TEKSTİL VE MÜHENDİS

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ELECTROSPINNING OF PVP/CARVACROL/LANOLIN COMPOSITE NANOFIBERS

PVP/LANOLİN/KARVAKROL KOMPOZİT NANOLİFLERİN ELEKTRO LİF ÇEKİMİ

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Online Erişime Açıldığı Tarih (Available online):31 Aralık 2023 (31 December 2023)

Bu makaleye atıf yapmak için (To cite this article):

Hülya KESİCİ GÜLER, Mustafa GEYSOĞLU, Funda CENGİZ ÇALLIOĞLU (2023): Electrospinning Of Pvp/Carvacrol/Lanolin Composite Nanofibers, Tekstil ve Mühendis, 30: 132, 302- 309.

For online version of the article: https://doi.org/10.7216/teksmuh.1406047

TMMOB Tekstil Mühendisleri Odası
UCTEA Chamber of Textile Engineers
Tekstil ve Mühendis
Journal of Textiles and Engineer

Yıl (Year) : 2023/4
Cilt (Vol) : 30
Sayı (No) : 132

Araştırma Makalesi / Research Article

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Gönderilme Tarihi / Received: 01.08.2023
Kabul Tarihi / Accepted: 15.12.2023

ABSTRACT: In this study, it was aimed to produce and characterize various concentrations of carvacrol and lanolin loaded PVP nanofibers via electrospinning. Various concentrations of carvacrol: lanolin were added to the PVP polymer solutions such as 2.5, 5, 7.5, 10, 12.5 and 15 wt %. Solution properties such as viscosity, conductivity, and surface tension were measured. In terms of characterization studies, SEM, FT-IR, TGA and DSC analysis were carried out. According to the results of the study, viscosity increased and conductivity decreased with carvacrol: lanolin concentration increasement. However, surface tension was not affected from carvacrol: lanolin concentration. Nanowebs quality improved with carvacrol: lanolin concentrations. Generally, nanofibers are quite fine, smooth, and uniform. FT-IR spectrums verified that PVP, carvacrol and lanolin exist in the structure of nanofibers chemically. DSC and TGA results demonstrated that the thermal stability of carvacrol and lanolin, which have poor thermal stability, was enhanced by incorporating them into the PVP nanofiber structures. Considering antibacterial properties of carvacrol and wound healing properties of lanolin, these composite nanofiber surfaces have a high potential for use in the biomedical field, especially as a wound dressing.

Keywords: Polyvinylpyrrolidone, carvacrol, lanolin, electrospinning, nanofiber.

PVP/LANOLİN/KARVAKROL KOMPOZİT NANOLİFLERİN ELEKTRO LİF ÇEKİMİ

ÖZ: Bu çalışmada, elektro lif çekim yöntemi kullanılaarak çeşitli konsantrasyonlarda karvakrol ve lanolin yüklü PVP nanoliflerin üretilmesi ve karakterize edilmesi amaçlanmıştır. PVP polimer çözeltilerine ağırlıkça %2.5, 5, 7.5, 10, 12.5 ve 15 gibi çeşitli konsantrasyonlarda karvakrol:lanolin eklenmiştir. Hazırlanan çözeltilerin viskozite, iletkenlik ve yüzey gerilimi gibi çözelti özellikleri ölçülmüştür. Karakterizasyon çalışmaları açısından SEM, FT-IR, TGA ve DSC analizleri gerçekleştirilmiştir. Çalışmanın sonuçlarına göre, çözeltilerdeki karvakrol:lanolin konsantrasyonu arttıkça viskozite artmış, iletkenlik ise azalmıştır. Ancak, yüzey gerilimi karvakrol:lanolin konsantrasyonundan etkilenmemiştir. Nano ağların kalitesi, karvakrol:lanolin konsantrasyonu ile artmıştır. Genel olarak, oldukça ince, pürüzsüz ve üniform yapıda nanolifler elde edilmiştir. FT-IR spektrumları PVP, karvakrol ve lanolinin kimyasal olarak nanoliflerin yapısında bulunduğunu doğrulamıştır. DSC ve TGA sonuçları, zayıf termal stabiliteye sahip olan karvakrol ve lanolinin termal stabilitesinin, PVP nanolif yapılarına ilave edilmesiyle artırıldığını göstermiştir. Karvakrolün antibakteriyel özellikleri ve lanolinin yara iyileştirici özellikleri göz önünde bulundurulduğunda, bu kompozit nanolifli yüzeyler biyomedikal alanda, özellikle de yara örtüsü olarak kullanım için yüksek bir potansiyele sahiptir.

Anahtar Kelimeler: Polivinilpirolidon, karvakrol, lanolin, elektro lif çekim, nanolif.

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***DOI:** https://doi.org/10.7216/teksmuh.1406047 www.tekstilvemuhendis.org.tr*

This study was presented at "8th International Technical Textile Congress (ITTC2022)", October 13-14, 2022, Online Congress. Peer review procedure of the Journal was also carried out for the selected papers before publication.

1. INTRODUCTION

Nanofibers have superior properties such as small fiber diameter (nm), small and open pores, high porosity, large specific surface area (m^2/g), and high loading capacity. With this high loading properties of nanofibers, different kinds of agents such as drugs, proteins, antibacterial agents, enzymes, and essential oils can be loaded nanofibers in terms of get much more functionally according to their end use application areas [1, 2]. In this study, carvacrol and lanolin were loaded to the biocompatible PVP nanofibers for their antibacterial and wound healing properties.

The main component of *Origanum minutiflorum* (O Schwarz and PH Davis), an endemic thyme species of the Lamiaceae family growing in Isparta/Sütçüler region, is carvacrol [3, 4]. Carvacrol is natural, inexpensive and easy to obtain. Both carvacrol and thyme essential oils have antibacterial, antifungal, odor, and pesticidal properties [5, 6]. In the literature, there are studies in which carvacrol is incorporated in the structure of nanofibers and nanofibers gain antibacterial properties. By using Zein/poly (lactic acid) [7], chitosan/poly(ethylene oxide) [8, 9], gelatin/ poly(vinyl pyrrolidone) (PVP) [10] polymers, and combinations, thyme essential oils loaded antibacterial nanofibers have been produced for different application areas such as food packaging and biomedical. Lanolin, an organic ester derived from sheep fleece after shearing, produces an air-permeable temporary barrier and promotes moist wound healing when applied to damaged skin. Actually, lanolin is a kind of wax formed which is also known as wool yolk, wool wax, or wool grease. It softens and treats dry skin on the lips and nipples. Generally, new mothers, breastfeeding mothers, chemotherapy patients with dry, cracked, and damaged skin, and those with burn wounds use products containing lanolin. Lanolin is commercially available in many medical such as ophthalmic eye drops, burn and wound healing creams, and cosmetic products such as lotions, makeup, sunscreen and shaving creams or gels [11, 12]. Studies on lanolin are quite new and limited in the literature. Akduman et al. [13] developed lanolin loaded cellulose acetate (CA), polyethylene oxide (PEO), polyethylene oxide/chitosan (PEO/chitosan) and thermoplastic polyurethane (TPU) based nanofibers which is used for nursing pads. The research group determined that lanolin loaded nanofibers can be used for nursing pads and CA nanofibrous surfaces could be preferred when better hydrophilicity and swelling are required. Another research group produced and characterized silk fibroin(SF)/poly (caprolactone) (PCL) blend nanofibers with various concentrations of lanolin. They indicated that smooth and fine nanofibers produced for all samples. However, as lanolin increased, the viability rate in the cell decreased. Because of this reason it is a solvent whose name is 1,1,1,3,3,3-hexafluoropropane-2-propanol(HFIP). They suggested that more suitable solvent should be used in terms of cell viability and antibacterial activity [14].

Poly(vinylpyrrolidone) (PVP) is not toxic polymer and environmental friendly [15], synthetic, hydrophilic [16], water-soluble [17], and biocompatible polymer [18]. PVP is chosen as the raw material for this study because of all of these features, which are important in biomedical and cosmetic applications [10, 19]. PVP polymer is widely used in the pharmaceutical industry [20] and biomedical applications [21, 22], such as medicine [23], drug delivery systems [24], wound dressing, tissue engineering [25], and the bioactive packaging industry [26].

Investigations on Carvacrol and Anhydrous Lanolin nanofibers have been published in the literature individually [7, 12, 27]. However, no study has been conducted on PVP/Carvacrol/ Lanolin nanofiber composites in combination. Therefore, the findings of this study are expected to be valuable in the literature.

2. MATERIALS AND METHODS

2.1 Materials

To produce nanofibers, PVP (360.000 g/mol) (Sigma–Aldrich (St. Louis, MO, USA)) was used as a polymer, Carvacrol (Süleyman Demirel University, Natural Products Application and Research Center, with 96 % purity) and Anhydrous Lanolin (Galenic Chemicals, İzmir, Turkey) were used as an additive, deionized water was used as a solvent and Cremophor RH 40 (Ersa Chemistry, İzmir, Turkey) was used as a surfactant.

2.2 Preparation and Characterization of Polymer Solutions

Various concentrations of carvacrol:lanolin added to the PVP polymer solutions (Table 1). According to our preliminary studies, optimum carvacrol:lanolin ratio was determined as 9:1. Then, solution properties such as viscosity, conductivity and surface tension were measured.

2.3 Production and Characterization of Nanofibers

Nanofiber productions were carried out by electrospinning set up under the optimum process parameters (Table 2).

After nanofibers production, characterization studies were achieved. In order to analyze nanofibers morphology, SEM images were taken with different magnifications. ImageJ software was used to measure the diameters of 100 fibers that were taken from different parts of the electrospun nanowebs. and in order to understand fiber diameter distribution, fiber diameter histograms curves were obtained via statistical analysis program. Then, average fiber diameter uniformity coefficient values were calculated using the formulas are given in below:

$$A_n = \frac{\sum n_i d_i}{\sum n_i} \ (number\ average) \quad (1)$$

$$A_w = \frac{\sum n_i d_i^2}{\sum n_i d_i} \ (weight\ average) \quad (2)$$

d_i: fiber diameter *ni: fiber number*

Table 1. Sample codes and composition of carvacrol:lanolin loaded PVP polymer solutions

Sample Codes	PVP Concentration (%)	Carvacrol:Lanolin Ratio	Carvacrol:Lanolin Concentration (%)
PVP0	12	9:1	0
PVP2.5	12	9:1	2.5
PVP5	12	9:1	5
PVP7.5	12	9:1	7.5
PVP10	12	9:1	10
PVP12.5	12	9:1	12.5
PVP15	12	9:1	15

Table 2. Process parameters of electrospinning

Voltage (kV)	Distance between electrodes (cm)	Feed Rate (mL/h)	Humidity (%)	Temperature (°C)	Needle Diameter (mm)
17	24.0	0.2	30-35	22-24	0.8

The ratio of A_w/A_n was used to calculate the fiber diameter uniformity coefficient. The closer the average fiber diameter uniformity coefficient value is to 1, the more uniform fibers can obtain [28]. FT-IR spectrums were carried out to understand chemical structure of PVP based nanofibers with Attenuated Total Reflection (ATR) technique between 4000 and 400 $cm^{-1.}$ Thermogravimetric analysis (TGA) was carried out with an Exstar SII TG DTA 7200 to analyze the thermal stability of PVP based nanofibers, polymer form of PVP, Carvacrol, and Anhydrous Lanolin in a nitrogen gas environment by increasing 10°C/min from room temperature to 600 °C. Differentiating Scanning Calorimetry (DSC) analysis was performed using a Perkin Elmer DSC 4000 in order to determine the glass transition, melting and decomposition temperatures, and enthalpies of PVP, lanolin, carvacrol, and nanofibers in a nitrogen gas environment from 24 °C to 600 °C, with a 10 °C/min temperature increase.

3. RESULTS AND DISCUSSION

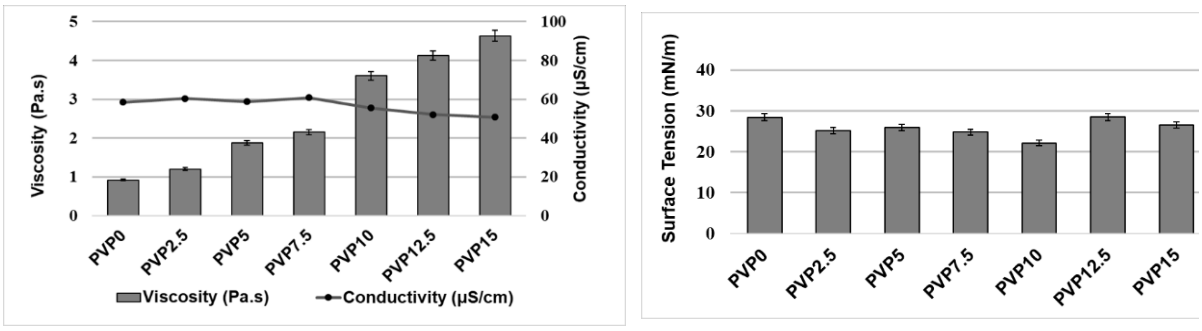
3.1 Solution Properties Results

The properties of polymer solutions, such as viscosity, electrical conductivity, and surface tension, all have an impact on the morphology of electrospun nanofibers. Therefore, within the scope of the study, the polymer solution properties were measured and the results are given in Figure 1.

According to Figure 1; viscosity increases and conductivity decreases while carvacrol:lanolin concentration increases. However, surface tension was not affected from carvacrol:lanolin concentration.

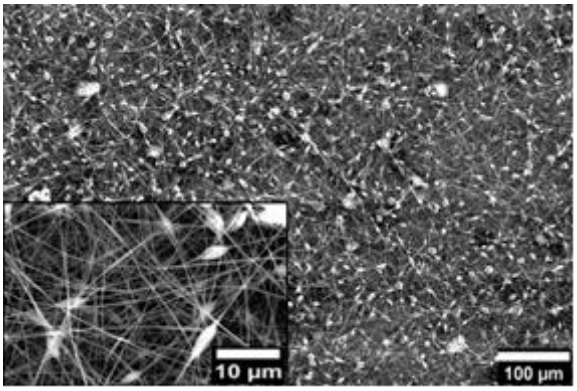
3.2 Fiber Morphology Results

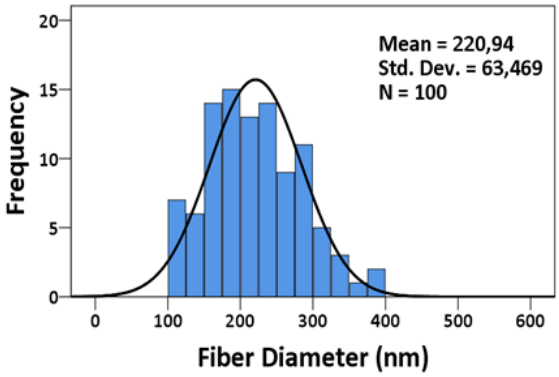
SEM images of various concentrations of carvacrol:lanolin nanofibers are given in Figure 2.



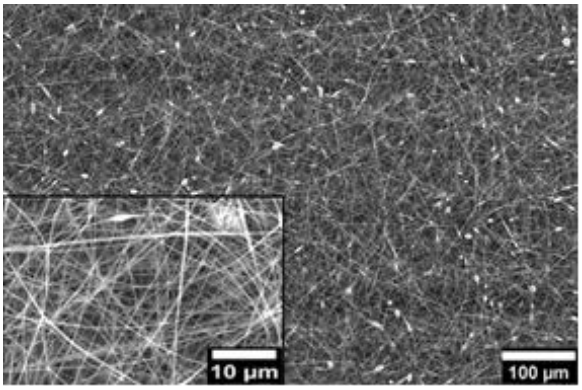
(a) (b)

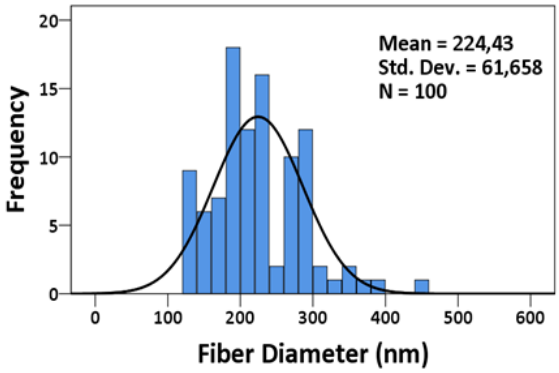
Figure 1. Solution properties results (a) viscosity and conductivity (b) surface tension



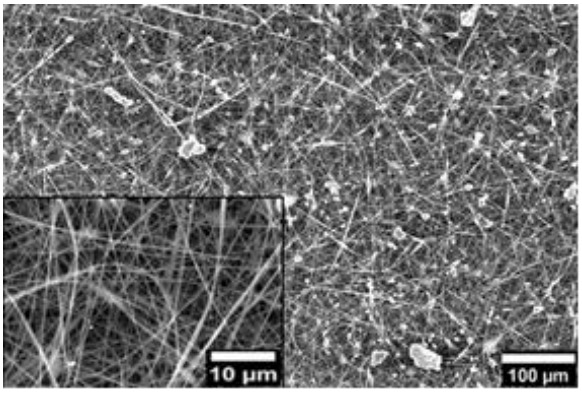


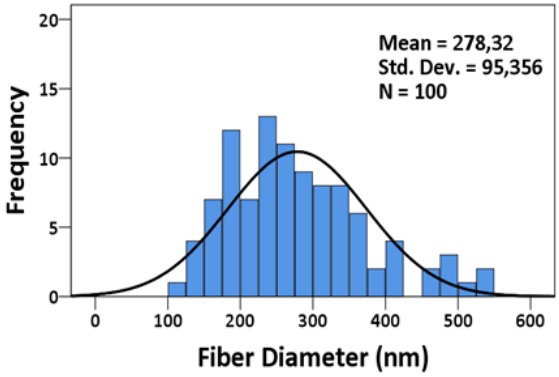
PVP0



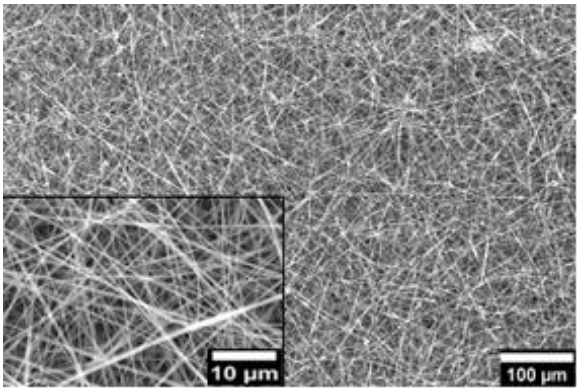


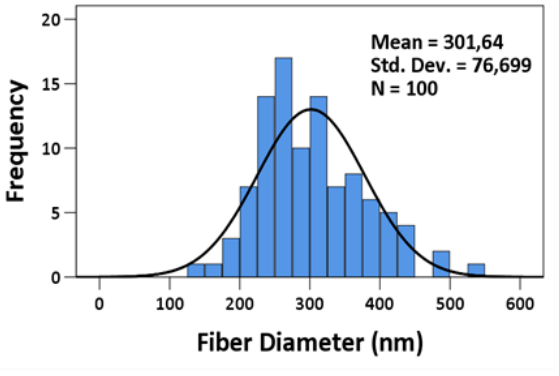
PVP2.5





PVP5





PVP7.5

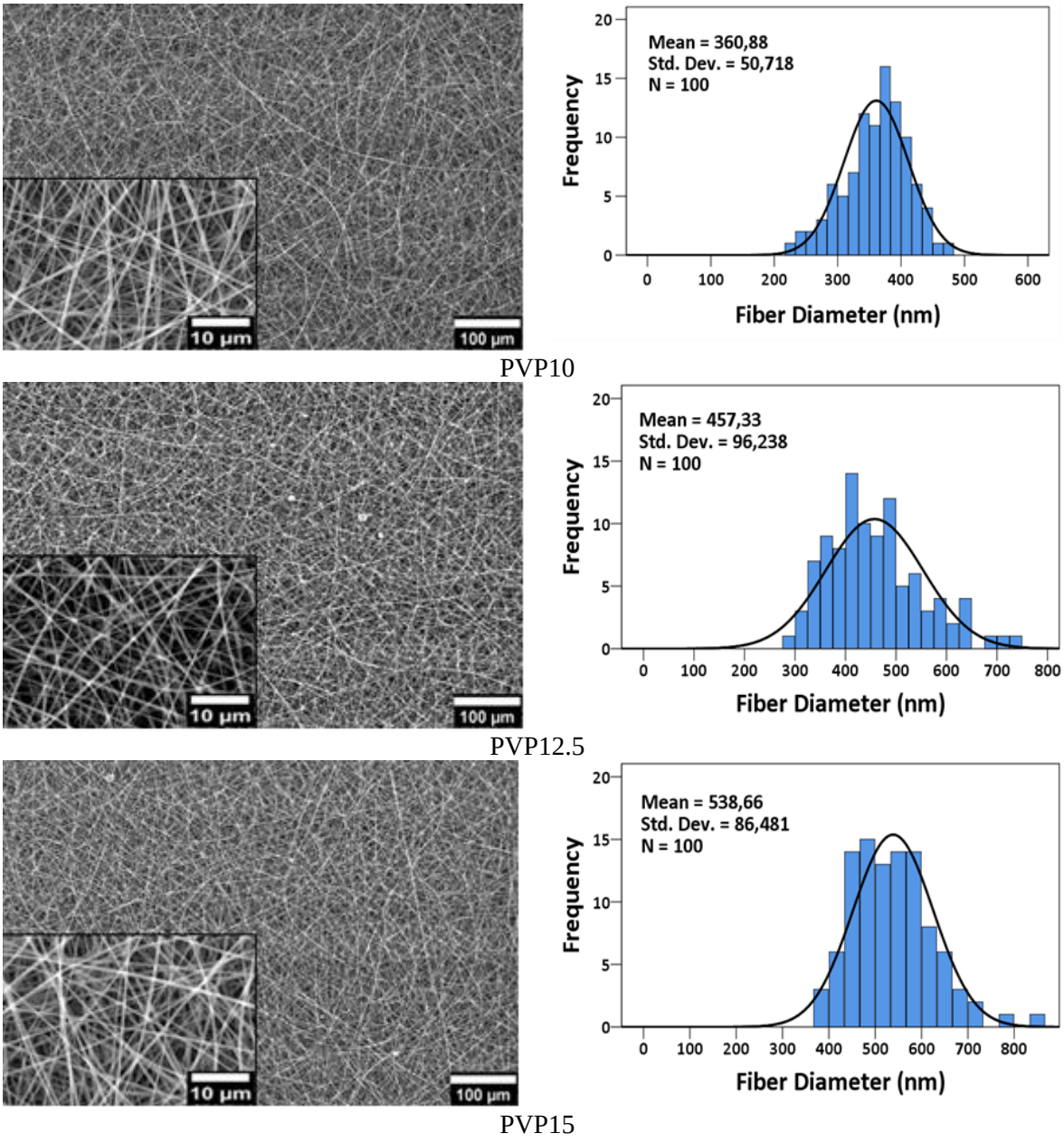


Figure 2. SEM images of PVP nanofibers with various concentrations of carvacrol:lanolin

It has been seen clearly that nanofiber morphology improved with carvacrol:lanolin concentrations. There are some bead defects in the nanoweb structure for concentrations of 0, 2.5, and 5 wt. % of carvacrol: lanolin. However, after these concentrations, nanoweb quality changed significantly. It is possible to say that nanofibers are quite fine, smooth and uniform for 7.5, 10, 12.5 and 15 wt % carvacrol:lanolin concentrations. Due to the fact that the interaction between macromolecules increased as solution viscosity increased, fiber spinning performance increased while beads decreased. Viscosity is known to influence the interaction between macromolecules [29]. In electrospinning, it is well known that higher viscosity and lower conductivity result in less stretching of the jet, therefore producing thicker nanofibers [1].

When the histograms are analyzed, it can be said that all samples have a single peaked and unimodal distribution. Average fiber diameter and fiber diameter uniformity coefficient graph is given in Figure 3.

According to Figure 3, it was determined that average fiber diameter increased and fiber diameter uniformity coefficient did not affect carvacrol:lanolin concentration. According to the viscosity results; it is expected that average fiber diameter increased from 220 (PVP0) to 538 (PVP15) nm with lanolin carvacrol concentrations. With 1.02, the most uniform nanofibers were obtained in the PVP10 sample. However, it is possible to say that the bead-free samples such as PVP7.5, PVP10, PVP12.5, and PVP15 are all uniform. As a nanofiber morphology results; it is

possible to say that PVP10 were selected as the optimum sample in order to average fiber diameter, fiber diameter distribution, fiber uniformity and fiber morphology.

FT-IR spectrums of PVP, lanolin (LAN), carvacrol (CAR), and PVP10 nanofibers are given in Figure 4.

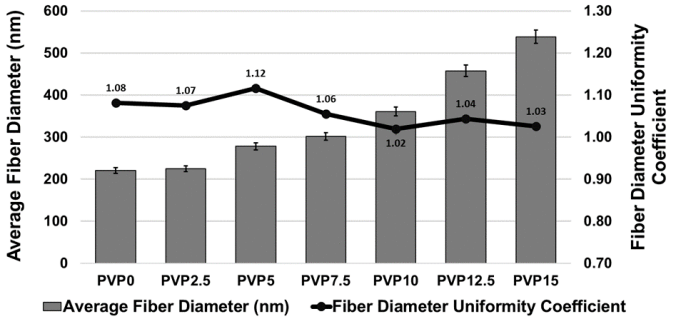


Figure 3. Average fiber diameter and fiber diameter uniformity coefficient of PVP nanofibers produced with various concentrations of carvacrol:lanolin

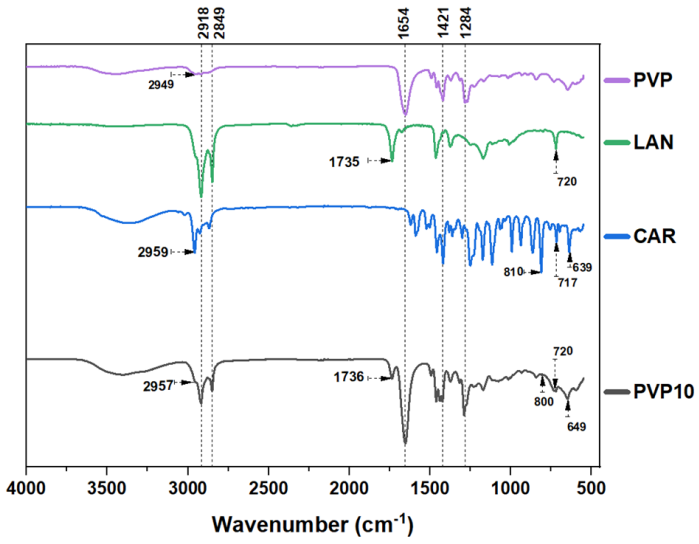


Figure 4. FT-IR spectra of PVP, LAN, CAR, and PVP10 nanofibers

FT-IR spectrums demonstrated that all characteristic peaks of PVP, lanolin and carvacrol arise in the spectra of the PVP10 nanofibers. This means, there were not undesirable reactions while preparing polymer solutions. In detail, there is a sharp peak at 3434 cm^{-1} in the spectra of PVP. The O-H peak can be attributed to the presence of water. The peak is determined at 3403 cm^{-1} in the PVP10 spectra with an increase of intensity. Because, at this wavelength, carvacrol also has a wide peak. Since these two peaks overlapped in the PVP10 sample, the intensity of the peak increased. Another strong peak at 1654 cm^{-1} identified the existence of heteroatomic molecules and carbonyl groups in the pyrrolidone ring of PVP as a sign of C=O stretching. The peak arises at the same wavelength in the spectra of PVP10 [26, 30, 31]. The most intense peak in the spectrum of carvacrol occurred at 811 cm^{-1} (C–H wagging vibrations). This peak also appears at 800 cm^{-1} in the spectra of PVP10. Additional peaks in the carvacrol spectrum at 639 cm^{-1} (C=C) and this peak also arise 649 cm^{-1} in the spectra of PVP10 [32, 33]. There are two characteristic absorption peaks at 2918 cm^{-1} and 2849 cm^{-1} could be attributed to $–CH_2–$ and $–CH_3$. Another characteristic two peaks are at 1735 cm^{-1} and 720 cm^{-1} namely cis-CH=CH and carbonyl compounds [34, 35]. All of these characteristic peaks also appeared in spectra of PVP10 nanofibers.

TGA thermograms of PVP based nanofibers, PVP polymer, Carvacrol, and Lanolin are given in Figure 5.

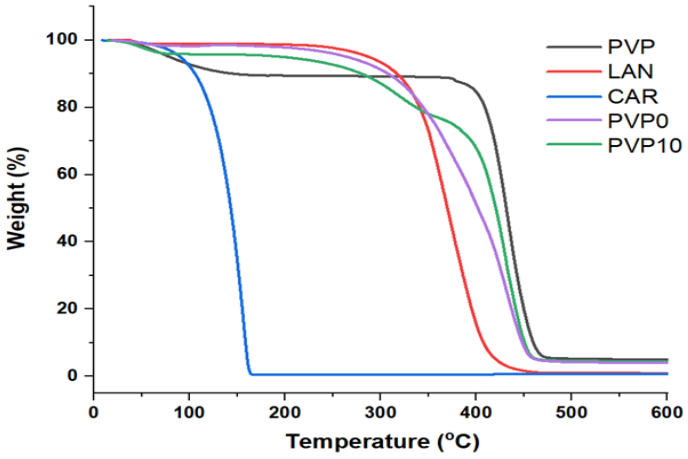


Figure 5. FT-IR spectra of PVP, LAN, CAR, and PVP based nanofibers

Carvacrol, known to be a volatile constituent, degraded between 44.43 and 164.52 degrees Celsius and left no residue, as expected. Anhydrous Lanolin, which is highly hydrophobic, undergoes almost no mass reduction up to 100 degrees Celsius. Lanolin, which started degradation at 231.72 degrees Celsius, completed degradation at 464.10 degrees Celsius and left a very small residue of 1.25%. PVP in polymer form lost 11.52% mass up to 100°C due to its hydrophilic structure, degraded between 354.6°C and 478.1°C and left 5.72% residue after degradation. When the thermograms of the nanofibers were examined, it was determined that the PVP0 coded nanofibers showed a thermal behavior similar to the PVP polymer in polymer form but degraded at lower temperatures (212-465 degrees) due to the rapid mass transfer due to the nanoscale fiber structure. The thermogram of carvacrol/lanolin-loaded and PVP-based nanofibers (PVP10) shows a two-step degradation. This is thought to be due to the fact that the starting temperatures of lanolin and PVP polymers are quite different from each other. While the first step degradation due to lanolin occurred between 204.74 and 377.43 degrees, the second step degradation due to PVP polymer occurred between 385.87 and 464.44 degrees. Both PVP0 and PVP10 coded nanofiber samples left 4.80% and 5.06% residue, respectively, similar to PVP in polymer form. In general, carvacrol and lanolin, which have very poor thermal stability, were incorporated into the structure of PVP nanofibers to increase their thermal stability.

DSC analysis of PVP polymer, Lanolin and PVP based nanofibers are shown in Figure 6.

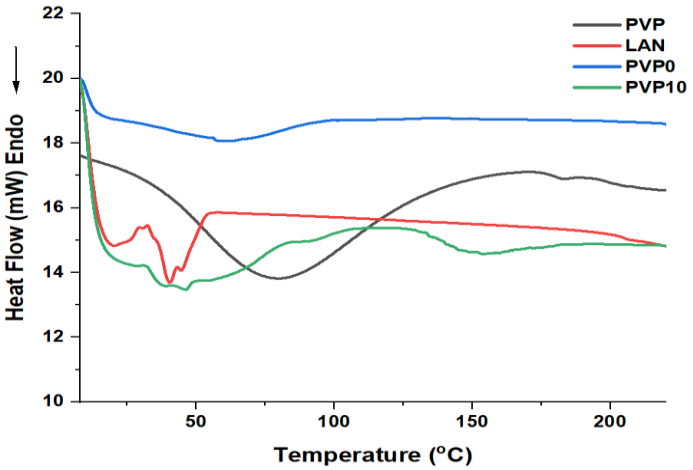


Figure 6. DSC thermograms of PVP, LAN and PVP based nanofibers

The thermal properties of PVP polymer, Lanolin, and PVP-based nanofibers were measured by DSC. Due to how volatile Carvcarol is, DSC measurements were unable to be performed effectively. According to the DSC thermograms in Figure 6, the PVP polymer did not exhibit any fusion peaks or phase transitions, similar to previous literature studies. Besides a wide endotherm brought on by dehydration, which takes place between 70 and 120 °C [36]. When the thermogram of PVP0 nanofibers was examined, it was observed that the endothermic peak width of the PVP0 nanofibers decreased compared to the thermogram of the PVP polymer. Additionally, in both nanofibers, the PVP polymer peak appeared at lower temperatures. This is assumed to be caused by a change in the chain structure of PVP polymers during the production of nanofibers. An exothermic peak was seen in the Anhydrous Lanolin thermogram between 25 and 35 °C, while an endothermic peak was seen between 45-50 °C [37]. According to the amount of lanolin it contains, as expected, the DSC thermogram of the lanolin-loaded PVP10 nanofibers was similar to the lanolin thermogram. But there was a slight increase in the degradation temperatures. The reason for this is that PVP nanofibers improve the low temperature stability of lanolin.

4. CONCLUSION

In this work, it is achieved to produce and characterize PVP based carvacrol and lanolin loaded composite nanofibers successfully. Pure PVP solution based nanofibers had a lot of bead defects but nanofibrous web quality was improved with addition of carvacrol:lanolin to the PVP polymer solutions with same PVP concentration. In this way, very smooth, fine, homogeneous, and uniform fibers were obtained without increasing the polymer concentration and without increasing the average fiber diameter prominently. In addition, the chemical structure of the nanowebs was investigated by FT-IR analysis. No undesirable reaction occurred between the components during the preparation of the polymer solution including many components, and all the components in the polymer solution in the produced nanowebs were chemically determined. In DSC and TGA analysis, it was determined that the thermal stability of carvacrol and lanolin increased with the inclusion of PVP nanofibers. Considering the properties of carvacrol and lanolin in the content of biocompatible nanofiber surfaces, it is thought that there is a potential for use in the medical application areas especially as a wound dressing.

ACKNOWLEDGMENT

This research was supported by Süleyman Demirel University Scientific Research Project Funding Unit, Project No: Project No: FYL-2018-5824. The authors would like to thank Süleyman Demirel University Natural Products Application and Research Center for supplying carvacrol.

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