

## Chemical Profile of *Ficus carica* L. Leaf Methanol Extract and Its Effect on *In vitro* Wound Healing

Handan UĞUZ BAYRAKÇEKEN<sup>1,2\*</sup>, Bahri AVCI<sup>2</sup>, Ayşe Nurseli SULUMER<sup>2</sup>,  
Esra PALABIYIK<sup>3</sup>, Hakan AŞKIN<sup>2</sup>, Murat ÇELİK<sup>4</sup>

<sup>1</sup> Ataturk University, Faculty of Agriculture, Department of Field Crops, 25240, Erzurum, Türkiye

<sup>2</sup> Ataturk University, Faculty of Science, Department of Molecular Biology and Genetics, 25240, Erzurum, Türkiye

<sup>3</sup> Ağrı İbrahim Çeçen University, Faculty of Science and Letters, Department of Molecular Biology and Genetics, 04100, Ağrı, Türkiye

<sup>4</sup> Ataturk University, Faculty of Science, Department of Chemistry, 25240, Erzurum, Türkiye

**Received:** 03/03/2025, **Revised:** 30/06/2025, **Accepted:** 30/09/2025, **Published:** 30/03/2026

### Abstract

*Ficus carica* L. is among the earliest known cultivated plants of the family *Moraceae*. It constitutes one of the most important angiosperm genera, encompassing more than 800 species of shrubs, epiphytes, and trees that have been planted in several tropical and subtropical regions across the globe. *Ficus carica* (FC) leaves have been used not only for tea but also for traditional medicine purposes throughout history. In study, the content of *Ficus carica* leaf methanol extract (M-FCL) and its proliferation and cell viability properties in Human Dermal Fibroblast (HDF) cell culture were examined. In the first stage of the study, different components in the structure of the extract were identified using gas chromatography-mass spectrometry (GC-MS) and secondly, HDF cell viability (WST-8) and wound healing activity were evaluated. HDF cell viability (WST-8) and wound healing activity were evaluated. To evaluate the wound healing effects of the extract, microscopic images were taken at three-hour intervals (0, 3, 6, 9, 12 and 15 hours) until the wound was closed in any group. Then, they were analyzed with Image J program. Fourteen chemical components were identified in the M-FCL extract by means of GC-MS. Analysis reveals that the main constituent of M-FCL is  $\alpha$ -tocopherol (28.181 %). As a result of WST-8 analysis, it was observed that cell viability increased significantly ( $p < 0.01$ ) at concentrations between 31.25-250  $\mu\text{g/mL}$ . It was observed that M-FCL accelerated wound healing at low concentrations.

**Keywords:** *Ficus carica* L., Gas Chromatography-Mass Spectrometry, HDF, Cell proliferation, Wound healing

## *Ficus carica* L. Yaprak Metanol Ekstraktının Kimyasal Profili ve *In vitro* Yara İyileşmesi Üzerindeki Etkisi

### Öz

*Ficus carica* (Fig.) L., *Moraceae* familyasının bilinen en eski kültür bitkileri arasındadır. Dünyanın çeşitli tropikal ve subtropikal bölgelerinde ekilen 800'den fazla çalı, epifit ve ağaç türünü kapsayan en önemli angiosperm cinslerinden birini oluşturur. *Ficus carica* (FC) yaprakları sadece çay olarak değil, aynı zamanda tarih boyunca geleneksel tıp amaçları için de kullanılmıştır. Çalışmada, *Ficus carica* yaprak metanol özütünün (M-FCL) içeriği ve insan dermal fibroblast (HDF) hücre kültüründe çoğalma ve hücre canlılığı özellikleri incelenmiştir. İyileştirme aşamasında, özütün yapısındaki farklı bileşenler gaz kromatografisi-kütle spektrometrisi (GC-MS) kullanılarak tanımlanmış ve ikinci olarak HDF hücre canlılığı (WST-8) ve yara iyileştirme aktivitesi değerlendirilmiştir. HDF hücre canlılığı (WST-8) ve yara iyileştirme aktivitesi değerlendirilmiştir. Ekstraktın yara iyileştirme etkilerini değerlendirmek için, herhangi bir grupta yara kapatılıncaya kadar üç saatlik aralıklarla (0, 3, 6, 9, 12 ve 15.saatler) mikroskopik görüntüler alınmıştır. Daha sonra, Image J programı ile analiz edilmiştir. M-FCL ekstraktında GC-MS aracılığıyla on dört kimyasal bileşen belirlendi. Analiz, M-FCL'nin ana bileşeninin  $\alpha$ -tokoferol (%28.181) olduğunu ortaya koymaktadır. WST-8 analizi sonucunda, hücre canlılığının 31.25-250  $\mu\text{g/mL}$  arasındaki konsantrasyonlarda önemli ölçüde ( $P < 0.01$ ) arttığı görüldü. M-FCL'nin düşük konsantrasyonlarda yara iyileşmesini hızlandırdığı görüldü.

**Anahtar Kelimeler:** *Ficus carica* L., Gaz Kromatografisi-Kütle Spektrometrisi, HDF, Hücre çoğalması, Yara iyileşmesi

## 1. Introduction

Wound is defined as an injury that disrupts the structure and function of a tissue at the cellular level [1]. Wounds can occur due to various reasons including physical impact, infection (microbes), immune system, chemical agent, heat, radiation, or electric shock, all of which impair tissue integrity [2]. Wounds are generally classified as either acute or chronic. While acute wounds heal without leaving a scar in a short time, chronic wounds may take longer to heal and may leave a scar [3]. Wound healing is a complex process that includes hemostasis, inflammation, proliferation and remodeling stages. The main purpose of wound healing is to repair damaged tissue [4, 5, 6].

In modern medical practice, both local and systemic drugs are employed to treat wounds and to regulate disrupted tissue integrity [7]. A range of pharmaceutical agents, including topical medicines, analgesics, antiseptics, antibiotics and herbal drugs, are utilised to ensure tissue integrity and facilitate wound healing [6, 8]. Herbal drugs refer to treatments that contain specific plant components or the entire plant [9].

The genus *Ficus* (family *Moraceae*) is one of the largest genera of angiosperms, comprising over 800 species of trees, epiphytes, and shrubs predominantly found in tropical and subtropical regions [10]. *Ficus carica*, a species widely cultivated in the Bursa region of Türkiye, thrives in diverse environments such as open fields, mixed forests, rocky slopes, valleys, and crevices. It is also one of the earliest domesticated fruit plants. Traditionally, its fruit, latex, and leaves have been used to treat various ailments since ancient times [11].

In the context of wound healing treatment, the objective is to augment the population of fibroblast cells through the utilization of antioxidants and antiseptic agents [12]. The release of oxygen radicals serves to eliminate invading foreign organisms and purge the fibrin matrix, thereby expediting the healing process [13]. Fibroblast cells, which initiate the proliferative phase, play an important role in the wound healing process [1]. In this context, studies on the *Ficus* genus have previously determined that extracts obtained from different parts of the plant have various pharmacological activities, including anti-inflammatory, hemostatic, antioxidant, antimicrobial and antiviral properties [15, 16, 17].

Phytochemical studies on *Ficus carica* have resulted in the isolation of a number of hydrocarbons, organic acids, phenolic compounds, phytosterols, anthocyanins, fatty acids, aliphatic alcohols, volatile compounds and other classes of secondary metabolites from various parts of the plant. The concentration of these phytochemicals is highest in the latex, followed by the leaves, fruits and roots [18].

In recent years, there has been a growing interest in the use of plant-based therapies in both cosmetic and medical applications. Although numerous studies have analyzed the chemical composition of *Ficus carica*, limited research has focused on the chemical content of leaves from the Bursa region and their biological activities using gas chromatography–mass spectrometry (GC-MS). Therefore, this study aims to identify the chemical constituents of the methanol extract of *Ficus carica* leaves collected from the Bursa province and to evaluate its

proliferative effects, oxidative stress modulation, and wound healing potential on Human Dermal Fibroblast (HDF) cells.

## 2. Materials and Methods

### 2.1.1. Reactives and solutions

Methanol solvent used for the study was provided by Isolab. In our study, HDF cells (PCS-201-012-ATCC) were used in *in vitro* wound experiments. Dulbecco's Modified Eagle Medium (DMEM), Penicillin-Streptomycin (Pen-strep) solution, Fetal Bovine Serum (FBS), Trypsin-EDTA (Trypsin-ethylenediaminetetraacetic acid) and Cell Viability Determination WST-8 kit were supplied by EcoTech Biotechnology.

### 2.1.2. Supply of plant material

Leaves of the *Ficus carica* plant, commonly known as “Bursa siyah inciri” (Bursa black fig), were collected from a fig garden in the Çamdibi neighborhood of Bursa's İznik district in September 2020. The location of the plant is 40.403°N 29.747°E.



**Figure 1.** *Ficus carica* leaves

### 2.1.3. Preparation of *Ficus carica* leaves for analysis

The leaves of the *Ficus carica* tree were collected and dried in a thin layer in conditions that maintained low temperatures and low levels of light. The dried leaf samples were then ground into a powder using a mortar and pestle and stored at -18°C for use in the subsequent study. Most of the studies under review were conducted within the Genetics and Organic Chemistry Laboratories of Atatürk University's Faculty of Science.

## 2.2. Methanol extraction of *Ficus carica* leaves

After the pre-dried *Ficus carica* leaves (100 g) were ground into powder, 1000 mL methanol (CH<sub>3</sub>OH) was added as a solvent. It was macerated for 72 h. Filtration was performed to remove waste materials in a reflux system (4 h, 60-80°C) [19]. An evaporator was used to remove the solvent in the sample (155 rpm, 40°C). After this procedure, it was prepared for GC-MS analysis, which was performed three times to increase the yield.

### 2.3. GC-MS system and chromatographic conditions

The 7820A GC-MS, 5977 Mass Spectrometry Detector and 7673 Series Autosampler were used to determine the chemical profile of Plantain. Injection and detector temperatures were set at 250 °C. The injection capability was determined as a 1 µl indivisible injection mode, helium as the carrier gas, a flow rate of 1ml/min and an ionization energy of 70 [20]. A variety of temperatures were used during the performance of the GC-MS procedure. Accordingly, the adjustment was made to increase by 50°C for 1 minute, to increase by 20°C per minute for 1 minute at 100°C, to increase by 10°C per minute for 1 minute at 180°C, and to increase by 5°C per minute for 1 minute. The components in the extract were identified by comparing the chromatograms and mass spectra with those of the reference standard substance [19].

### 2.4. Cell culture experiments

The *in vitro* wound study was conducted on healthy Human Dermal Fibroblast (HDF) cell line ATCC® PCS-201-012™). The aim of the study was the proliferation of HDF cells in an incubator environment containing 5% CO<sub>2</sub> at 37°C. DMEM (Dulbecco's Modified Eagle Medium) containing 10% FBS (Fetal Bovine Serum) and 1% Pen-STREP (100 U penicillin /0.1 mg/mL streptomycin) was used in the study.

The cells were initially seeded in T25 cell culture flasks and incubated in an incubator at 37 °C and 5% CO<sub>2</sub>. Once the cells had reached 70–80% confluence, they were transferred to T75 flasks via passage.

### 2.5. Cell seeding and counting

Cell counting was performed using an automatic cell counter. Adherent cells were detached using 0.1% trypsin/EDTA in a T75 culture dish and spun at 1200 rpm for 5 minutes. The resulting cell solution was mixed with trypan blue at a ratio of 1:1, laminated, and the total cell number was determined.

96-well plates were seeded with 5 x 10<sup>3</sup> HDF cells in 100 µl medium each. After cell seeding was completed, the plates were placed in an incubator containing 5% CO<sub>2</sub> at 37°C.

### 2.6. Preparation of the M-FCL extract and application to cells

1 mg of methanol extract (M-FCL) obtained from *Ficus carica* leaves was dissolved by adding 100 µL of DMSO and 900 µL of DMEM and filtered through a 0.22 µm filter. Then, stock solutions at concentrations of 15.625; 31.25; 62.5; 125; 250 and 500 µg/mL were prepared by serial reduction method. HDF cells was exposed to M-FCL at concentrations ranging from 15.625–500 µg/mL for 48 hour.

### 2.7. WST-8 Analysis

WST-8 analysis was performed to determine cell proliferation, i.e. the amount of live cells, and Ecotech Cell Viability Determination Kit-8 (Cat No: CVDK-8) was used for the analysis. HDF

cells were incubated in 96-well plates at  $5 \times 10^3$  cells per well. When the cell density was 70-80% confluent, extracts were applied at 6 different concentrations. The first well was used as a control group in which no substance was applied. After 48 hours of incubation, 10% of the well volume of WST-8 solution was added and the plates were incubated at 37 °C, 5% CO<sub>2</sub> for 3 hours. During the 3-hour incubation period, absorbance values were measured at 450 nm wavelength every hour. The process was repeated at 3 different times and viability/cytotoxicity values were calculated.

$$\text{Cell viability \%} = \left[ \frac{\text{Absorbance (dose well)}}{\text{Absorbance (control dose)}} \right] * 100$$

## 2.8. Scratch Assay

Concentrations obtained from *F. carica* leaf methanol extraction WST-8 analysis were used to determine the migration capacity of healthy fibroblast cells using scratch analysis.  $5 \times 10^5$  cells were seeded in 2 ml medium in 3 replicates in each well of 6 well plates [1]. Then, the cells in the plates were incubated in a 5% CO<sub>2</sub> and 37°C oven until they reached 100% occupancy rate. A wound was created by making a single scratch with a sterile 0,1-10 µl pipette tip in a monolayer cells. Then, the wound area was created, and the lifted cells and their residues were washed with PBS and removed. Then, the medium containing drugs at the determined concentrations were added to the wells. The cells were observed with an inverted microscope (Zeiss AXIO Vert A1, Axiocam 208 Color) at 0., 3., 6., 9., 12. and 15. hours. The images were recorded.

In our study, after creating a wound model with scratch assay, *Ficus carica* leaf methanol extract was applied at two different concentrations. The wound area was then photographed at regular intervals using an inverted microscope. The migration percentage of cells was calculated with the following formula by measuring with Image J program.

$$\% \text{ Migration} = \frac{\text{0. hour intercellular distance} - \text{desired time intercell distance}}{\text{0. hour intercellular distance}} * 100$$

## 2.9. Statistical Analysis

All experiments were performed in three repetitions. Subsequent to the completion of the experiments, a One-Way Analysis of Variance (ANOVA) and a Tukey's Multiple Comparison test were performed based on the data obtained as a result of the experiments. The analysis was conducted utilising the GraphPad Prism version 8.0.2 package program (GraphPad Software, San Diego, USA) to facilitate the evaluation of the data. All results were expressed as mean  $\pm$  standard deviation. The statistical significance level was evaluated as  $p < 0.01$ .

### 3. Results and Discussion

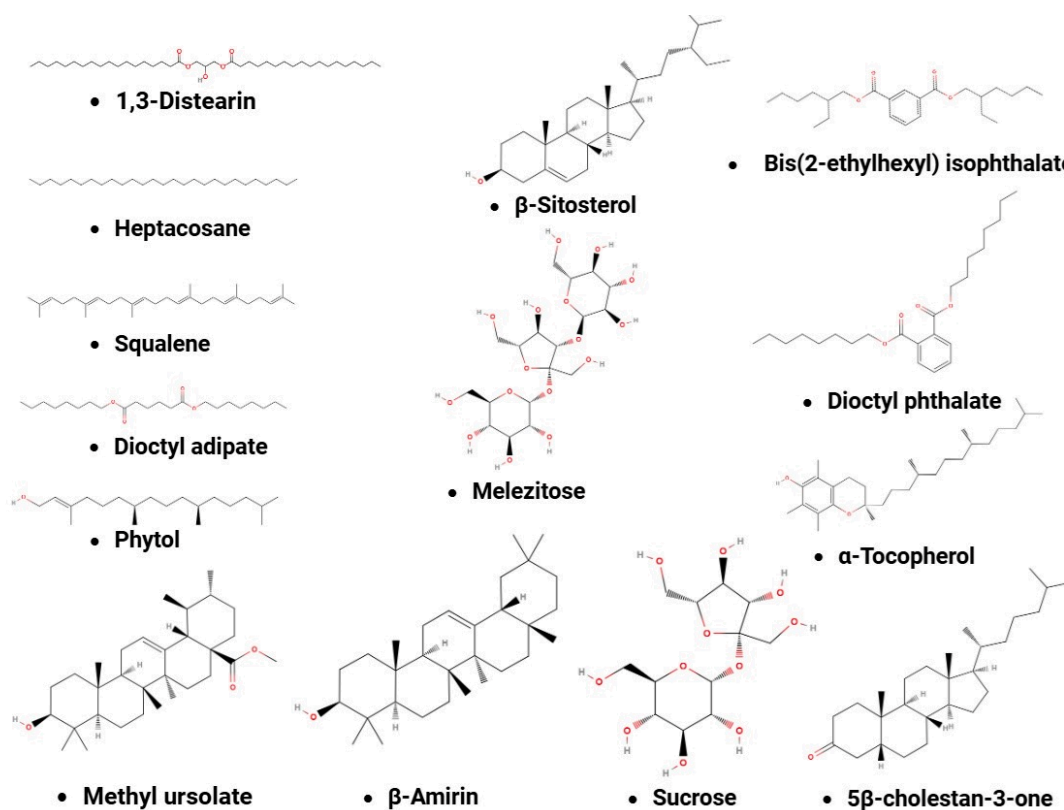
#### 3.1. GC-MS analysis of *Ficus carica* leaves methanol extract

A GC-MS analysis was performed to determine the chemical content of *Ficus carica* leaf methanol extract, and fourteen compounds were identified. These compounds are listed in detail in Table 1.

**Table 1.** Structural formulas of compounds in *Ficus carica* L. leaf methanol extract

Peak	Retention Time(min.)	% of total	Compound	Molecular Formula
1	12.614	22.6	Sucrose	C <sub>12</sub> H <sub>22</sub> O <sub>11</sub>
2	13.053	0.479	Melezitose	C <sub>18</sub> H <sub>32</sub> O <sub>16</sub>
3	19.005	3.803	Phytol	C <sub>20</sub> H <sub>40</sub> O
4	27.796	2.625	Diethyl adipate	C <sub>22</sub> H <sub>42</sub> O <sub>4</sub>
5	30.411	4.479	Diethyl phthalate	C <sub>24</sub> H <sub>38</sub> O <sub>4</sub>
6	32.223	2.154	Heptacosane	C <sub>27</sub> H <sub>56</sub>
7	33.181	3.426	1,3-Distearin	C <sub>39</sub> H <sub>78</sub> O <sub>4</sub>
8	32.775	4.045	Bis(2-ethylhexyl) isophthalate	C <sub>19</sub> H <sub>38</sub> O <sub>4</sub>
9	33.537	6.891	Squalene	C <sub>30</sub> H <sub>50</sub>
10	34.671	1.989	5 $\beta$ -cholestan-3-one	C <sub>27</sub> H <sub>46</sub> O
11	35.487	3.355	Methyl ursolate	C <sub>31</sub> H <sub>50</sub> O <sub>3</sub>
12	36.470	28.181	$\alpha$ -Tocopherol	C <sub>29</sub> H <sub>46</sub> O
13	38.800	13.607	$\beta$ -Sitosterol	C <sub>29</sub> H <sub>50</sub> O
14	39.349	2.366	$\beta$ -Amirin	C <sub>30</sub> H <sub>50</sub> O

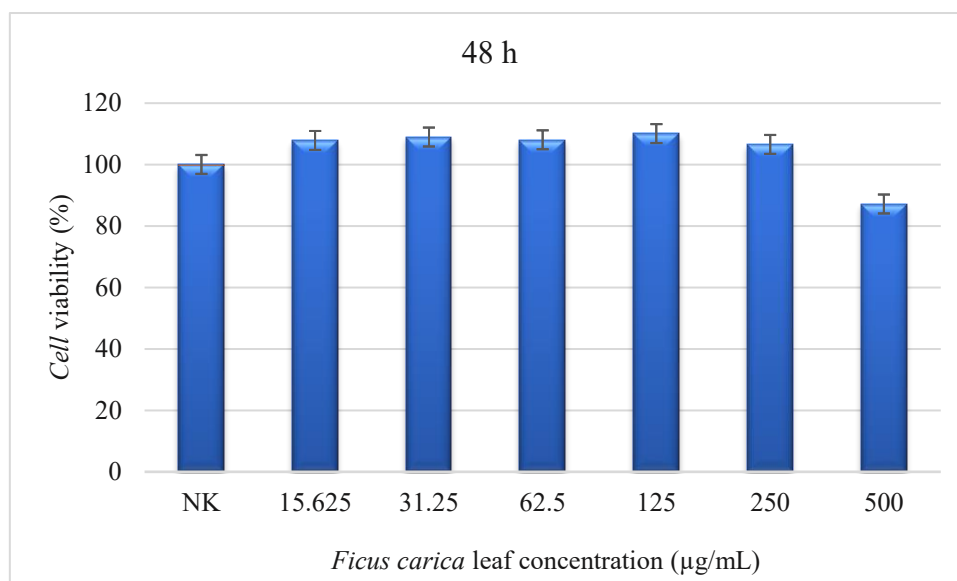
In the 40-minute GC-MS analysis period, the components began to appear between 12 and 40 minutes. In this period,  $\alpha$ -tocopherol (28.181%) was identified as the main component. Following this component, respectively; Sucrose (22.6%),  $\beta$ -sitosterol (13.607%), squalene (6.891%), methyl ursolate (3.355%), 1,3-distearin (3.426%), phytol (3.803%), diethyl phthalate (4.479%), bis(2-ethylhexyl) isophthalate (4.045%), diethyl adipate (2.625%),  $\beta$ -Amirin (2.366%), heptacosane (2.154%), 5 $\beta$ -cholestan-3-one (1.989%) and melezitose (0.479%).



**Figure 2.** Bioactive components of M-FCL extract

### 3.2. WST-8 Analysis Results for Proliferative Effect

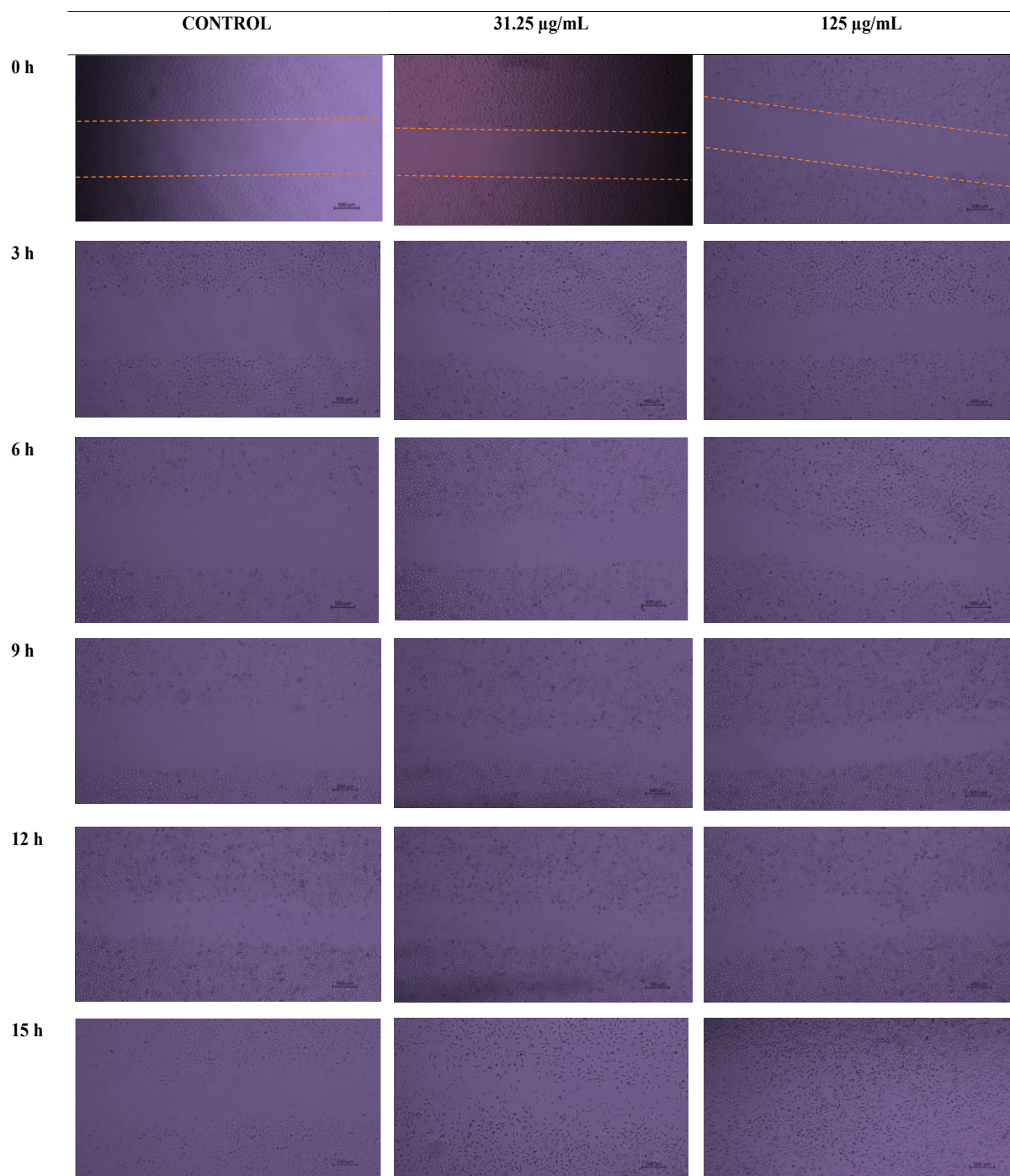
WST-8 test was used to determine the effect of M-FCHL extract on cell viability and cell proliferation on HDF cells. The results of WST-8 test obtained after 48 hours at concentrations of 31.25 to 500  $\mu\text{g/mL}$  of M-FCHL extract are presented in Fig. 3. The findings revealed that the application of a methanol leaf extract obtained from plant leaves at a concentration range of 31.25-250  $\mu\text{g/mL}$  for 48 hours had a stimulatory effect on the migration of HDF cells. As a result, it was shown that M-FCHL extract exerted a proliferative effect on HDF cell line at each applied dose.



**Figure 3.** The effect of M-FCL extract on HDF cell viability and proliferation was assessed by WST-8 assay.

### 3.3. Scratch Assay Results for Wound Healing Effects

The effect of M-FCL extract on wound healing was investigated using scratch test. In the scratch test, the concentrations that showed the highest cell viability and proliferation as a result of WST-8 analysis were used. These concentrations were 31.25 and 125 µg/mL. The observations showed that cell proliferation and migration started in 6 hours at high concentrations and in 9 hours at low concentrations. In addition, complete closure of the wound area was recorded in 15 hours (Fig. 4). In addition, the cell migration results were consistent with the WST-8 results and it was found that 31.25 and 125 µg/mL concentrations of M-FCL extract had a high effect on both cell viability and wound healing.

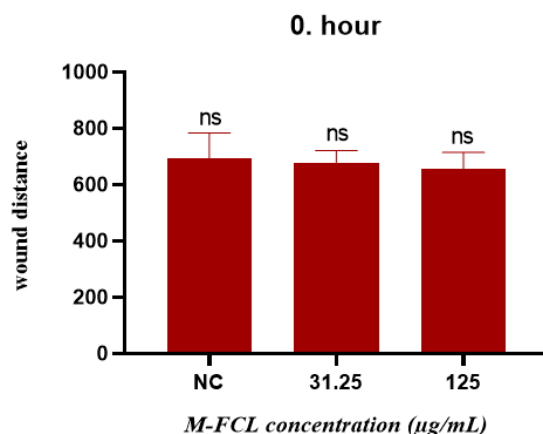


**Figure 4.** The present study investigates the effect of M-FCL extract on HDF cell migration. Monolayers of confluent HDF cells were exposed to different concentrations of M-FCL extract using 0.1–10 µL plastic micropipette tip to form an *in vitro* wound model. Photographs were taken under a microscope at 4x magnification at 0, 3, 6, 9, 12 and 15 h.

#### 3.4.Determination of Wound Healing Rate by Image J Analysis

After creating a wound model with scratch assay, M-FCL mixture was applied at 2 different concentrations. The created wound area was photographed with an inverted microscope at

three-hour intervals. The numerical data obtained by measuring with Image J program were compared with NC and analyzed statistically.



**Figure 5.** Initial wound area measurement at 0 h.

As a result of the statistical comparison of the wound width between all groups at hour 0, it was determined that there was no significant (relative) difference between the groups. However, at hour 15, it was observed that the 125 µg/ml concentration closed the wound area more quickly and effectively than the other groups, and at the same time a homogeneous closure was achieved.

Türkiye is quite rich in terms of plant diversity [14]. Some plant species are not only used for nutritional purposes among the public, but also for medical purposes [21]. For one of these, the *Ficus* species, it is known that the above-ground parts (fruit, latex and leaves) are used for various purposes among the public [18] *Ficus carica* is a plant that grows in various tropical and subtropical regions, including Bursa, Türkiye.

In this study, the effects of methanol extract of *Ficus carica* leaves from Bursa region on cytotoxicity, cell viability and wound healing on Human Dermal Fibroblast (HDF) cells were evaluated for the first time. As a result of GC-MS analysis, a total of 14 compounds were identified in the M-FCL. In a study conducted on *Ficus carica* leaf extract in the literature, Turkoglu *et al.* (2017) detected 126 compounds in the aqueous extract in the analyses performed on HaCaT cells; they reported that this extract exhibited anti-inflammatory, anti-angiogenic, antimitotic and antiandrogenic effects [50]. Differences in the number of compounds may vary depending on the abiotic (climate, soil, light etc.) and biotic (microorganism, insect interaction etc.) conditions of the region where *Ficus carica* grows, as well as the solvent and extraction method used. One of the main compounds identified in our study,  $\alpha$ -tocopherol, or vitamin E, is a natural antioxidant [51].  $\alpha$ -tocopherol prevents oxidative damage by stabilizing the cell membrane, increases collagen cross-linking and prevents lipid peroxidation, and contributes to the delay of skin aging [52]. In addition, it has been reported in the literature that  $\alpha$ -tocopherol is used in the healing of sunburn and skin wounds, in the reduction of photo immune suppression, in the prevention of carcinogenesis and as a dermal immune system supporting agent [52].

Similarly,  $\beta$ -sitosterol, another important organic limited in our extract, is known to have antioxidant [41], antimicrobial [42] and anti-inflammatory [43] properties. Squalene is known to increase skin moisture, provide immune system modulation and protect against UV-induced damage [32]. The activities of other images obtained as a result of GC-MS are given in detail in Tab. 2.

**Table 2.** Reported activite of chemical compounds reported in M-FCL.

Chemical compounds	Reported Activities	References
<i>Sucrose</i>	Antimicrobial activity Antifungal activity Anti-tumor activity, Antioxidant activity Anti-depressive activity Neuro-protective activity,	[22, 23, 24, 25, 26, 27, 28, 29s]
<i>Melezitose</i>	Anticancer activity	[30]
<i>Phytol</i>	Antioxidant activity	[31]
<i>Squalene</i>	Antioxidant activity Anti-tumor activity Skin hydration	[32]
<i>5<math>\beta</math>-cholestan-3-one</i>	Hyperglycemia and Hyperinsulinemia	[33]
<i>Methyl ursolate</i>	Anti-microbial activity,	[34]
<i><math>\alpha</math>-Tocopherol</i>	Gene-Regulatory activity, Anti-inflammatory activity, Antioxidant activity, Wound healing activity	[35, 36, 37, 38, 39, 40]
<i><math>\beta</math>-Sitosterol</i>	Antioxidant activity, Antimicrobial activity, Anti-inflammatory activity, Anti-hyperlipidemia activity, Wound healing activity,	[41, 42, 43, 44, 45]
<i><math>\beta</math>-Amirin</i>	Anti-inflammatory activity, Antinociceptive activity, Antibacterial activity, Hepatoprotector activity,	[46, 47, 48, 49]

According to the WST-8 test, the extract in the range of 31.25–250  $\mu$ g/mL significantly increased HDF viability permanently ( $p < 0.01$ ). The obtained data show that the extract does not harm fibroblasts and even shows cell proliferation. Similarly, in the literature, cases where *Ficus* species extracts increase cell proliferation [16, 45]. In addition, scratch assay intervention showed that this proliferative effect is also reflected in wound healing. Especially in the 125  $\mu$ g/mL content, it was observed that the wound was completely closed within 15 hours. During this period, it is significantly shorter than the control group, and it is seen that fibroblast migration is adapted by the extract. In addition, the antioxidant capacity of phytol contained in the extract [31], the antimicrobial effect of methyl ursolate [34], and the anti-inflammatory and

hepatoprotective effects of  $\beta$ -amyryn [46-49] may have synergistic effects on the eyes. This synergy makes the wound healing powers of the extract stronger.

However, there are some limitations. First of all, there is the use of only *in vitro* systems. Since cell culture does not contain genetic, wound healing immune, medical or hormonal properties, these results need to be confirmed *in vivo* models. In addition, the fact that the extract was prepared with methanol solvent means that it only covers compounds with certain polarity, which may be the reason why the biological effects of non-polar images are ignored. In the future, extraction and fractionation with different solvents will allow for a clearer determination of the specific images that show effects.

#### 4. Conclusion

This study investigated the effects of *Ficus carica* leaf methanol extract on cell viability, proliferation, and *in vitro* wound healing in the HDF cell line by GC-MS content analysis obtained, *Ficus carica* leaf methanol extract accelerated wound healing by increasing cell viability and cell migration even at low concentrations. It is thought that the reason for this proliferative effect is due to the fatty acids, phenolic compounds, and vitamins found in the structure of *Ficus carica* leaf methanol extract. These compounds show prooxidant activity at high concentrations. For this reason, it was concluded that *Ficus carica* leaf methanol extract can be used as an alternative candidate agent in the treatment of wound healing in the pharmaceutical and cosmetic industry if supported by future *in vivo* studies.

#### Ethics in Publishing

This study contains plant material obtained from leaves of the species *Ficus carica*, which is not currently protected. Therefore, no official permit was required for the collection of this material. Furthermore, there are no ethical issues regarding the publication of this study.

#### Author Contributions

**H.U.B, H.A, and M.Ç:** Conceptualization and design of the study. **H.U.B, B.A, A.N.S and E.P:** Data collection and analysis. **H.U.B, B.A and A.N.S:** Interpretation of data, drafting of the article, and revision of the manuscript for substantial intellectual approval. All authors approved the manuscript to be submitted and published.

#### Acknowledgements

This work was supported by Atatürk University (grant number FCD-2021-9401). We would like to thank Assoc. Prof. Dr. Ufuk ATMACA, Faculty Member of the Chemistry Department of the Faculty of Science, for his support with the plant extraction and Prof. Dr. Bilal YILMAZ, Faculty Member of the Faculty of Pharmacy of Atatürk University, for his GC-MS analyses.

## References

- [1] Miloglu, F. D., Akpınar, A., Güven, L., Demirkaya, A. K., Gundogdu, G., Nalcı, K. A., & Hacimuftuoglu, A. (2023). Evaluation the effects of *Helichrysum plicatum* subsp. *pseudoplicatum* on an *in-vitro* wound model using human dermal fibroblast cells. *The International Journal of Lower Extremity Wounds*, 22(2), 401-408.
- [2] Foncerrada, G., Capek, K. D., Herndon, D. N., Lee, J. O., Sirvent, R. Z., & Finnerty, C. C. (2017). The state of the art on burn wound healing. *Journal Avid Science*, 4-52.
- [3] Raziyeva, K., Kim, Y., Zharkinbekov, Z., Kassymbek, K., Jimi, S., & Saparov, A. (2021). Immunology of acute and chronic wound healing. *Biomolecules*, 11(5), 700.
- [4] Sinno, H., & Prakash, S. (2013). Complements and the wound healing cascade: an updated review. *Plastic Surgery International*, 2013(1), 146764.
- [5] Bielefeld, K. A., Amini-Nik, S., & Alman, B. A. (2013). Cutaneous wound healing: recruiting developmental pathways for regeneration. *Cellular and Molecular Life Sciences*, 70, 2059-2081.
- [6] Oryan, A., Alemzadeh, E., & Moshiri, A. (2017). Burn wound healing: present concepts, treatment strategies and future directions. *Journal of Wound Care*, 26(1), 5-19.
- [7] Chin, J. S., Madden, L., Chew, S. Y., & Becker, D. L. (2019). Drug therapies and delivery mechanisms to treat perturbed skin wound healing. *Advanced Drug Delivery Reviews*, 149, 2-18.
- [8] Negut, I., Grumezescu, V., & Grumezescu, A. M. (2018). Treatment strategies for infected wounds. *Molecules*, 23(9), 2392.
- [9] Yuan, H., Ma, Q., Ye, L., & Piao, G. (2016). The traditional medicine and modern medicine from natural products. *Molecules*, 21(5), 559.
- [10] Singh, D., Singh, B., & Goel, R. K. (2011). Traditional uses, phytochemistry and pharmacology of *Ficus religiosa*: A review. *Journal of Ethnopharmacology*, 134(3), 565-583.
- [11] Hajam, T. A., & Saleem, H. (2022). Phytochemistry, biological activities, industrial and traditional uses of fig (*Ficus carica*): A review. *Chemico-Biological Interactions*, 368, 110237.
- [12] Oliveira, A., Simões, S., Ascenso, A., & Reis, C. P. (2022). Therapeutic advances in wound healing. *Journal of Dermatological Treatment*, 33(1), 2-22.
- [13] Fadilah, N. I. M., Phang, S. J., Kamaruzaman, N., Salleh, A., Zawani, M., Sanyal, A., Maarof, M., & Fauzi, M. B. (2023). Antioxidant biomaterials in cutaneous wound healing and tissue regeneration: A critical review. *Antioxidants*, 12(4), 787.

- [14] Kahraman, A., Onder, M., & Ceyhan, E. (2012). The importance of bioconservation and biodiversity in Turkey. *International Journal of Bioscience, Biochemistry and Bioinformatics*, 2(2), 95.
- [15] Jaradat, N. A. (2005). Medical plants utilized in Palestinian folk medicine for treatment of diabetes mellitus and cardiac diseases. *Al-Aqsa University Journal (Natural Sciences Series)*, 9(1), 1-28.
- [16] Lazreg Aref, H., Gaaliche, B., Fekih, A., Mars, M., Aouni, M., Pierre Chaumon, J., & Said, K. (2011). In vitro cytotoxic and antiviral activities of *Ficus carica* latex extracts. *Natural Product Research*, 25(3), 310-319.
- [17] Chawla, A., Kaur, R., & Sharma, A.K., (2012). *Ficus carica* Linn, a review on its pharamacognostic, phytochemical and pharmacologica aspects. *International Journal of Pharmaceutical and Phytopharmacological Research*, 1:215–232.
- [18] Badgujar, S. B., Patel, V. V., Bandivdekar, A. H., & Mahajan, R. T. (2014). Traditional uses, phytochemistry and pharmacology of *Ficus carica*: A review. *Pharmaceutical Biology*, 52(11), 1487-1503.
- [19] Palabıyık, E., Uğuz, H., Avcı, B., Sulumer, A. N., Yılmaz, B., & Aşkın, H., (2024). Bioactive component analysis of seed coat hexane extract of Ardahan (Türkiye) walnut. *Frontiers in Life Sciences and Related Technologies*, 5(2), 89-94.
- [20] Rutkowska, E., Wołejko, E., Kaczyński, P., Łuniewski, S., & Łozowicka, B. (2023). High and low temperature processing: Effective tool reducing pesticides in/on apple used in a risk assessment of dietary intake protocol. *Chemosphere*, 313, 137498.
- [21] Ferreira, I. C., Morales, P., & Barros, L. (Eds.). (2017). Wild plants, mushrooms and nuts: functional food properties and applications. John Wiley & Sons.
- [22] Teng, Y., Lan, P., White, L. V., & Banwell, M. G. (2023). The useful biological properties of sucrose esters: Opportunities for the development of new functional foods. *Critical Reviews in Food Science and Nutrition*, 64(22), 8018–8035.
- [23] Habulin, M., Šabeder, S., & Knez, Ž. (2008). Enzymatic synthesis of sugar fatty acid esters in organic solvent and in supercritical carbon dioxide and their antimicrobial activity. *The Journal of Supercritical Fluids*, 45(3), 338-345.
- [24] Holley, J. D., R. R. King, and R. P. Singh. (1987). Glandular trichomes and the resistance of *Solanum berthaultii* (PI 473340) to infection from *Phytophthora infestans*. *Canadian Journal of Plant Pathology*, 9(4), 291–294.
- [25] Zhu, J.-P., M.-Y. Liang, Y.-R. Ma, L. V. White, M. G. Banwell, Y. Teng, and P. Lan. (2022). Enzymatic synthesis of an homologous series of long-and very long-chain sucrose

esters and evaluation of their emulsifying and biological properties. *Food Hydrocolloids*, 124, 107-149.

[26] Fabre, N., P. Urizzi, J. P. Souchard, A. Fréchar, C. Claparols, I. Fourasté, and C. Moulis. (2000). An antioxidant sinapic acid ester isolated from *Iberis amara*. *Fitoterapia*, 71(4), 425–428.

[27] Hu, Y., H. B. Liao, G. Dai-Hong, P. Liu, Y. Y. Wang, and K. Rahman. (2010). Antidepressant-like effects of 3,6'-disinapoyl sucrose on hippocampal neuronal plasticity and neurotrophic signal pathway in chronically mild stressed rats. *Neurochemistry International*, 56 (3), 461–465.

[28] Tian, Y., W. Liu, Y. Lu, Y. Wang, X. Chen, S. Bai, Y. Zhao, T. He, F. Lao, Y. Shang, (2016). Naturally occurring cinnamic acid sugar ester derivatives. *Molecules*, 21(10), 1402.

[29] Zhao, X., Y. Cui, P. Wu, P. Zhao, Q. Zhou, Z. Zhang, Y. Wang, and X. Zhang. (2020). *Polygalae Radix*: A review of its traditional uses, phytochemistry, pharmacology, toxicology, and pharmacokinetics. *Fitoterapia*, 147, (104759).

[30] Zhou, Q., Wu, Z., Cheng, X., Zuo, Z., & Fan, C. (2024). Exploring Melezitose as a Potential Therapeutic Agent in Lung Cancer: Inhibitory Effects on Cell Proliferation and EMT-Mediated Signaling in A549 Cells. *Pharmacognosy Magazine*, 09731296241251545.

[31] Costa, J. P., Islam, M. T., Santos, P. S., Ferreira, P. B., Oliveira, G. L., Alencar, M. V., Paz, M. F., Ferreira, É. L., Feitosa, C. M., Citó, A. M., Sousa, D. P., & Melo-Cavalcante, A. A. (2016). Evaluation of Antioxidant Activity of Phytol Using Non- and Pre-Clinical Models. *Current Pharmaceutical Biotechnology*, 17(14), 1278–1284.

[32] Huang, Z. R., Lin, Y. K., & Fang, J. Y. (2009). Biological and pharmacological activities of squalene and related compounds: potential uses in cosmetic dermatology. *Molecules*, 14(1), 540-554.

[33] Nagao, K., Inoue, N., Suzuki, K., Shimizu, T., & Yanagita, T. (2021). The cholesterol metabolite cholest-5-en-3-one alleviates hyperglycemia and hyperinsulinemia in obese (db/db) mice. *Metabolites*, 12(1), 26.

[34] Ramadwa, T. E., Elgorashi, E. E., McGaw, L. J., Ahmed, A. S., & Eloff, J. N. (2017). Antimicrobial, anti-inflammatory activity and cytotoxicity of *Funtumia africana* leaf extracts, fractions and the isolated methyl ursolate. *South African Journal of Botany*, 108, 126-131.

[35] Rimbach, G., Moehring, J., Huebbe, P., & Lodge, J. K. (2010). Gene-regulatory activity of  $\alpha$ -tocopherol. *Molecules*, 15(3), 1746-1761.

[36] Reiter, E., Jiang, Q., & Christen, S. (2007). Anti-inflammatory properties of  $\alpha$ - and  $\gamma$ -tocopherol. *Molecular Aspects of Medicine*, 28(5-6), 668-691.

- [37] Huang, S. W., Hopia, A., Schwarz, K., Frankel, E. N., & German, J. B. (1996). Antioxidant activity of  $\alpha$ -tocopherol and trolox in different lipid substrates: bulk oils vs oil-in-water emulsions. *Journal of Agricultural and Food Chemistry*, 44(2), 444-452.
- [38] Ehterami, A., Salehi, M., Farzamfar, S., Samadian, H., Vaez, A., Ghorbani, S., Ai, J., Sahrapeyma, H. (2019). Chitosan/alginate hydrogels containing Alpha-tocopherol for wound healing in rat model. *Journal of Drug Delivery Science and Technology*, 51, 204-213.
- [39] Stanizzi, A., Bottoni, M., Torresetti, M., Campanati, A., & Di Benedetto, G. (2015). Topical use of  $\alpha$ -tocopherol acetate in delayed wound healing. *International Wound Journal*, 12(6), 746.
- [40] Stanizzi, A., Bottoni, M., Torresetti, M., Campanati, A., & Di Benedetto, G. (2015). Topical use of  $\alpha$ -tocopherol acetate in delayed wound healing. *International Wound Journal*, 12(6), 746.
- [41] Gupta, R., Sharma, A. K., Dobhal, M. P., Sharma, M. C., & Gupta, R. S. (2011). Antidiabetic and antioxidant potential of  $\beta$ -sitosterol in streptozotocin-induced experimental hyperglycemia. *Journal of Diabetes*, 3(1), 29-37.
- [42] Bumrela, S., & Naik, S. (2012). Hepato protective activity of methanolic extract of *Dipteracanthus patulus* (Jacq) Nees: Possible involvement of antioxidant and membrane stabilization property. *International Journal of Pharmacy and Pharmaceutical Sciences*, 4, 685-690
- [43] Valerio, M., & Awad, A. B. (2011).  $\beta$ -Sitosterol down-regulates some pro-inflammatory signal transduction pathways by increasing the activity of tyrosine phosphatase SHP-1 in J774A. 1 murine macrophages. *International Immunopharmacology*, 11(8), 1012-1017.
- [44] Palabiyik, E., Sulumer, A. N., Uguz, H., Avci, B., Askin, S., & Askin, H. (2024). Walnut fruit diaphragm ethanol extract ameliorates damage due to Triton WR-1339-induced hyperlipidemia in rats. *European Journal of Lipid Science and Technology*, 126(1), 2300105.
- [45] Abbas, M. M., Al-Rawi, N., Abbas, M. A., & Al-Khateeb, I. (2019). Naringenin potentiated  $\beta$ -sitosterol healing effect on the scratch wound assay. *Research in Parmaceutical Siences*, 14(6), 566-573.
- [46] Melo, C. M., Carvalho, K. M. M. B., de Sousa Neves, J. C., Morais, T. C., Rao, V. S., Santos, F. A., Brito, G. A. C., & Chaves, M. H. (2010).  $\alpha$ ,  $\beta$ -amyrin, a natural triterpenoid ameliorates L-arginine-induced acute pancreatitis in rats. *World Journal of Gastroenterology: WJG*, 16(34), 4272.

- [47] Pinto, S. H., Pinto, L. M. S., Guedes, M. A., Cunha, G. M. A., Chaves, M. H., Santos, F. A., & Rao, V. S. (2008). Antinoceptive effect of triterpenoid  $\alpha$ ,  $\beta$ -amyrin in rats on orofacial pain induced by formalin and capsaicin. *Phytomedicine*, 15(8), 630-634.
- [48] Abdelkader, M., Ahcen, B., Rachid, D., & Hakim, H. (2014). Phytochemical study and biological activity of sage (*Salvia officinalis* L.). *International Journal of Biological, Biomolecular, Agricultural, Food and Biotechnological Engineering*, 8(11), 1231-1235.
- [49] Oliveira, F. A., Chaves, M. H., Almeida, F. R., Lima Jr, R. C., Silva, R. M., Maia, J. L., Brito, G. A. A. C., & Rao, V. S. (2005). Protective effect of  $\alpha$ -and  $\beta$ -amyrin, a triterpene mixture from *Protium heptaphyllum* (Aubl.) March. trunk wood resin, against acetaminophen-induced liver injury in mice. *Journal of Ethnopharmacology*, 98(1-2), 103-108.
- [50] Turkoglu, M., Pekmezci, E., Kilic, S., Dundar, C., & Sevinc, H. 2017. Effect of *Ficus carica* leaf extract on the gene expression of selected factors in HaCaT cells. *Journal of Cosmetic Dermatology*, 16(4), e54-e58.
- [51] Simru, E. D. (2007). Recent advances in critical care for severely burn patients. *Turkish Journal of Medical Sciences*, 3(1), 32-35.
- [52] Tuzcu, A. K. (2019). Antioksidanların Doğal ve Takviye Şeklinde Kullanımı. *Türkiye Klinikleri Kozmetik Dermatoloji Özel Dergisi*, 12(2).