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**Research Paper / Araştırma Makalesi**

# Effect of Freeze-Drying and Oven-Drying on Volatiles, Bioactive and Structural Properties of Hawthorn (*Crataegus tanacetifolia*) Fruit

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## ABSTRACT

Hawthorn (*Crataegus*) fruit, which is traditionally used as a folk medicine, has been commonly used in pharmaceuticals due to its positive neuro- and cardio-selective effects in recent years. Drying is a practical approach for the long-term storage of hawthorn fruits due to their high annual abundance. This study aimed to compare the effects of oven-drying and freeze-drying methods on the quality, bioactivity, volatiles, and surface structure of hawthorn fruits. Total phenolic content (TPC), total flavonoid content (TFC), and antioxidant activity ($IC_{50}$) were used to compare the bioactive properties of the fresh and dried fruits. The volatiles of fresh and dried hawthorn fruits were determined by GC-MS. The TPC and TFC of freeze-dried fruit extracts were markedly higher than those of fresh and oven-dried fruit extracts ($p<0.05$). The $IC_{50}$ value of the freeze-dried fruit extract was 480 µg/mL, considerably lower than the fresh (530 µg/mL) and oven-dried fruit extracts (500 µg/mL) ($p<0.05$). Freeze-drying preserved hawthorn fruit color with the highest L* value and the lowest a* and b* color values. Many volatile compounds were identified as esters, aldehydes, hydrocarbon, terpene, etc., and their concentration decreased significantly with oven-drying, in contrast to freeze-drying. Scanning electron microscopy showed that the microstructure of freeze-dried samples was relatively homogenous and more uniform while oven-dried samples were unevenly broken pieces. Consequently, freeze-drying was determined as the best method for the dehydration of hawthorn fruits in terms of the bioactive content and natural color of fruits, and volatile preservation.

**Keywords:** Hawthorn, Freeze-drying, Oven-drying, Bioactive compounds, Volatiles

## Dondurarak ve Fırında Kurutmanın Alıç (*Crataegus tanacetifolia*) Meyvesinin Uçucu Maddeleri, Biyoaktif ve Yapısal Özellikleri Üzerine Etkisi

## ÖZ

Geleneksel olarak bir halk ilacı olarak kullanılan alıç (*Crataegus*) günümüzde pozitif nöro- ve kardiyo-selektif etkileri nedeniyle farmasötiklerde yaygın olarak kullanılmaktadır. Kurutma alıç meyvesinin yıllık bol miktarda olması nedeniyle uzun süreli depolama için uygun bir yaklaşımdır. Bu çalışmanın amacı, fırında kurutma ve dondurarak kurutmanın alıç meyvelerinin kalitesi, biyoaktivitesi, uçucu bileşenleri ve yüzey yapısı üzerindeki etkilerini karşılaştırmaktır. Taze ve kurutulmuş meyvelerin biyoaktif özelliklerini karşılaştırmak için toplam fenolik içeriği (TPC), toplam flavonoid içeriği (TFC) ve antioksidan aktiviteleri ($IC_{50}$) kullanılmıştır. Taze ve kurutulmuş alıçların uçucu madde içeriği GC-MS kullanılarak incelenmiştir. Dondurularak kurutulmuş meyve özütlerinin TPC ve TFC değerleri, taze ve fırında kurutulmuş meyvelerinkinden belirgin şekilde daha yüksek bulunmuştur ($p<0.05$). Dondurularak kurutulmuş meyve özütünün $IC_{50}$ değeri 480 µg/mL olup, taze (530 µg/mL) ve fırında kurutulmuş (500 µg/mL) meyve özütünden oldukça düşük bulunmuştur ($p<0.05$). Dondurarak kurutma en yüksek L* değeri ve en düşük a* ve b* değerleri ile alıç rengini koruyabilmiştir. Birçok uçucu bileşik ester, aldehit, hidrokarbon, terpen vb. olarak tanımlanmış ve bunların

konsantrasyonu, dondurarak kurutmanın aksine fırında kurutma yöntemiyle önemli ölçüde azalmıştır. Taramalı elektron mikroskobu, dondurularak kurutulmuş numunelerin mikro yapısının nispeten homojen ve daha düzgün olduğunu, fırında kurutulmuş numunelerin ise düzensiz kırılmış parçalar olduğunu gösterdi. Sonuç olarak, alıçta biyoaktif içerik, doğal renk ve uçucu koruma açısından en iyi dehidrasyon yöntemi dondurarak kurutma olmuştur.

**Anahtar Kelimeler:** Alıç, Dondurarak kurutma, Fırında kurutma, Biyoaktif bileşikler, Uçucu maddeler

## INTRODUCTION

Hawthorn fruit, also known as haw berries or haws, are the small, red or yellow berries that grow on the hawthorn tree. Hawthorn, which belongs to the Rosaceae family's Crataegus genus, has 1000 species and has been cultured for 1700 years [1]. Hawthorn fruit is native to Europe, Asia, and North America but is now widely cultivated in many other parts of the world. It is a hardy and adaptable plant that can grow in various soil types and climates. Different species of hawthorn fruit may have additional environmental requirements, and not all species will grow well in all regions.

Hawthorn trees' leaves, blossoms, and berries have been traditionally used in medicine and consumed by people due to their anti-inflammatory, antibacterial, anti-proliferative, and mutagenic effects [2]. Hawthorn fruit has traditionally been used to treat heart diseases such as hypertension and heart problems. Researchers have proved their effectiveness in reducing blood cholesterol and the risk of cardiovascular illnesses [3]. However, more research is needed to understand its effects fully. The popularity of hawthorn is increasing, and utilizing methods such as vinegar, pomace are started to take attention of consumers. However, because hawthorn species and variants are not authorized in Turkey, its production is confined to trees that grow naturally in the wild [1].

Although there is a large amount of hawthorn in Turkey, the ripe fruit is not harvested due to the lack of usage area, and it dries up and disappears on the branch [4]. Harvested fruits and vegetables can be consumed fresh, as they are perishable foods, and can be processed in different ways and offered for consumption by increasing their shelf life. Drying fruits and vegetables inhibits microbiological, enzymatic activity and quality deterioration. Because dried material has less water activity, physical and chemical changes are minimized during storage, increasing shelf-life [5]. Drying can be done in a few different ways, such as using a traditional (sun drying, oven drying etc.) and modern techniques (freeze-drying etc.). The general use of hawthorn is jams, jellies, and vinegar production. Drying hawthorn fruit is a way to preserve the fruit for later use. For this reason, this study aims to expand the usage of hawthorn fruit and increase its consumption. By this way, drying hawthorn fruit can improve its economic value.

Obtaining high-quality dried products is one of the most important concerns of commercial manufacturers of dried fruits. The process of drying hawthorn fruits can impact their quality. Nowadays, the most common method for drying fruits is sun-drying. In this scenario, drying takes an extended period of time, and the fruits are exposed to environmental contaminants such as dust, mice, birds, and bacteria. Hence, the quality of the dried items may be seriously reduced. Hot-air dryers offer an alternative to open-air sun-drying and can contribute to the solution of these issues. Freeze-drying, a modern sublimation-based drying technique, has become an important technology. It creates a high-quality, shelf stable product with a distinctive texture [6]. Because there are so many hawthorn fruits that need to be dried yearly and the drying process is not affected by weather, hot-air drying is a good way to dry them [7].

This study aimed to analyze the impact of two drying techniques (oven and freeze-drying) on hawthorn fruits' qualitative characteristics, bioactive properties, and volatile profile. Scanning electron microscopy was used to evaluate the effect of the drying technique on fruit structure. Total phenolic content, total flavonoid content, and antioxidant activity were used to compare the nutritional content of the fresh and dried fruits.

## MATERIALS and METHODS

Mature hawthorn fruits (*Crataegus tanacetifolia*) were collected from different trees in Acaroba village near Gaziantep, Turkey (Figure 1), in September 2021. Fruits were taken to Gaziantep University and kept at 4°C until used. Fresh fruits were cut into small pieces (1×1 cm), and analyzed immediately. Analytical solvents, Folin–Ciocalteau reagent, 2,2-diphenyl-1-picrylhydrazyl (DPPH), sodium carbonate were supplied from Merck (Darmstadt, Germany). The chemical standards were purchased from Sigma-Aldrich (St. Louis, MO, USA).

### Drying Methods

Fresh fruits were cut and frozen at -78°C in a freezer (New Brunswick Scientific, U570 Premium, England) for 24 h before freeze drying. A freeze drier (alpha 1–4 LD plus, Christ, Osterode am Harz, Germany) was used to dry the frozen samples during 48 h. The freeze-drying process was set up to work at -58°C for the freezing step, 10°C for the drying step, and 10 Pa for the pressure in the chamber during the drying step. The dried fruits were ground in a blender (Waring, Model HGB55E, Torrington, USA) and sieved through a 250-mesh sieve. The products were kept in a freezer at -20°C before usage. The fruits were put on a plate and dried at 60°C (Japan Synthetic Rubber Co., Ltd, JSOF 100, Japan) for oven drying.


Figure 1. Acaroba village, Gaziantep, Turkey. The map was illustrated using the free version of Scribblemaps.

## Extraction

Extraction was performed using a modified method suggested by Bushra et al. [8]. The fresh, freeze-dried, and oven-dried samples (1 g) were extracted using 30 mL methanol: water (80:20) for 90 min at 25°C in a rotary incubator (New Brunswick Scientific, Nova 40, Edison, NJ, USA). The residues were centrifuged by a centrifuge (Eppendorf, 5810 R, Vienna, Austria) for 10 min at 4°C and 6000 rpm. The supernatants were utilized in the further experiments.

## Total Phenolic Compounds (TPC)

Folin-Ciocalteau colorimetric method was modified to quantify total phenolic compounds [9]. 200 µL extract solution and 1 mL Folin Ciocalteu's reagent were combined in a test tube. The solution was vortexed with 0.8 mL of 7.5% $Na_2CO_3$ after 3 min. Then, the vortexed mixture was kept under darkness for 30 min. The absorbance of extracts was measured at 765 nm using UV/Vis spectrophotometer (Lambda 25, PerkinElmer, Connecticut, USA). A calibration curve was plotted using different concentrations of gallic acid (12.5-400 mg/mL). The concentration of total phenolic compounds was indicated as milligrams of gallic acid equivalent per hundred grams of dried weight (mg GAE/100 g DW).

## Antioxidant Activity

The antioxidant activity of the fruits was determined using DPPH based on a method suggested by Roesler et al. [10]. Each aliquot (0.5 mL) of extract solution was prepared in methanol at different concentrations mixed with 3.5 mL of 0.2 mM DPPH methanolic solution. The tubes were firmly shaken and left for 30 min at 25°C in the dark. UV/Vis spectrophotometer (Lambda 25, Perkin Elmer, Connecticut, USA) measured sample absorbance at 517 nm. A control measurement was done with methanol. For each different concentration, the percent reduction in absorbance was noted. The percent quenching of DPPH was then determined from the percent reduction in absorbance of the radical. In order to determine radical scavenging, we used the following formula and reported it as a percentage called the inhibitory percentage:

$$Inhibition\ (\%) = \frac{A_c - A_s}{A_c} \cdot 100 \qquad (1)$$

where $A_c$ and $A_s$ are the absorbances at 517 nm of the control and sample, respectively. The sample concentration required to scavenge 50% of DPPH radicals, $IC_{50}$ (half-maximal inhibitory concentration), was used to demonstrate DPPH radical-scavenging activity. This was found by doing a linear regression analysis between the extract's concentration and the inhibition percentage. The $IC_{50}$ values are given as the concentration of the extract, which is evaluated in µg/mL of dried extracts necessary to reduce the initial concentration of DPPH by 50%.

## Total Flavonoid Content

The total flavonoid content was analyzed using the method Chang et al. reported [11] with some modifications. Each 2.0 mL diluted extract was added to 0.1 mL 10% (w/v) aluminum chloride and mixed. Then 0.1 mL of 0.1mM potassium acetate was added and diluted to 4 mL using water. The absorbance of each mixture was measured at 415 nm after being incubated at room temperature for 30 min, using a spectrophotometer Lambda 25 UV/Vis spectrophotometers (Perkin Elmer, Connecticut, USA). Varying quantities of quercetin ranging from 0.02 to 1 mg/mL were employed to make a calibration curve. The findings were expressed as the amount of quercetin present in one milligram for every one hundred grams of dried weight (mg quercetin/100 g DW).

## Color

Color measurements of about 25 g of crushed fruit samples were taken using a HunterLab ColorFlex (A60-1010-615 Model Colorimeter, HunterLab, Reston VA) in

accordance with the International Commission on Illumination (CIE) methodology. The measurements were obtained at three distinct time points from a transparent glass sample cup and subsequently averaged. The instrument was calibrated against black and white standard tiles (L*= 93.41, a* =-1.12, b* =1.07). L* = lightness, +a* = redness, -a* = greenness, +b* = yellowness, and -b* = blueness are the parameters measured. The following equation was used to calculate the total color differences (ΔE):

$$\Delta E=[(L_f^*-L_0^*)^2 + (a_f^*-a_0^*)^2 + (b_f^*-b_0^*)^2]^{1/2} \quad (2)$$

where the subindices o and f represent the values for the initial (fresh fruit) and final (dehydrated fruit) samples [12].

### Volatile Compositions by Gas Chromatography

Volatile compounds in fresh, freeze-dried, and oven-dried hawthorn fruit were analyzed using solid phase microextraction (SPME). SPME fiber was coated with 50/30 µm Carboxen/ Divinylbenzene/ Polydimethyl-siloxane (CAR/DVB/PDMS). The fibers were inserted into the headspace of a 20 mL vial that contained 3 grams of the sample and 5 µL of the internal standard (4-nonanol in EtOH, 8.3 g 100 $mL^{-1}$). Supelco (Bellefonte, Pennsylvania, United States) supplied the fiber, which was conditioned in accordance with the manufacturer's specifications by remaining in the GC injector. The SPME working settings included an extraction temperature of 60°C for 15 min, a rotational speed of 250 rpm, an agitation period of 1 s, and a desorption temperature of 250°C for 5 min. Analyses were carried out on an Agilent 7890B gas chromatograph that was outfitted with an Agilent 7010B network mass selective detector (EI) (electron energy = 70eV). The mass range that was analyzed was 20-550 amu. A DB-Wax column with dimensions of 60 m x 0.25 mm and a thickness of 0.25 m was used by J&W Scientific of Folsom, California, in order to separate volatile compounds. The following working conditions were established: the injector and FID temperatures were set to 270 and 280°C, respectively. High-purity helium (99.999%) was employed as the carrier gas during the splitless injection mode that lasted for 0.5 min. Carrier gas (He) flow was maintained at 1.5 mL $min^{-1,}$ and the column oven temperature was maintained at 50°C for 5 min before being elevated to 200°C at a rate of 5°C $min^{-1}$ and then held for 5 min. The temperature at the MS interface was 250 °C, whereas the ion source temperature was 180°C. The mass spectra were obtained by scanning rapidly at 2.0 scans $s^{-1}$ in the electron ionization (EI) mode with an energy setting of 70 eV. The 30–300 m/z range was covered. The mass spectra of the compounds were compared to those in the Wiley 9, National Institute of Standards and Technology (NIST) 14, and an in-house library of alkane standards to determine which compounds they were. The amounts of volatile compounds were then determined using the internal standard method with 4-nonanol (43.3 g/kg). The area of the peak ratio was corrected using response factors for each component, which were calculated using the intensity ratio of each compound to 4-nonanol [13-14].

### Scanning Electron Microscopy

The structural and morphological properties of oven- and freeze-dried fruit were examined using SEM (Quanta Field Emission 650, FEI, USA). Prior to SEM analysis, the freeze- and oven-dried hawthorn fruit (100 g) were processed into powder using stainless steel electronic grinder. Samples were fixed and coated with gold using a coater (Q150R Plus Sputter Coater Combined System, Quorum Technologies, UK). The surface morphology images were photographed in a high vacuum at 15 kV and 300X, 1000X and 5000X magnification [15].

### Statistical Analysis

SPSS 22.0 software (SPSS Inc., Chicago, IL, USA) was used to conduct all of the statistical analyses, and Duncan's Multiple Range Test was employed as the post-hoc test. The results of the data analysis were reported as the mean value standard deviation of three independent determinations. A significant difference was determined to exist when the p-value was less than 0.05 ($\alpha = 0.05$).

## RESULTS and DISCUSSION

### Color

The effects of drying methods on the color values of dried hawthorn fruits compared to fresh ones are given in Table 1. The fresh hawthorn's L*, a*, and b* values were 71.72, 11.46, and 47.36, respectively. The drying processes considerably impacted the color parameters of the samples. The decrease of the L* value in oven drying led to dried fruits having a dark color compared to fresh samples (Figure 2). The unavoidable Maillard and non-enzymatic browning events during drying procedures can explain the decrease in L* value of dried samples [16].

Meanwhile, freeze-dried samples presented significantly higher L* values than those obtained from oven-dried and fresh samples ($p<0.05$). This was attributed to the freeze-dried sample's shallow moisture content and well-preserved porosity, which caused greater light scattering [17]. When looking at the fresh sample, freeze and oven-dried samples had lower a* and b* values. The decrease in b* showed the loss of yellowness in fruit. Deterioration of color pigments was typically reflected in a reduction in a* and b* values [5]. The freeze-dried samples exhibited higher total color changes (ΔE) compared to the oven-dried samples, mostly attributed to the significantly elevated L* value observed in the freeze-dried sample. Baeghbali et al. [18] observed similar result for freeze- dried okra slices as compared to oven-dried samples.

Table 1. Bioactive and color properties of fresh and dried hawthorns

| Properties | Samples[1] | | |
|---|---|---|---|
| | FH[2] | FDH | ODH |
| Color | | | |
| L* | 71.72±0.06[b] | 85.97±1.10[a] | 63.03±0.10[c] |
| a* | 11.46±0.02[c] | 2.46±0.29[a] | 8.35±047[b] |
| b* | 47.36±0.04[c] | 23.00±0.27[a] | 33.01±0.27[b] |
| ΔE | - | 29.62±0.18a | 17.06±0.24b |
| TPC (mg gallic acid/100 g DW) | 950.02±28.03[a] | 1173.92±25.23[b] | 992.85±32.52[a] |
| TFC (mg quercetin eq/100 g DW) | 330.36±0.57[a] | 332.04±0.85[b] | 331.95±0.57[c] |
| TFC/TPC ratio | 0.34 | 0.28 | 0.33 |
| $IC_{50}$ (µg/mL) | 530.01±2.88 | 480.01±5.57 | 500.12±51.30 |

[1]FH: Fresh hawthorn, FDH: Freeze-dried hawthorn, ODH: Oven-dried hawthorn. [2]Different letters in the same row indicate significant differences ($p < 0.05$). Data are presented as means ±SD (Standard deviation) (n=3).


Figure 2. Photograph of hawthorn fruits; fresh (A), freeze-dried (B), oven-dried (C)

## Total Phenolic Content

The effects of freeze- and oven-drying on total phenolic content were investigated and compared to fresh samples. According to the TPC results gained from the spectrophotometric method, freeze-dried hawthorn fruit exhibited a higher concentration of total phenolics (1173.2 g GA/ 100 g DW) than oven-dried fruit (992.85 g GA/ 100 g DW) and fresh fruit (950.02 g GA/ 100 g DW) ($p<0.05$). Similarly, Asami et al. [19] , Chan et al. [20] and Garcia et al. [21] found that phenolic content was lost less in the freeze-drying process than in the oven-drying process. Asami et al. [19] explained the reason for higher TPC content in the freeze-dried sample compared to fresh and oven-dried samples as the high extraction efficiency because ice crystals formed within the plant matrix can rupture cell structure, allowing cellular components to exit and solvent access, resulting in better extraction. The TPC of oven-dried showed a higher TPC than that of the fresh sample, but this result was statistically non-significant ($p>0.05$). The reason may be explained by the phenomenon that exposure to high temperatures might cause the development of new phenolic compounds (low molecular weight) [22]. Hawthorn fruits exhibited considerably higher TPC compared to the reported TPC of fruits such as apples, strawberries, mulberry, apricot, and plum, which were also extracted using a methanol: water (80:20) solvent mixture as in our study [8].

## Total Flavonoid Content

The total flavonoid contents (TFC) of fresh and dried fruit samples are given in Table 1. Similar to the TPC results, freeze-dried fruit had the highest TFC (332.04 mg QE/ 100 g DW), followed by oven-dried (331.95 mg QE/ 100 g DW) and fresh fruit (330.36 mg QE/ 100 g DW). It is clear from the data that there was a significant response regarding the flavonoid content in relation to the drying method. The samples that were dried in an oven with hot air showed a lower TFC than freeze-dried samples. According to Hirsch et al. [23], the loss of flavonoid content is caused by the activation of oxidative enzymes such as polyphenol oxidase during the hot air oven drying process. The polyphenol oxidase enzyme's activity was reduced during the freeze-drying process since it was carried out at a lower temperature [24].

The flavonoid-phenolic ratio showed that flavonoids comprise a high proportion of the phenolics. These results showed the importance of flavonoids in the total phenolic content of hawthorn fruits. Similarly, high flavonoid/phenolic ratios were noticed for different fruits such as berries, grapes, and pears in literature [25].

## Antioxidant Activity

DPPH technique was applied to compare the antioxidant activity of hawthorn fruit extracts dried by different methods. Comparisons in antioxidant assays were done using $IC_{50}$ values, which are the concentrations of sample extract required to prevent 50% of DPPH scavenging. As a result, a lower $IC_{50}$ value indicates that the samples have higher radical scavenging activity [26]. As shown in Table 1, freeze-dried samples recorded the lowest $IC_{50}$ value (480 µg/mL) with a significant difference compared with the fresh sample (530 µg/mL), and oven-dried sample (500 µg/mL) ($p<0.05$). Thus,

freeze-dried samples showed the highest antioxidant capacity due to their ability to scavenge 50% free radical activity at the lowest concentration. Samples that were dried in an oven had the second-highest antioxidant capacity. Therefore, freeze-drying favors the antioxidant activity of hawthorn fruits, suggesting its prospective application as a drying technique.

## Volatile Composition

The major volatile components of fresh, freeze-dried, and oven-dried hawthorn fruits were given in Table 2. All samples' dominant volatile group was esters, followed by alcohols, aldehydes, and terpenes. Table 2 showed (E)-2-hexenal, ethyl hexanoate, hexyl acetate, cis-3-hexenyl acetate, (Z)-2-hexenyl acetate, 1-hexanol, cis-2-hexenol, hexyl butyrate, trans-2-hexenyl butyrate, and hexyl hexanoate were the most abundant volatiles in fresh hawthorn fruit. Hexyl acetate had the highest amount of these compounds, measuring 999.25 µg/kg. Similarly, Horvat et al. [27] reported that hexanal, butyl acetate, (E)-2-hexenal, butyl butyrate, linalool, butyl hexanoate, methyl octanoate, pentyl hexanoate, and hexyl hexanoate were nine major components in raw hawthorn fruit.

When compared to fresh hawthorn fruit, both drying processes caused alterations in the volatile profile and amount. In the case of freeze-dried hawthorn fruit, the main volatile compounds detected were butyl acetate, ethyl hexanoate, hexyl acetate, cis-3-hexenyl acetate, (z)-2-hexenyl acetate, 1-hexanol, hexyl butyrate, hexyl hexanoate, d-limonene, and styrene (ethenyl benzene) (Table 2). Results showed an increase in esters like butyl acetate, hexyl butyrate, hexyl hexanoate, and d-limonene and the formation of styrene (ethenyl benzene) in the freeze-dried fruit.

The major volatile compounds in the oven-dried hawthorn fruit include butyl acetate, hexyl acetate, 1-hexanol, hexyl butyrate, d-limonene, styrene (ethenyl benzene), dodecane, furfural/3-furaldehyde, and dihydro-3-methylene-5-methyl-2-furanone. For the oven-dried fruits, a loss of compounds such as (e)-2-hexenal, ethyl hexanoate, cis-3-hexenyl acetate, (z)-2-hexenyl acetate, trans-2-hexenyl butyrate, hexyl hexanoate was observed. The formation of dodecane, furfural/3-furaldehyde, dihydro-3-methylene-5-methyl-2-furanone, and the increase of d-limonene and styrene (ethenyl benzene) was also observed. Furfural obtained only in oven-drying is one of the main byproducts of ascorbic acid, and it is partially responsible for why fruit turns brown [28].

## Microstructure

The information on microstructure changes is critical for better process control and appearance improvement. Microstructure analysis of the dried fruit surface was performed to investigate the effects of the drying method on the microstructure changes of the samples. Figure 3 shows the morphology and structure of oven-dried and freeze-dried hawthorn samples as observed under a scanning electron microscope at different magnifications. The particles of oven-dried and freeze-dried fruits significantly differed in structure, reflecting the SEM results. Freeze-dried fruits developed a porous or honeycomb-like structure. The microstructure of freeze-dried fruits were relatively homogenous, clear and more uniform (Figure 3A-C) The oven-dried samples had unevenly broken pieces or flake-like structure (Figure 3D-F). The oven-dried sample shrank more than the freeze-dried sample due to the excessive microstructural stress generated by high moisture gradients inside the product.

The porous structure is caused by ice sublimation in the freeze-dried fruits, and voids are likely formed due to the sublimation process [29]. Higher porosity in food, on the other hand, means that it has more surface area. Thus, it has a shorter shelf life because the surface is exposed and a higher rehydration ratio since it has more open pores that act as capillaries to take in water [30]. Therefore, it is vital to regulate the drying process to produce the desired porosity and meet the intended objectives.

Table 2. The composition of major volatile compound in fruits of hawthorn for different drying techniques

| Compound | Chemical Family | Concentration (µg/kg)* | | |
|---|---|---|---|---|
| | | FH[1] | FDH | ODH |
| Butyl Acetate | Ester | 119.37±2.2$^{a}$ | 345.60±5.5$^{b}$ | 77.77±0.8$^{b}$ |
| Ethyl Hexanoate | Ester | 183.37±1.1$^{a}$ | 55.16±0.3$^{b}$ | nd |
| Hexyl Acetate | Ester | 999.25±28$^{a}$ | 599.52±10.8$^{b}$ | 57.79±0.1$^{c}$ |
| Cis-3-Hexenyl Acetate | Ester | 583.07±14$^{a}$ | 202.10±2.8$^{b}$ | nd |
| (Z)-2-Hexenyl Acetate | Ester | 408.54±6.8$^{a}$ | 55.07±0.3$^{b}$ | nd |
| Hexyl Butyrate | Ester | 246.72±3.8$^{a}$ | 358.86±5.7$^{b}$ | 16.71±0.1$^{c}$ |
| Trans-2-Hexenyl Butyrate | Ester | 85.08±1.2 | nd | nd |
| Hexyl Hexanoate | Ester | 153.91±2.3$^{a}$ | 166.36±1.6$^{b}$ | nd |
| 1-Hexanol | Alcohol | 159.80±2.5$^{a}$ | 119.80±1.9$^{b}$ | 47.26±0.2$^{c}$ |
| Cis-2-Hexenol | Alcohol | 104.97±1.5 | nd | nd |
| (E)-2-Hexenal | Aldehyde | 103.55±5.0 | nd | nd |
| Furfural/3-Furaldehyde | Aldehyde | nd[2] | nd | 465.36±10 |
| Styrene (Ethenyl Benzene) | Hydrocarbon | nd | 156.31±2.2$^{a}$ | 522.98±6.5$^{b}$ |
| Dodecane | Hydrocarbon | nd | nd | 117.17±0.7 |
| D-Limonene | Terpene | 16.56±0.1$^{a}$ | 61.91±0.1$^{b}$ | 443.84±5.2$^{c}$ |
| Dihydro-3-Methylene-5-Methyl-2-Furanone | Furan | nd | nd | 81.02±3.2 |

*Different letters in the same row indicate significant differences ($p < 0.05$). Data are presented as means ±SD (Standard deviation) (n=3). [1]FH: Fresh Hawthorn, FDH: Freeze-dried hawthorn, ODH: Oven-dried hawthorn, [2]not detected

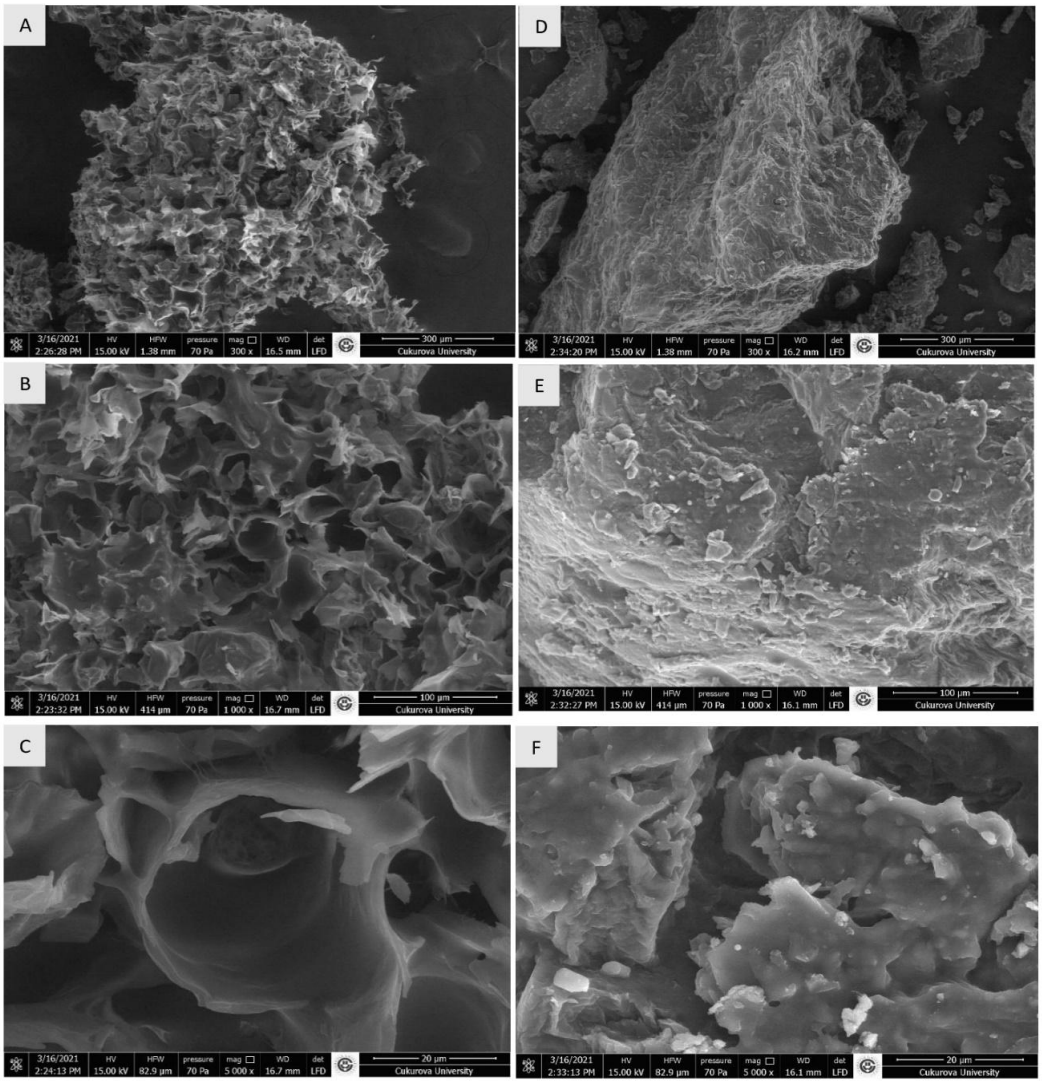


Figure 3. Scanning electron microscope images of freeze-dried hawthorn fruits (A) 300 X magnification (B) 1000 X and (C) 5000 X magnification and oven-dried hawthorn fruits (D) 300 X magnification (E) 1000 X and (F) 5000 X magnification

## CONCLUSION

Various drying techniques exist within the food industry. Drying techniques using heat are undesirable due to the detrimental impact on the bioactive characteristics of the dried sample. In this study, freeze-drying was superior to oven-drying, being rich in total phenolics and flavonoids and having a high antioxidant capacity. Freeze-drying preserved the hawthorn color with the highest L* value and the lowest a* and b* values. Esters and aldehydes were the most common group of compounds susceptible to giving flavor. Freeze-dried samples had more volatile compounds than those that were oven-dried. As a result, freeze-drying can be a promising option for drying hawthorn to retain the maximum quantity of naturally existing bioactive components, antioxidant activity, volatiles, and color. The results of this study hold significant practical significance for various industries, including those associated with food production, pharmaceuticals, traditional medicine, and agriculture. The results provided that freeze-drying was the most effective drying technique for hawthorn fruit to preserve its bioactive characteristics, color, and flavor, enhancing its value for producers and consumers across several industries.

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