinning was used to produce nanofibers made of pullulan/poly (vinyl alcohol) (PVA) from their aqueous solutions. The study examined whether varying polymer concentrations and mix ratios affect the morphology and properties of pullulan/PVA nanofibers. The total polymer concentrations were also varied at a given blend ratio to determine the best electrospinning conditions for producing uniform and bead-free nanofibers. Fourier transform infrared spectroscopy (FTIR), X-ray Diffraction (XRD), Scanning electron microscopy (SEM) and Differential scanning calorimetry (DSC) were used to characterise the nanofibers. The mechanical qualities were also investigated using tensile strength testing. A 10% total polymer concentration produced uniform and bead-free pullulan/PVA nanofibers with greater mechanical and thermal properties. *In vitro* assay was performed to access the wound healing. Pullulan/PVA shows wound healing factors in fibroblast cell line (L929). Pullulan/PVA/Gentamicin according to the wound dressing 10% is the most efficient achieving 90% after 5 h of use while maintaining an approximate 70% absorption rate.

KEYWORDS: Electrospinning, Wound dressing, Poly (vinyl Alcohol), Pullulan, Drug delivery.

INTRODUCTION

Since electrospinning is an easy and successful way to produce nanofibers which have been demonstrated to be beneficial in a number of applications, such as biomedical engineering, filtration, protective apparel, catalysis reactions and sensors it has attracted the interest of both academic and industrial scientists.[1] Electrospinning involves applying an electric field to a polymer solution that is kept stationary at the end of a capillary tube by surface tension. The electric field induces electrons to accumulate on the liquid's surface. Mutual charge repulsion produces a force that is distinct from surface tension. The hemispherical surface of the solution at the capillary tube tip elongates as the electric field intensity grows, resulting in a conical taylor cone shape. Nanoparticle surfaces may be changed and scanned, whereas nanofibrous materials with a large porous surface area are ideal for medicinal applications.[2] Another technique for loading active pharmaceutical ingredients for medication release is electrospinning. For the delivery of drugs, coaxial electrospinning is a widely utilised technology. Polymer nanofibers with their invisible properties including enhanced surface area per volume and surface characteristics hold promise for a range of biological applications. To reduce adverse effects medications can be released gradually with the help of nanofiber composites that are filled with pharmaceuticals. In drug delivery systems, the compatibility of the polymer with the chemical behaviour of the medication is important.[3] The conventional technique for creating nanofibers with different morphologies is electrospinning, which is also a productive way of producing nanofiber matrices. Poly (vinyl alcohol) is a semi-crystalline, hydrophilic polymer that is chemically and thermally stable. It is an extremely biocompatible, non-toxic polymer that has excellent water permeability that is also simple to produce. PVA is widely utilised in the food, pharmaceutical, medical, cosmetic and packaging industries due to its ability to form physical gels in a wide range of solvents. It is a typical synthetic polymer that is with other materials with weak physical properties to improve their due to its high flexibility and durability.[4] Because PVA with functional groups is simple to prepare as a bulk material, films and fibers, it is helpful in practical studies of functional polymers. Figure 1 illustrates the way electrospinning nanofibers is used to make polymer. Natural polymers such as pullulan are becoming increasingly important because to their biocompatibility and usage as proteins in biotechnological materials and biomedical applications. The yeast-like fungus *Aureobasidium pullulans* produces pullulan an extracellular microbial polysaccharide. The linear a-glucan structure consists of three glucose units connected by α-(1,4) and maltotriose units joined by α-(1,6). Pullulan's amazing properties make it a popular low-calorie food component, gelling

agent, food and pharmaceutical coating and packaging ingredient, fertiliser binder, and tablet oxidation inhibitor.[5] Contact lenses, plywood, biodegradable foil, water solubility enhancer and enhanced oil recovery are some of the other applications. A further technique for loading active pharmaceutical ingredients for these releases is electrospinning which results in tiny fibers with specific surface features in the sub-micron to nanometer range. The appropriate polymer is dissolved in a solvent and exposed to a high-voltage electric field to create nanofibers. A method has been developed that involves applying a strong electric field to a solution containing the required polymer or melting the polymer and exposing it to the electric field if there is not enough solvent to make micrometre to nanometer-thick fibers with carefully controlled topography. Polymer nanofibers are promising options for many important biological applications because of their exceptional qualities. Polymer fibers with a diameter of less than a micrometre or nanoscale may exhibit hitherto unobserved properties like flexibility in surface characteristics and an enhanced surface area to volume ratio. It has previously been demonstrated that a variety of polymers with leukemia-like bioactivities can be used to treat cancer cells. In order to create a targeted and efficient drug delivery system for the treatment of wound dressings the objective of this study was to synthesise pullulan/PVA nanofibers and investigate their possible use in wound dressing by assessing their drug encapsulation and efficiency-controlled release characteristics and effectiveness in inhibiting fibroblast cell growth. Electrospinning techniques are frequently used in drug delivery systems. The feed exchange polymer solution is connected to two needles with two capillaries in the traditional electrospinning procedure.[6] With a nozzle design many pathways are possible for both the external and internal structures. Nozzle clogging may be a concern while handling electro-spraying. A continuous jet with viscoelasticity and bending instability is produced via electrospinning. Numerical analyses reveal that when electrospinning takes place at a jet's surface electrical charges move. Three primary parts make up an electrospinning device: a high-voltage power source a grounded collector attached to the collection plate in between and a metallic needle known as a spinneret. Nanofibers are recognised as having numerous benefits, such as facilitating the development of tissues and the transportation of drugs. When compared to other methods, electrospinning is recommended for the manufacturing of long continuous nanoscale hollow fibers because template directed procedures are only compatible with short fibers. To produce long continuous nanoscale hollow fibers using electrospinning, specific threshold values or ranges can be defined for classifying fibers.[7] For example, a fiber is categorised as long if it is more than a certain length, such as 1 mm. Furthermore, each fiber's aspect ratio, which is

determined by dividing its length by its diameter can be used as a parameter. Fibers with an aspect ratio larger than that can be classified as long, while those with a ratio smaller than that are classified as short provided a minimum value such as 100 is set. It is essential to assess the fiber structure for breaks, discontinuities or abnormalities. Long fibers are distinguished from short fibers by designating a predefined length throughout which the fiber should show continuous continuity, whereas those with breaks or irregularities are categorised as short fibers.[8,9] These quantitative criteria allow for consistent classification of fibers and allow evaluation of the way electrospinning produces continuous long nanoscale hollow fibers.

When gentamicin is developed at high concentrations *in vivo* it may constrict and either kill or prevent the growth of bacteria. To lessen the negative consequences of high toxicity gentamicin can be released gradually over an extended period of time thanks to drug-loaded nanofiber composites.[10] Based on the polymer's compatibility with the drug's chemical behaviour the polymers are chosen to provide consistent loading following manufacture. Gentamicin is used in a nano-drug delivery system with stable nanofiber structure release characteristics and mechanical qualities at pH 7.4 at pH 5, the medication can be released rapidly.

MATERIALS AND METHODS

MATERIALS

Pullulan (food-grade) was manufactured by Hayashibara Biochemical Laboratories Inc (Okayama, Japan) and poly (vinyl alcohol) with a number-average degree of polymerisation (completely hydrolysed, degree of saponification = 99.9%, Mw 1,25,000) was purchased from Otto Chemical Co., India. Gentamicin was acquired from Sigma Aldrich (J01GB03, Wockhardt UK Ltd). The polymer solutions were prepared using double distilled water. The mouse fibroblast cell line (L929) was obtained from the National Centre for Cell Science (NCCS), Pune and cultured in Eagles Minimum Essential Medium with 10% Fetal Bovine Serum (FBS).

CHARACTERISATION

Solution preparation

Pullulan/PVA was dissolved in D.H_2O and agitated for at least 24 h at 30°C using a magnetic stirrer. The concentration of the solution is 10 wt.%.

Viscosity measurement

A digital viscosimeter (WI-52301, Haryana, India) was used to measure the pullulan/PVA solution for 2 minutes at ambient temperature.

Electrospinning

Pullulan/PVA were combined at a concentration of 10 wt.% each in distilled water by magnetic stirring and heating. Pullulan/PVA solutions were individually prepared and combined. A PVA solution (10 wt.%) was produced by dissolving PVA in distilled water and stirring at 80°C for 2 h. This solution was chosen for production. A pullulan solution (10 wt.%) was produced by dissolving it in distilled water and stirring for 2 h at room temperature. The nanofibers were obtained using a needle electrospinning machine (ESPIN NANO, V1VH, Tamilnadu, India). The polymer syringe was then connected to a voltage of 25 kV. The needle tip was positioned 20 mm away. The solution flow ratio was 0.5 ml/h same procedure was followed to prepare the drug loaded nanofibers. The volume of pullulan/PVA =10ml + 0.5 µl of gentamicin.

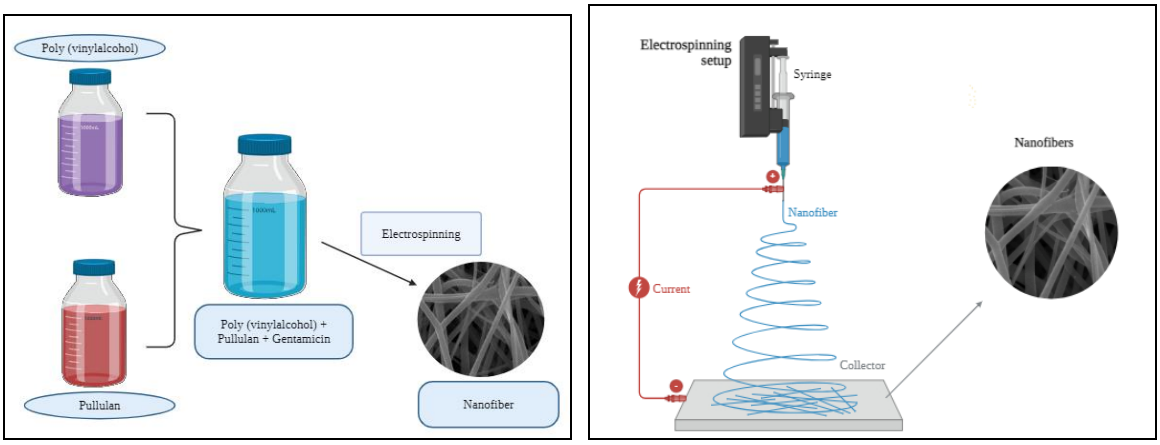

Figure 1: Preparation of polymer in pullulan/PVA solutions.

Fourier Transform Infrared Spectroscopy analysis

FT-IR spectroscopy was utilised to investigate the pullulan/PVA composite membrane (FTIR, Jasco 7084-J002A) in 64 scans with a spectral resolution of 4 cm^{-1} and a scanning range of 4000-600 cm-1.

X-Ray Diffraction method

Intermolecular and intramolecular structure was identified using of hydrogen. Collect the diffraction data using a XRD over a range of angles (2θ) (DOPS2 goniometer).

Surface morphology

A scanning electron microscope (SEM) (Carl Zeiss Microscopy GmbH Germany, EVO 18, 1x to 1000000x, United Kingdom) functioning at high electrospun nanofiber morphology was examined in vacuum. The working distance was 15cm and a 15 kV acceleration voltage was used. The distribution of fiber size and their average diameter were obtained by randomly measuring the diameters using the nano measure ImageJ programme for each sample.

Tensile strength

A material's wear resistance correlates positively with its tensile strength, according to researchers. The tensile strength and modulus values found here were utilized to assess the surface layer's wear resistance. Specimens for the tensile test were specifically designed to fit the tensile tester (30 mm length × 5 mm width). Three materials were tested: freeze-dried pullulan/PVA (~5mm thick), electrospun nanofibrous layer (~1mm thick), and composite (nanofibers on freeze-dried layer, ~6mm thick). In samples with aligned nanofibers, the tensile load was parallel to fiber alignment. Tensile strength analysis was utilised to the characterise nanofiber physical and mechanical properties. The pullulan/PVA nanofibers tensile strength increases as the weight ratio increases. This tensile strength analysis was determined to be approximately 13.9 mPa.s using a pullulan/PVA mass ratio of 10wt.% in the total polymer concentration.

Drug release studies

The drug release functions of the indicated gentamicin-containing nanofibers were examined as follows. 100 cells from each nanofiber were weighed in separate test tubes before being combined with 3 mL of pH 7.2 PBS. The amount of gentamicin released from the nanofiber was evaluated by removing the PBS from the test tubes and replacing it with 3 mL of new buffer after an hour. The same procedure was used after 20, 40, 60, and 80 h. Gentamicin release concentration was measured spectrophotometrically at 255 nm. The standard curve was used to calculate the drug release concentration.

Cell line study of the electrospinning nanofibers

The cells were developed at 36°C in an atmosphere moistened with Co_2 on the fifth day of culture in RPMI 1640 medium (Sigma Aldrich, Czech Republic) supplemented with 10% fetal bovine serum F1283, 100 IU µL-1 streptomycin and 100 IU mL-1 penicillin (Sigma Aldrich, Czech Republic) per millilitre. After cultured for 12 hours, the cell monolayers were taken out of the culture media cleaned with PBS ice and distilled water, then fixed for 10

minutes with a new 1.20 wt.% glutaraldehyde (GA) solution. After removing the fixative fresh GA was applied and the cells were fixed for a further ten minutes. Three PBS and distilled water washing were performed on the cell monolayers. The cells were allowed to dry in the lab at 36°C after the media was removed. Samples were examined using SEM following fixing and drying.

RESULTS

Characterization of the nanofiber

The electrospinning setup's functions such as polymer concentration, collector distance and applied voltage, can all have an impact on the morphology of the nanofibers. During the experiment, the TCD and applied voltage were constant at 20 mm and 25 kV respectively. Pullulan/PVA mixed nanofibers have a pH 5.[11] This pH value is a crucial value due to its influence on the properties and functioning of the nanofibers. Selecting a pH of 5 for the electrospinning solution is important as it facilitates the formation of nanofibers with the correct parameters, stability, and morphology. Maintaining a pH of 5 in the pullulan/PVA solution optimized the electrospinning process and ensured consistent and continuous nanofiber production. The viscosity of the solution had a substantial impact on fiber morphology. Changes in polymer material affect the viscosity of a solution. Thus, the relationship between the solution's viscosity and the pullulan, PVA, and pullulan/PVA 10% was studied. Figure 2 shows the solution's viscosity changed with the pullulan/PVA 10% concentration. The solution's viscosity improved as the concentration of PVA increased. The result may include an increase in the spinning solution's internal friction resistance and a reduction in molecular fluidity.

The polymer pH of the produced nanofibers has a direct impact on their alignment and surface qualities. The findings demonstrate that when H^+ ions in the acid solution produce a high electric current that pulls droplets of the mixed solution to the collector's end a strong electric field is produced. Because of the greater H^+ ion concentration in the acidic solution, there is more electrospinning allowing it to attract chemical compound solution droplets from the collector tip in the direction of the negative electrode after applying a field.[12] Since pullulan has a pH range of neutral to alkaline, a carrier polymer was not used for electrospinning. One important variable is the variety of viscosities of different polymer solutions while electrospinning is carried out. When extracted pullulans were electrospun, their higher viscosity helped them produce continuous, smooth fibers, while lesser

consistency pullulans produced beaded fibers.[13] Pullulan exhibited a viscosity of 40 mPa.s. Because of its film forming and encapsulating qualities, this polysaccharide finds employment in a variety of industries such as the food and pharmaceutical industries. The viscosity of PVA is 6 mPa.s; it is particularly 87-89% hydrolyzed and has a molecular weight range of 1,25,000 g/mol. PVA is a synthetic polymer that has good film-forming, emulsifying and adhesive qualities in addition to being soluble in water.[14] Understanding the flow properties of polymers is essential for understanding their behaviour in diverse applications and processing methods and these values offer valuable insights into this area.

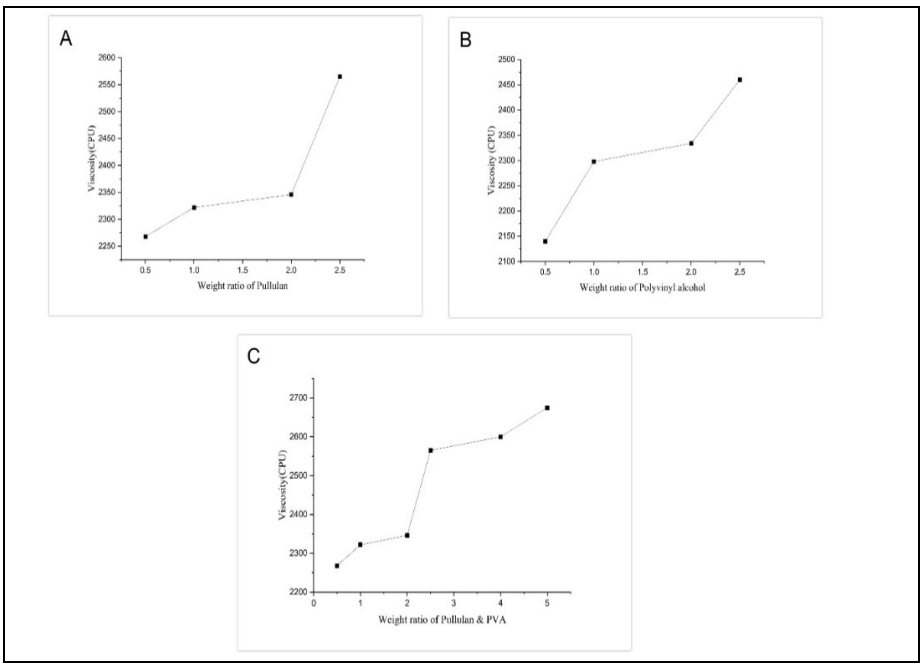

Figure 2: A) Viscosities of pullulan solutions B) Viscosities of the PVA C) Viscosity of the pullulan/PVA polymer solution.

FT-IR Analysis of pullulan/polyvinyl alcohol nanofiber

The vibrational modes and structural interaction of pullulan/polyvinyl alcohol nanofiber were investigated through FT-IR spectroscopy. Pullulan/polyvinyl alcohol nanofiber FT-IR spectra recorded in the wave number range of 4000 to 600cm^{-1}. Figure 3 illustrate the FT-IR spectra of pullulan/PVA nanofibers.[15] Herein, the appeared strong absorption peak at 1091.41cm^{-1}, indicates the strong C-O vibration. Additionally, the absorption peak of 2943.24cm^{-1} corresponds to the stretching vibration of the CH_2 group. The broad stretching vibration of hydroxyl groups (-OH) was suggesting to the wave number range of 3330-3320cm^{-1}. An absorption band at 847.70cm^{-1} in the spectra of pullulan/PVA is indicative of the α-glucopiranosid units. The pullulan produced the strong bond at 3300.82cm^{-1}. The gentamicin

produced the strong bond at 1145.33 cm^{-1}. PVA often produces the following distinctive bands: str (C=O) at 1019.50cm^{-1}, ν(O-H) at 2915.27cm^{-1} and ν(O-H) at 3290.83cm^{-1}. The bands exhibiting CH/CH2 deformation vibrations within the range 1300-1500cm^{-1}. Further, the hydroxyl group (-OH) is attributed to the large adsorption peak at 3290.83cm^{-1}. Table 1 shows the illustration of pullulan/PVA nanofibers in functional group identification.

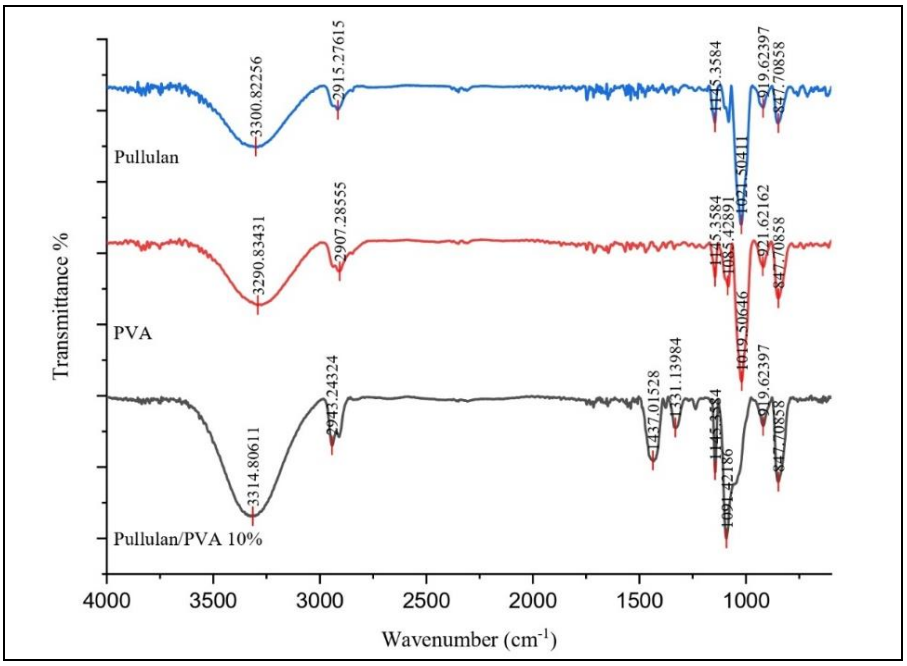

Figure 3: An analysis of the Fourier transforms infrared (FT-IR) spectra of pullulan/PVA nanofibrous mats in the wave number range of 4000-600cm^{-1}.

Table 1: FT-IR values of pullulan/PVA range.

Functional Group	Wave number (cm^{-1})	Material Identification	Reference
-OH	3300	Pullulan	16
C-O	1019	PVA	17
-OH	3330-3320	Pullulan/PVA	18

X-Ray Diffraction method

Figure 4 shows the XRD analysis for the structural differences in the pullulan/PVA nanofibers. To assess the nanofibers, the XRD outlines were compared with those of PVA. It is commonly known that amorphous polymers exhibit broad peaks in X-ray diffraction, whereas a polymer having a crystalline portion exhibit sharper peaks with higher intensities.[19] The pullulan (19.1◦), PVA (19.1) and pullulan/PVA nanofibers is represented by the peak at 2θ = 19.1◦. When mixed with the location and sharpness of the important hybrid peak, proving the consistency of the crystalline structure; nevertheless, FTIR also confirm the chemical bonding of pullulan/PVA nanofibers.[20]

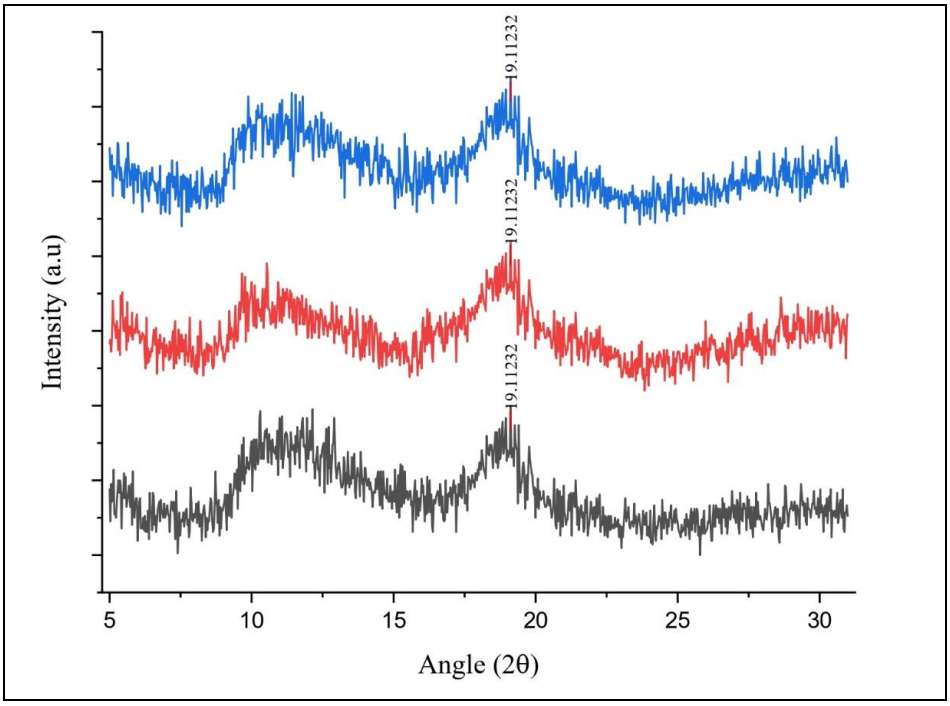

Figure 4: shows the results of an X-ray diffraction of the following materials powdered electrospun pullulan/PVA and nanofibers 10wt.%.

Surface morphology

This carrier polymer cannot break up under normal biological conditions is cost-effective and is stable both chemically and thermally. Figure 5 illustrate the SEM images of A) Pullulan B) PVA C) pullulan/PVA D) pullulan/PVA and gentamicin mixed nanofiber. Using ImageJ software to analyse the SEM images discovered that the PVA-bonded amylopectin nanofibers have a mean diameter between 220 and 230 nm.[21] The electrospun amylopectin and PVA fiber diameters held greater significance than their combinations. As the amount of amylopectin decreased the diameter reduced. The pullulan/PVA nanofiber structures exhibited homogeneity and the absence of beads. Figure 5 (A) shows the pullulan nanofibers at range of 240±41 nm. Which confirm clear appearance without bead. Figure 5 (B) shows the PVA nanofibers at range of 282±40 nm. Which confirm clear appearance without bead Figure 5 (C) shows the pullulan/PVA nanofibers at range of 280±41 nm.[22] Which confirm clear appearance without bead. The drug delivery pullulan/PVA nanofibers Figure 5 (D) confirm the present of pullulan and PVA nanofibers drug with the 290±41 nm. All the SEM images confirm the eventual structures of nanofibers without any beads and clear structure.

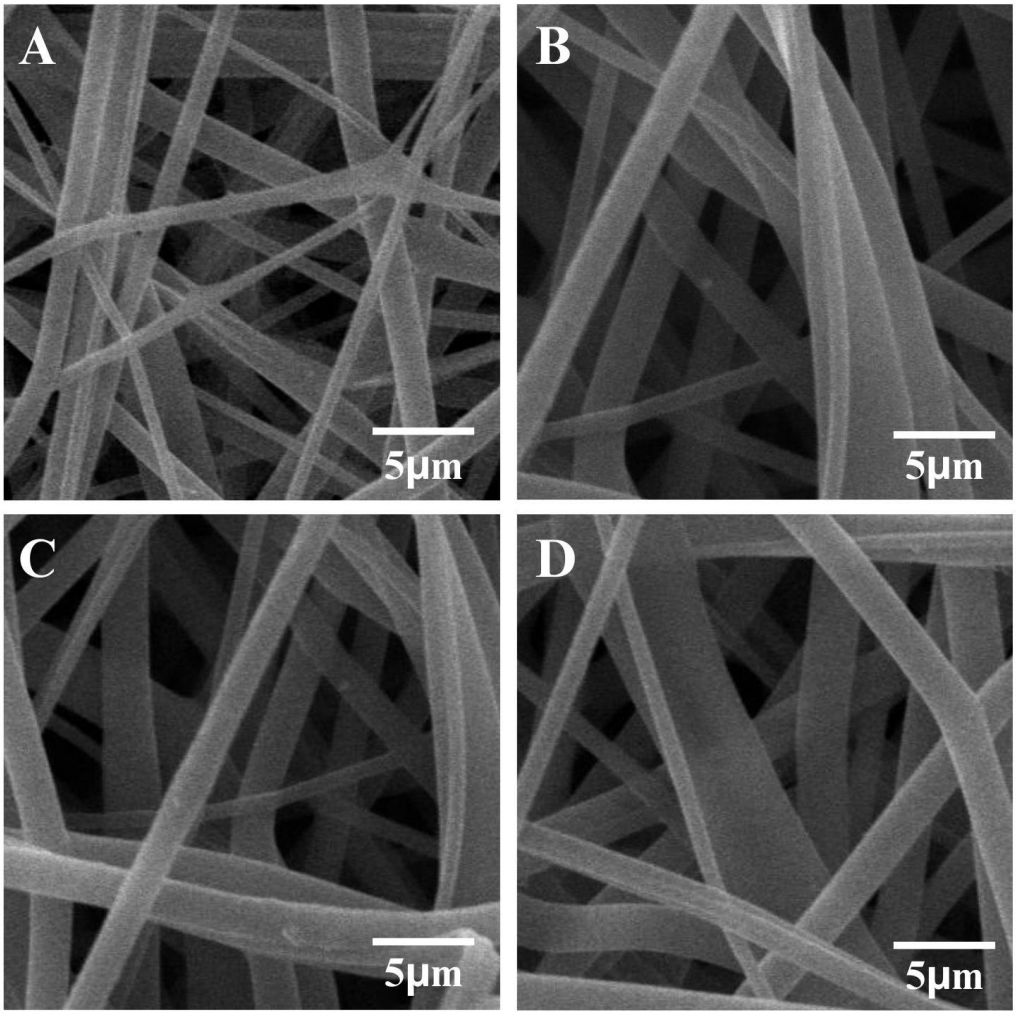

Figure 5: SEM images of Nanofibers A) Pullulan B) PVA C) Pullulan/PVA D) Pullulan/PVA with Gentamicin.

Tensile Strength Analysis

Tensile strength analysis (Figure 6) shows that the electrospun pullulan/PVA nanofibers as a function of PVA %. Tensile strength increases proportional with PVA concentration.[23] Pullulan nanofibers had a maximum tensile strength of 5.5mPa, PVA nanofibers had a maximum tensile strength of 8.0mPa, while pullulan/PVA nanofibers with a 10wt.% produced a strength of 13.5 mPa. This finding suggests that PVA inclusion enhances the mechanical performance of pullulan/PVA nanofibers.[24] Pullulan/PVA interfacial adhesion improves with PVA content, resulting in increased tensile strength.

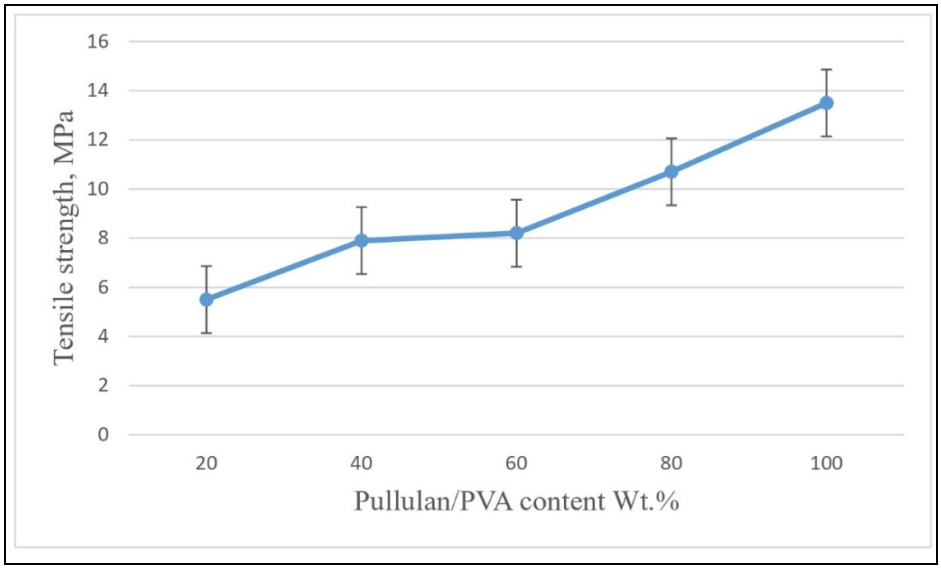

Figure 6: Tensile strength studies on electrospun pullulan/PVA nanofibers (applied voltage = 15 kV, TCD = 15 cm, total polymer concentration of 10 wt.%)

Differential scanning calorimetry (DSC) analysis

DSC thermogram of electrospun pullulan/PVA nanofibers at 10wt%. Figure 7 shows a significant endothermic peak at around 225ºC, indicating the melting of PVA. The DSC thermogram of pullulan showed an important peak for the melting transition at 95℃. This finding is quite consistent with the supplied DSC data.[25] Adding 10 wt.% pullulan/ PVA produced changes of the peak in 236℃, 246℃ and 225ºC. The continuous shift of these endothermic peaks to lower temperatures as pullulan content increases indicates a decrease in the melting temperature of the nanofibers. The lower melting temperature was likely caused by non-crystalline chains formed during electrospinning, which solidified fast.[26] The explanation above comes to the conclusion that in order to produce pullulan/PVA mix nanofibers with improved thermal stability, the PVA mass proportion has to be higher than the pullulan mass proportion.

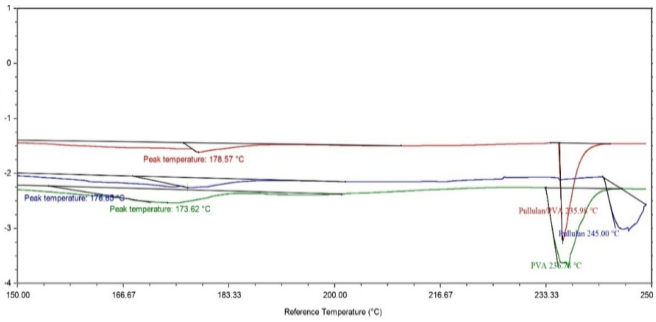

Figure 7: DSC data of electrospun pullulan/PVA nanofibers at 10wt.% ratios (total polymer concentration = 10%, applied voltage = 15 kV and TCD = 15 cm).

Gentamicin standardization

The concentration of gentamicin in each vial was 10 mg/0.2 mL. To be able to prepare the stock, 1 mL was separated from the vial and dilute to 5 mL with distilled water.[27] Various aliquots of 0.5, 1, 1.5 and 2.0 mL were then taken and diluted to half the concentration, with measurements at 255 nm. The range of the O.D. value is 100–400µg.

***In vitro* drug release**

The drug release pattern for gentamicin from pullulan/PVA nanofibers at 20, 40, 60, and 80 hrs with different PVA concentrations is shown in Figure 8. The high PVA content raised the medication release rate in both situations. Furthermore, both formulations drug release patterns exhibit a burst release at initially, followed by a continuous release, which is explained by drug diffusion through the polymer wall and polymer degradation within the nanofiber.[28] The nanofiber with the drug releases much more medication than the nanofiber with gentamicin. For example, while the gentamicin itself showed 70-700 µg, the drug release from the nanofiber containing gentamicin was 8,500-20,000 µg.

Due to drug diffusion in the polymeric nanofiber, gentamicin was released in a burst from all of the nanofibers that contained it, followed by a continuous release from nanofibers P1, P2 and P3.[29] This demonstrates the drug's continuous release over the course of the 80h investigation. A significantly higher concentration of gentamicin is released by the nanofiber. A stronger interaction between the drug and the polymer than gentamicin is the reason for the slower rate of release of gentamicin. The release of nanofibers also proceeded initially.

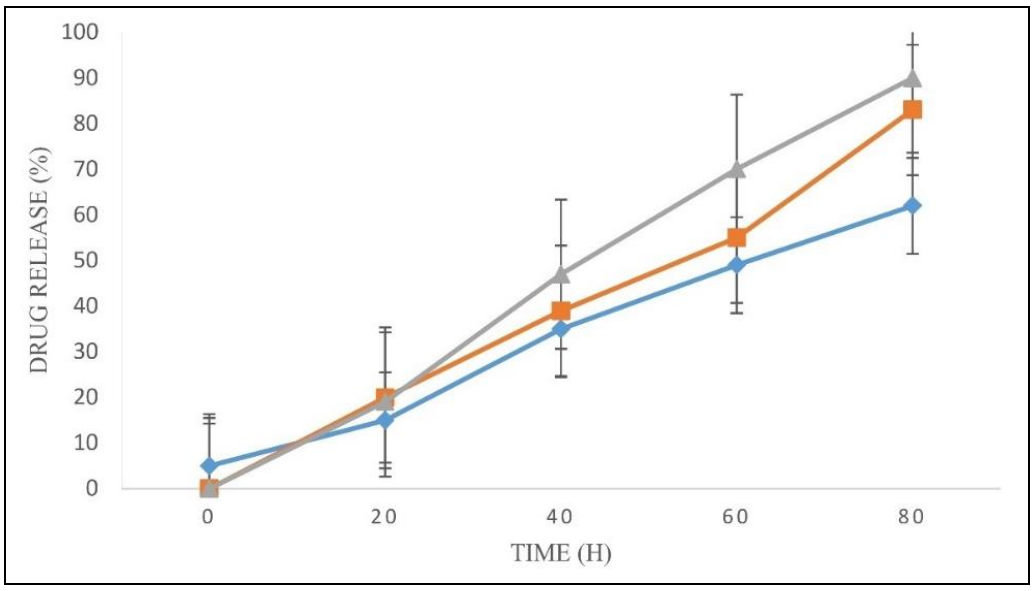

Figure 8: Pullulan/PVA and gentamicin nanofibers with concentration of 10wt.%

The release profile for PVA demonstrates controlled exponential behaviour, where the percent release slowly increases over time and plateaus. The release from pullulan is linear but at a controlled rate indicating a steady and slow release of the drug. This is advantageous for therapeutic applications that require stable drug concentrations over time. The composite material exhibited a controlled sigmoidal release profile. The slow initial release followed by a more rapid but controlled release phase and a final plateau suggest a multistage release mechanism.[30] The composite material released its contents more quickly and thoroughly than either pullulan/PVA alone based on the release profiles of pullulan/PVA and the pullulan/PVA/gentamicin composite during a 48h period (figure 8). A more effective and targeted drug delivery system that maximises the therapeutic impact while minimising potential side effects in wound dressing treatment is provided by the composite material made of pullulan/PVA and gentamicin.[31] This material was chosen for its enhanced release profile because it combines the mechanical stability and biocompatibility of PVA, the biodegradability and encapsulation efficiency of pullulan and the therapeutic action of gentamicin. The result is a controlled and gradual release of the wound healing drug.

Cell Line Studies

Fibroblast L929 cells were shown to retain their normal morphology for a duration of one day when exposed to pullulan/PVA nanofibrous drug-loaded nanofibers, which were formed at a concentration of 10wt.% (Figure 9A). This is due to the fiber's three-dimensional structure, which increases the surface area accessible for conduction. Four days after planting, some cell states were altered (Figure 9D). It's spindle shape gave way to a circular one, signifying the end of apoptosis. Release of gentamicin from the fibers as the polymer breaks down, killing of organism.[32] Cell morphology could not be detected after 9 days after extraction (Figure 9F), which indicates that the medication had killed the fibroblast cells. Table 3 illustrate the concentration of different day's cell growth. So, the wound treatment frequently yields favourable results. The fiber structure improves sustained drug release, which gives an advantage for various treatments in medical science.[33] After three days of growth, the fibers produced significantly declined in a physiological context, which is promising for tissue engineering applications.

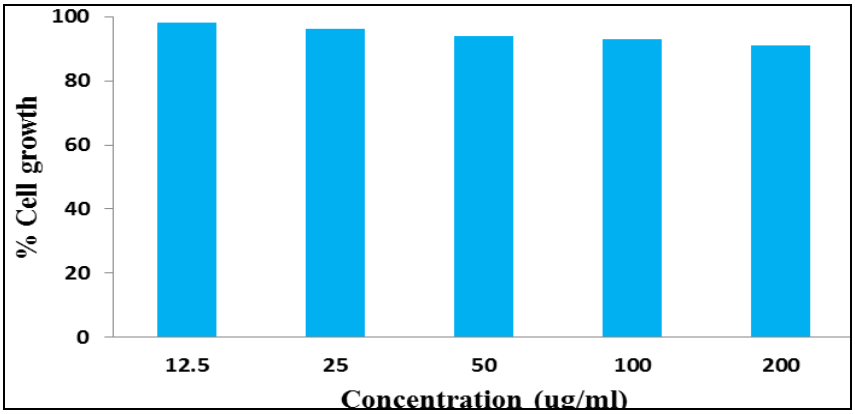

Table 2: Concentration of the cell growth.

Conc	12.5 µg	25 µg	50 µg	100 µg	200 µg	Cont
ABS	0.339	0.331	0.324	0.319	0.315	0.345
	0.337	0.332	0.323	0.321	0.313	0.347
	0.338	0.333	0.324	0.321	0.313	0.343
Avg	**0.338**	**0.332**	**0.323667**	**0.320333**	**0.313667**	**0.345**

Table 3: Concentration of the cell growth.

Conc (µg/ml)	% Cell Growth
Control	100
1 day	97.97101
3 day	96.23188
5 day	93.81643
7 day	92.85024
9 day	90.91787

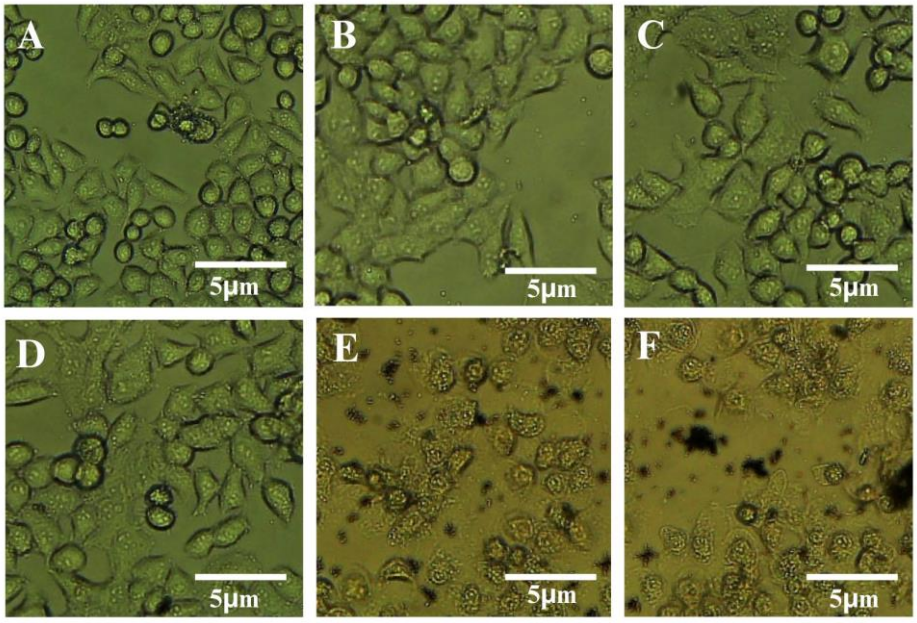

Figure 9: Pullulan/PVA and gentamicin nanofibers with concentration of 10wt.% seen in SEM images of fibroblast L929 Cells.

DISCUSSION

A study on pullulan/PVA and pullulan as carrier polymers for drug delivery more especially for the wound-healing medication gentamicin is presented in this article. The drug-loaded nanofibers that were produced by electrospinning with pullulan/PVA nanofibers were successful in preventing the adhesion and growth of fibroblast L929 cells. Pullulan/PVA was selected as the carrier polymers because of their relative affordability, chemical and thermal stability and resistance to degradation in the majority of physiological conditions. Additionally, Pullulan's homogeneous fiber structure achieved by combining with PVA and its beaded structure on the nano-surface obtained by electrospinning alone make them excellent for drug delivery. Gentamicin has a well-established wound healing impact, but its cardio toxicity prevents it from being used for extended periods. This restriction is removed by encapsulation in drug delivery methods, which lessen gentamicin harmful effects on healthy cells. The results of this investigation indicate that the generated nanofibers have potential as secure and eco-friendly medication delivery systems against solid tumours and wound dressings. The absence of any chemical interactions between pullulan/PVA was also revealed by the FT-IR study results, which is crucial for preserving the stability of the carrier polymer. Overall, this work offers insightful information about the effectiveness of electrospinning in producing drug-loaded nanofibers for wound dressing medications as well as the usage of pullulan/PVA and gentamicin as carrier polymers for drug delivery. To improve the drug-release characteristics of these nanofibers and enhance therapeutic outcomes, more research is required to determine the safety and effectiveness of these materials *in vivo*.

CONCLUSION

This study investigated the possibility of encapsulating gentamicin, a popular medication for wound healing, utilising pullulan/PVA as carrier polymers for drug delivery systems. The study effectively created homogeneous fibers using electrospinning, which prevented any chemical reactions between pullulan/PVA. By using this technique, the gentamicin solution was able to be encapsulated within the nanofibers, lessening the negative effects of the medication on healthy cells. The created nanofibers demonstrated their potential as a secure and focused drug delivery system for treating organisms by successfully blocking the adhesion and proliferation of fibroblast L929 cells, a kind of wound healing. The mean diameter of the PVA-bonded amylopectin nanofibers, which varied from 220 to 230 nm and the estimated fiber diameters, which were roughly 250 nm, was among the quantitative data

from this work. The nano-surface of pullulan produced by electrospinning alone has a beaded structure, whereas the nanofiber structure produced by combining with PVA is more uniform. There were no chemical interactions between pullulan/PVA, according to FT-IR research. Gentamicin is a popular antibiotic with the ability to heal wounds. To get beyond this restriction, encapsulation in drug delivery methods works better than drug-independent tissues. A pullulan/PVA was employed to create nanofibers using electrospinning. It is a great polymer carrier since it is affordable, chemically and thermally stable. A wide range of people utilise pullulan, which makes drug management easier to acquire. In normal cells, encapsulation can lessen the negative effects of gentamicin. Medication nanostructures could be to be effective and sustainable medication delivery methods for the management of acute illnesses. Fibroblast cell adhesion and proliferation were effectively inhibited by the drug-loaded nanofibers. In summary, the resulting nanofibers are viable, safe and eco-friendly drug delivery systems against living things.

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