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Production and Morphological Characterization of Nanofiber Membrane with Natural Wound Healing

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Wound is defined as the cessation of the structure and functional function of living tissue as a result of the deterioration of the entire skin for various reasons. Wound treatment started with human history. The main purpose in wound treatment is; It is to correct the deteriorated skin structure and functional properties by providing tissue repair as soon as possible. For this reason, there are many different techniques in wound treatment today.

In this study, the production of composite membranes, in which additives with known natural wound healing properties were added to various polymers, was achieved by nanotechnological electrospinning technique. Material properties were determined by performing morphological (Scanning Electron Microscope-SEM) analysis on the produced membranes. Our nanofiber composite material produced can be an ideal material that can lead various sectors, especially the health sector, with its functional properties.

Keywords: *Natural wound healing, polymer, electrospinning, nanofiber membrane.*

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1. Introduction

Wound; It is defined as the disruption of the integrity of the skin or mucosa with any cutting, piercing, wounding and similar objects on the inner and outer surfaces of the soft tissues. It is known as maintaining the integrity of the skin or mucous membrane damaged by accident or injury. Wound healing; It occurs as the inflammation-inflammation phase (lasting 1-5 days), proliferation-cellular proliferation phase (5-14 days) and maturation-tissue maturation phase (after 14 days) [1].

Biomaterial; They are materials that can be produced naturally or synthetically, which are in constant contact with living tissue in order to fulfill the functions of living tissues in a living system, support or treat their functional part. Biomaterials are divided into four main classes. These; metals, ceramics, polymers and composites [2].

Composite; It is a type of biomaterial created by two or more materials by preserving their tests. It can consist of reinforcement and matrix structures. In the electrospinning method, polymers are used as matrix materials, while additives are used as reinforcing elements [3].

Electrospinning method

It is the process of converting the fluid into nano-sized fibers by applying tension in kilovolt to viscous liquids at very small flow rates. In these applications, polymer solution or melt is used as viscous liquid. The solution arriving at the tip of the medical syringe at uniform flow forms a spherical droplet under the influence of surface tensions, where, under the effect of electrical forces, it is tapered (taylor cone) and transferred to the grounded collector at a certain distance as nanofiber [4].

Natural wound healing

Clover (*Medicago sativa*)

Its name in international terminology is "Alfalfa" of Arabic origin. From legume family; Although it is widely used as both wet and dried animal feed due to its high protein and mineral content, it has many benefits for human health. It is a plant that accelerates the healing of diabetic injuries.

Legume (*Agrimonia eupatoria*)

It is used for healing other skin problems such as wounds. For this purpose, a poultice is prepared by crushing the fresh aboveground parts of the plant and applied externally to the wounds. It grows naturally in almost every region of Turkey.

Chamomile (*Matricaria chamomilla*)

As the European Medicines Agency (EMA) confirmed a few years ago, chamomile is highly effective in improving benefits and treating eczema and all forms of inflammation when used topically. One of the best properties of chamomile is that it is anti-allergic.

Garlic (*Allium sativum*)

It is a great anti-bacterial and anti-fungal and can help heal almost any wound. Garlic can damage your skin, so make sure you don't leave it on the skin for longer than 20-25 minutes.

Honey

It is an ideal product to accelerate wound healing.According to the literature, honey is actually more effective than antibiotics. In lab tests, it has been found that honey kills most bacterial cells and prevents infections from occurring. One of the important points is that raw honey must be used to dress the wound.

Liver grass (*Pulmonaria officinalis*)

It is a perennial herbaceous plant species from the Boraginaceae family, which blooms between April and May, and is 10–50 cm tall. It provides faster drying and recovery of the inflamed areas in various parts of the body. Thanks to its antiseptic properties, it can create a germicidal effect and support open wounds to heal faster and healthier.

Mighty Pomegranate (*Momordica charantia*)

Being from the cucurbit family, which is grown in many regions of Asia and Africa and in some cities in our country, the mighty pomegranate is a type of plant used as food and medicine. It can be consumed as a fruit in the summer and thanks to its seeds, potion oil and paste are made and used. This plant species, which has many benefits, can prevent bacterial growth in the stomach and intestines and treat digestive system infections. In addition, its antibiotic properties have been proven and it can heal wounds and burns on the skin in a short time.

Pine gum (*Poix Regine*)

It is produced spontaneously among the bark of fir and spruce trees, which are members of the pine family, and it prevents the tree trunk from being damaged by closing the cracks in the tree trunk. Besides benefiting the tree, it has many effects on human health. It can renew the skin and prevent wrinkles by enabling the facial muscles to work. It protects dental health by strengthening the teeth. It provides a quick healing of wounds by making it into a paste form for wounds and has an antiseptic feature as it is a powerful product [5]. Figure 1.1. Contains natural wound healing.

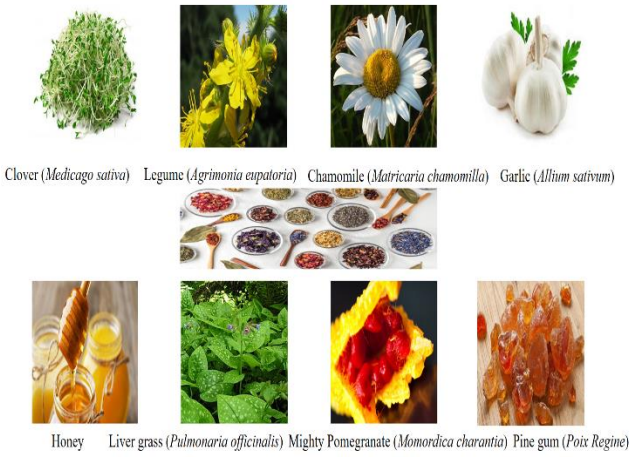

Figure 1.1. *Overview of natural wound healing*

In this study, polymeric nanofiber production will be carried out by electrospinning technique using natural wound healing additives. The nanofiber membranes to be obtained are aimed to be ideal wound healing and to be used in various sectors with their functional properties.

2. Material and Method

2.1. Material

Natural wound healing agents such as alfalfa, pubicum, chamomile, garlic, honey, liver out, mighty pomegranate, pine gum were supplied from herbalists and plant growers around us. Polycaprolactone (PCL), polyvinylalcohol

(PVA), polyacrylonitrile (PAN), polyethyleneoxide (PEO), polyvinylpyrrolidone (PVP), polyurethane (PU), shape memory polyurethane (SMPU), polystyrene (PS) and polylactic acid (PLA). Various solvents for dissolving polymers and polymers were obtained from Sigma / Aldrich. In the electrospinning stage, oil paper was used as the substrate.

2.2. Method

2.2.1. Preparation of wound healing solutions

Natural wound healing solutions were prepared by reinforcing various polymers with the values listed in Table 2.1.

Table 2.1.: Preparation parameters of wound healing solutions

Polymer / Additive	Solvent	Mixture temp. (˚C)	Mixture time (minute)
10% PCL	DMF / Chloroform	60	60
10% PVA	Pure water	60	60
10% PAN	DMF / Chloroform	60	65
10% PEO	Pure water	70	70
10% PVP	Pure water		
10% PU	DMF / Chloroform	70	70
10% SMPU	DMF / Chloroform	70	70
10% PS	DMF / Chloroform	70	70
10% PLA	DMF / Chloroform	70	70
10% PCL-5% Clover	DMF / Chloroform	60	60
10% PVA-5% Pubic Grass	Pure water	60	60
10% PAN-5% Chamomile	DMF / Chloroform	60	65
10% PEO-5% Garlic	Pure water	70	70
10% PVP-5% Honey	Pure water		
10% PU-5% Liver Grass	DMF / Chloroform	70	70
10% SMPU-5% Mighty Pomegranate	DMF / Chloroform	70	70
10% PS-5% Pine Gum	DMF / Chloroform	70	70
10% PLA-1% Clover-1% Groin Out-1% Chamomile-1% Garlic-1% Honey-1% Liver Grass-1% Power Pomegranate-1% Pine Gum	DMF / Chloroform	70	70

2.2.2. Wound healing nanofiber membrane production by electrospinning method

The electrospinning parameters required for wound healing nanofiber membrane production are shown in Table 2.2.

Table 2.2.: Electrospinning parameters required for wound healing nanofiber membrane production

Polymer / Additive	Working distance (cm)	Flow rate (ml/hour)	Voltage (kV)
10% PCL	15	2.5	30
10% PVA	15	2.5	30
10% PAN	15	3.0	30
10% PEO	15	3.0	30
10% PVP	15	2.5	30
10% PU	15	2.5	30
10% SMPU	15	3.0	30
10% PS	15	3.0	30
10% PLA	15	2.5	30
10% PCL-5% Clover	15	2.5	30
10% PVA-5% Pubic Grass	15	3.0	30
10% PAN-5% Chamomile	12	3.0	31.0
10% PEO-5% Garlic	12	3.0	31.0
10% PVP-5% Honey	12	3.0	31.0
10% PU-5% Liver Grass	12	3.0	31.0
10% SMPU- 5% Mighty Pomegranate	12	3.0	31.0

10% PS-5% Pine Gum	12	3.0	31.0
10% PLA-1% Clover-1% Groin Out-1% Chamomile-1% Garlic-1% Honey-1% Liver Grass-1% Power Pomegranate-1% Pine Gum	12	3.0	31.0

The production stages of wound healing nanofiber membrane by electrospinning method are given in Figure 2.1.

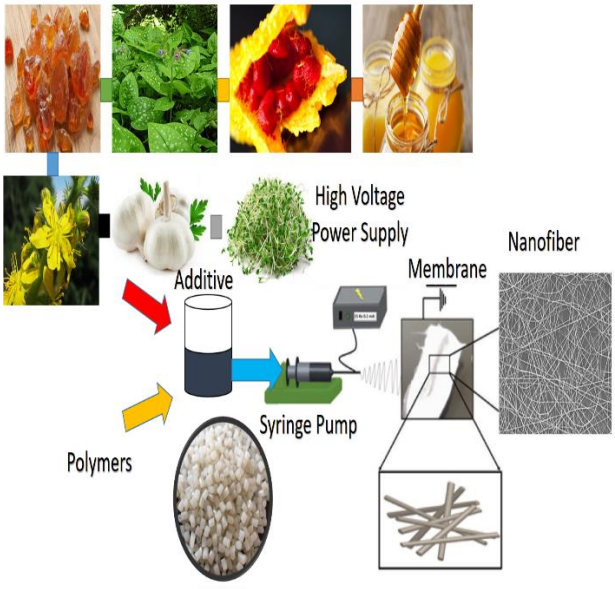

Figure 2.1. *Wound healing nanofiber membrane production stages by electrospinning method*

2.2.3. Characterization studies
* Morphological characterization

The nanofiber diameters of the produced wound healing nanofiber membranes were investigated in FEI FEGSEM QUANTA 250 brand device at 7 kV potential. 40 nanofiber diameters were measured from the images and their arithmetic averages were obtained and average nanofiber diameter ranges were determined.

3. Results and Discussion
* SEM analysis

Nanofiber formation was observed in all samples. In addition to the presence of different polymers and solvents, the natural wound healing and electrospinning working parameters affect the morphology of nanofiber membranes. The nanofiber diameter distribution range of the samples was extracted by calculating the diameters of 40 nanofibers and taking arithmetic averages. Natural wound healing agents used as additives have achieved nanofiber thinning by reducing the viscosity of the polymer solution [6-10]. The diameter distribution range values of nanofiber membranes are shown in Table 3.1. SEM morphological images of all samples are shown in Figure 3.1.

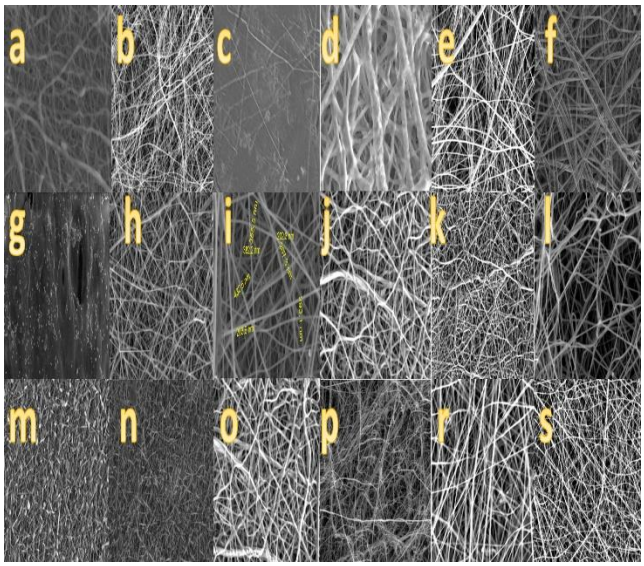

Figure 3.1. *(a) PCL, (b) PAN, (c) PEO, (d) PLA, (e) PS, (f) PU, (g) SMPU, (h) PVA, (i) PVP, (j) 10% PCL-5% Clover, (s) 10% PVA-5% Pubic Grass, (l) 10% PAN-5% Daisy, (m) 10% PEO-5% Garlic, (n) 10% PVP-5% Honey , (o) 10% PU-5% Liver Grass, (p) 10% SMPU-5% Mighty Pomegranate, (r) 10% PS-5% Pine Gum, (s) 10% PLA-1% Clover-1% Groin Out-1% Chamomile-1% Garlic-1% Honey-1% Liver Grass-1% Peppermint-1% Pine Gum SEM image*

Table 3.1.: Diameter range values of nanofiber membranes

Polymer / Additive	Nanofiber diameter distribution range of samples (nm)
10% PCL	120-345
10% PVA	180-490
10% PAN	50-350
10% PEO	150-550
10% PVP	220-487
10% PU	350-640
10% SMPU	140-910
10% PS	190-400
10% PLA	450-750
10% PCL-5% Clover	90-280
10% PVA-5% Pubic Grass	150-350

10% PAN-5% Chamomile	40-240
10% PEO-5% Garlic	110-500
10% PVP-5% Honey	135-345
10% PU-5% Liver Grass	250-545
10% SMPU- 5% Mighty Pomegranate	90-450
10% PS-5% Pine Gum	155-300
10% PLA-1% Clover-1% Groin Out-1% Chamomile-1% Garlic-1% Honey-1% Liver Grass-1% Power Pomegranate-1% Pine Gum	40-220

4. Conclusion

Nanofiber formation was observed successfully in all samples. Nanofiber diameters were thinned with the addition of natural wound healing compared to polymers. In addition to different polymers and solvents, electrospinning operating parameters can change the morphology of membranes. The thinnest nanofibers in the study are in the range of 40-220 nm, 10% PLA-1% Clover-1% Groin Out-1% Chamomile-1% Garlic-1% Honey-1% Lung Grass-1% Mighty Pomegranate-1% Pine Gum observed in the sample. The results obtained can be used in sectors such as textile, food, agriculture, filtration, defense as well as the health sector as an ideal wound healing of the polymeric matrix composites produced.

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