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Investigation of Some Biological Properties of Propolis in Hawthorn Vinegar Extract

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Propolis, Hawthorn Vinegar, Antioxidant, Phenolic, Flavonoid

Abstract: With the determination of the biological activity of different components in the chemistry of propolis, its importance has increased day by day and its use in the field of integrative medicine has become widespread. Propolis is not used in its crude form due to its physical properties, it has to be extracted. The type of solvent used in the extraction process is quite important for the efficiency of the biological activity of propolis. Solvents commonly used in propolis extraction; are water, ethanol, and methanol. Besides being quite easy to extract propolis components using ethanol; extraction of propolis with ethanol creates a usage limit for children, pregnant women, and Muslim people that don't use alcohol. The limited use of ethanol and the fact that it has some harm to health have led researchers to search for different types of solvents. As a result of this research, vegetable oils, and various kinds of vinegar have started to be in the literature as alternative solvents. In our study, the vinegar of hawthorn fruit, which is rich in flavonoids and has been used in traditional medicine for many years, was used in the extraction of propolis. The solubility of propolis in hawthorn vinegar was investigated in terms of its physicochemical properties (pH, titratable acidity, brix, and color) and bioactive properties (phenolic, flavonoid contents, and antioxidant activity). As a result of the investigation, it was observed that the addition of propolis to hawthorn vinegar caused changes in the physicochemical properties of vinegar at an acceptable level for the consumer and increased its bioactive properties. It is thought that propolis-added hawthorn vinegar will provide functionality in dishes where vinegar is used today, in salads, and even in foodstuffs such as brine and pickles.

Propolisin Alıç Sirkesi Ekstraktında Bazı Biyolojik Özelliklerinin İncelenmesi

Anahtar Kelimeler
Propolis, Alıç Sirkesi, Antioksidan, Fenolik, Flavonoid

Öz: Propolisin yapısındaki farklı bileşenlerin biyolojik aktivitesinin belirlenmesiyle, önemi gün geçtikçe artmış ve tamamlayıcı tıp alanında kullanımı yaygınlaşmıştır. Propolis fiziksel özelliklerinden dolayı ham halde kullanılamamakta, ekstraksiyon işleminden geçirilmesi gerekmektedir. Ekstraksiyon işleminde kullanılan çözücü tipi, propolisin biyolojik aktivitesinin etkinliği açısından oldukça önemlidir. Propolis ekstraksiyonunda yaygın olarak kullanılan çözücüler; su, etanol ve metanol olarak gösterilebilir. Etanol kullanarak propolis bileşenlerinin ekstraksiyonu oldukça kolay olmasının yanı sıra; propolisin etanol ile ekstraksiyonu çocuklar, hamileler ve alkol kullanmayan müslümanlar için kullanım sınırı oluşturmaktadır. Etanolün sınırlı dozda kullanımı ve sağlığa bazı zararlarının olması, araştırmacıları farklı çözücü tipleri aramaya yöneltmiştir. Bu araştırmalar sonucunda alternatif çözücüler olarak, bitkisel yağlar ve çeşitli sirkeler literatürde yer almaya başlamıştır. Çalışmamızda propolisin ekstraksiyonunda, flavonoidler bakımından zengin ve özellikle geleneksel tıp alanında uzun yıllardır kullanılan bir meyve olan alıç meyvesinin sirkesi kullanılmıştır. Alıç sirkesinde propolisin çözünürlüğü, fizikokimyasal özellikler

(pH, titrasyon asitliği, ºbriks ve renk) ve biyoaktif özellikler (fenolik ve flavonoid içerikleri ile antioksidan aktivite) açısından araştırılmıştır. Araştırma sonucunda alıç sirkesine propolis ilavesinin sirkenin fizikokimyasal özelliklerinde tüketici için kabul edilebilir düzeyde değişikliğe neden olduğu ve biyoaktif özelliklerini arttırdığı gözlemlendi. Propolis katkılı alıç sirkesinin; günümüzde sirkenin kullanıldığı yemeklerde, salata ve hatta salamura, turşu gibi gıda maddelerinin yapımında fonksiyonellik sağlayacağı düşünülmektedir.

1. INTRODUCTION

Propolis is a bee product in colors ranging from light yellow to dark brown, formed by the honey bee (Apis mellifera) combining resinous substances collected from different trees and plants with pollen, wax and its own sap [1]. Bees use propolis as an antiseptic against microbial infections and intruders that may occur in the hive. The chemical composition of propolis varies according to the collected flora, season and geographical region [2]. Crude propolis contains more than 300 components, and more than 180 compounds, most of which are polyphenols, have been identified as components of propolis. The main polyphenols in the content of propolis are phenolic acid and its esters, flavonoids, phenolic aldehydes, and ketone groups [3, 4]. It is not possible to use crude propolis directly due to the high amount of waxy substances it contains, the low proportion of the bioactive composition, and the pollution it contains [5]. Therefore propolis has to be extracted before consumption. With the extraction process, the phenolic components of propolis appear, and increases in antioxidant and antimicrobial activities occur. The most preferred solvent in the extraction process is ethanol, as it contains a large dipole moment in propolis content [6]. Apart from ethanol, other solvents such as water, chloroform [7], dimethylsulfoxide, methanol, ether, acetone [2], dichloromethane [8], ethyl acetate [9], and propylene glycol [10] can also be used in propolis extraction. Although the use of ethanol in propolis extraction is a simple and effective method, the intensity of the smell of alcohol combined with the smell of propolis limits its use in the cosmetics and pharmaceutical industries, as well as its use in pregnant women and children. For consumers with halal-haram sensitivity, the use of ethanol as a solvent in the extraction of propolis poses a problem [6]. Therefore, there is a need for a non-alcohol-based an effective extraction technique.

Vinegar is a special fermentation product produced by yeasts and acetic acid bacteria, which can be obtained from different raw materials that have fermentable carbon sources [11,12]. Due to the many phenolic compounds, amino acids, vitamins, organic acids, and melanoidin, it contains vinegar has positive effects on human health with its antioxidant, antimicrobial, anticarcinogenic, and antidiabetic properties [13]. Vinegar, which is among the fermented products, according to the raw materials used in its production; wine vinegar, fruit vinegar, fruit wine vinegar, cider vinegar, alcohol vinegar, grain vinegar, malt vinegar, flavored vinegar, and other vinegar are listed as [14].
Our country is very rich in terms of wild fruit species diversity, and one of these wild fruit varieties is hawthorn (*Crataegus* spp. L.). The Hawthorn genus is distributed in the northern hemisphere with 200 species, and in Turkey with 17 different species and many taxa that grow naturally [15,16]. The main components of hawthorn fruit, which is generally consumed fresh, are flavonoids, triterpene acids, proanthocyanidins, organic acids, and some amines [17,18]. In addition to these, hawthorn fruit is also very rich in phenolic substances such as apigenin, quercetin, chlorogenic acid, gallic acid, vitexin, coumaric acid, caffeic acid, naringenin and cratenacin [19]. As for hawthorn vinegar, it is a local product obtained from the fruits collected from the hawthorn tree and is among the lesser-known vinegar in Turkey because it is produced on a local scale.

The number of propolis extracts that don't contain ethyl alcohol is increasing rapidly in the market. These are aqueous extract, herbal oil extracts (especially olive oil), essential oil extracts, extracts of multiple alcohols (glycerol, glycol, etc.) and less frequently are used, soaking in beverages, solving in products such as vinegar, brewing in different liquid mixtures and combinations of all these [5].

In our study, hawthorn vinegar obtained from hawthorn that a fruit frequently consumed in the Bingol region was used in propolis extraction. It is thought that the use of propolis in vinegar extraction will increase its use in future studies, by obtaining a product with increased nutritional value.

2. MATERIAL AND METHOD

2.1. Materials

Propolis (Bingöl Provincial Beekeepers' Association), hawthorn vinegar (Tijda Gıda Ltd. Şti), acetic acid ($C_2H_4O_2$), phenolphthalein ($C_{20}H_{14}O_4$), sodium hydroxide (NaOH), folin & ciocalteu′s phenol reagent, sodium carbonate (Na_2CO_3), gallic acid ($C_6H_2(OH)_3CO_2H$), sodium nitrite ($NaNO_2$), aluminium chloride ($AlCl_3$), quercetin ($C_{15}H_{10}O_7$), DPPH (2,2-diphenyl-1-picrylhydrazil), BHT (Butylated Hydroxytoluene), methyl alcohol (CH_3OH). The propolis sample used in this study was collected from the Bingöl region, and hawthorn vinegar was purchased from a local market in Bingöl, Türkiye. All chemicals were used in analytical purity.

2.2. Methods

2.2.1. Extraction method

The samples to be used in the study were stored at +4 °C throughout the analysis period. 2 g of powdered propolis was added to 40 mL of hawthorn vinegar (HV) and the mixture was homogenized for 1 minute with the help of a homogenizer. The homogeneous mixture was stirred for 24 hours with the help of a magnetic fish in a stirrer at 150 rpm at 60 °C. At the end of the waiting period, the mixture was centrifuged at 3000 rpm for 10 minutes and the supernatant part was filtered with roughing filter paper thus propolis added hawthorn vinegar (PHV) was made ready for use.

2.2.2. Physiochemical analysis

To determine the physicochemical properties of hawthorn vinegar (HV) and propolis added hawthorn vinegar (PHV); pH, titratable acidity, brix, conductivity, and color measurement analyses were performed.

2.2.2.1. pH and titratable acidity

pH values of HV and PHV samples were measured using a Thermo Scientific pH meter. The titratable acidity of the HV and PHV samples was calculated as acetic acid equivalents. Phenolphthalein was added to a 5 mL sample as an indicator and titrated with 0.1 N NaOH. By noting the volume of NaOH used in the titration, the percent titratable acidity was calculated in Equation (1) [20].

Titratable acidity (%) = (V × E × 100) / M (1)
V: volume of NaOH used, mL
E: 0.006005 (acid equivalent to 1 mL 0.1 N NaOH, g)
M: volume of sample, mL

2.2.2.2.Brix

The amount of water-soluble solids (Brix) was analyzed for HV and PHV using an ISO-LAB handheld refractometer. The results were given in brix.

2.2.2.3. Conductivity

Conductivity measurement was measured using Mettler Toledo brand portable conductivity meter for HV and PHV samples. Results were given in µS/cm.

2.2.2.4. Color measurement

Color measurement was analyzed for HV and PHV samples using the Konica Minolta CM-5 colorimeter. Results were given as L* (0: darkness, 100: lightness), a* (-: greenness, +: redness), and b* (-: blueness, +: yellowness).

2.2.3 Bioactive analysis

To determine the bioactive properties of hawthorn vinegar (HV) and propolis added hawthorn vinegar (PHV) samples; total phenolic content, total flavonoid content and antioxidant activity (%DPPH) analyzes were performed in 3 repetitive.

2.2.3.1. Total phenolic content and total flavonoid content

According to the method of Hajji et al., [21] total phenolic content analyzes were performed for HV and PHV samples. 500 µL HV and 500 µL PHV samples were taken into test tubes and 500 µL of Folin reagent was added to them. Then, each test tube was vortexed by adding 500 µL of Na_2CO_3 and 3500 µL of distilled water. The test tubes were kept in a dark laboratory environment for 90 minutes. At the end of the waiting period, 300 µL of samples were pipetted into 96 well plates and measured at 760 nm in a microplate reader (Spectramax Plus 384 ELISA). Results were given as gallic acid equivalents.

According to the method of Zhishen et al., [22] Total flavonoid content analyses were performed for HV and PHV samples. 250 µL of HV and 250 µL of PHV were taken into test tubes. 150 µL of $NaNO_2$ was added to them and after waiting for 6 minutes, 75 µL of $AlCl_3$ was added. Test tubes were kept for 5 minutes in a laboratory environment, and 1000 µL of NaOH and 2500 µL of distilled water were added. After each addition, the samples were vortexed. Then, 300 µL of samples were pipetted into 96 well plates and measured at 510 nm in a microplate reader (Spectramax Plus 384 ELISA). Results were given as quercetin equivalents [23].

2.2.3.2. DPPH radical scavenging activity

According to the method of Hatano et al., [24] on top of 0.3 mL of HV and PHV samples, 2.7 mL of freshly prepared DPPH solution was added to the test tubes and the samples were vortexed, kept in the dark laboratory environment for 60 minutes. At the end of the waiting period, measurements were taken in a UV-Vis spectrophotometer (Shimadzu) at an absorbance of 517 nm. As a control, distilled water was used instead of the sample. Results were given as % inhibition.

3. RESULTS

3.1. Physiochemical Properties

3.1.1. pH and titratable acidity

The pH values of the HV sample and the PHV sample were determined respectively as 2.96 and 2.98. In studies in the literature, pH values of hawthorn vinegar samples are 3.28 [25], 3.76 [26], 3.3 [20], 2.69 [27], 2.76 [28], 3.63 [29], 3.67 (red) and 3.25 (black) [30]. The results obtained in our study are similar to the literature findings.

The titratable acidity values as the acetic acid equivalent of the HV sample and the PHV sample were determined respectively as 2.58% and 2.22%. In similar studies, titratable acidity values of hawthorn vinegar samples

were as acetic acid equivalent; it has been reported as 2.2% [20], 0.82% [26], 2.29% [28], and 4.13% [29]. The titratable acidity results obtained in our study were similar to other studies. It has been stated that this parameter differs according to the production method and the raw material used [30].

3.1.2. Brix

The amount of water-soluble solids in HV and PHV samples was determined as approximately 4 brix. In similar studies, the amount of water-soluble solids in hawthorn vinegar samples; was 5.33 [25], 1.26 [26], 2.24 [27], 3.17 [28], 5.45 [29], 9.3 (red), and 15.6 (black) [30] as brix was represented by high variability. The brix values obtained in our study were in the range of brix values of hawthorn vinegar in the literature (1.26-15.6).

3.1.3. Conductivity

The conductivity values for HV and PHV samples were found respectively as 2.27 and 3.20 µS/cm. In studies in the literature, the conductivity value of hawthorn vinegar was found to be 3.86 [25], and 1.36 [27] µS/cm. The conductivity values of both the HV sample and the PHV sample in our study are similar to other studies.

3.1.4. Color measurement

The color properties of vinegar are very important in terms of consumer perception, so color measurement was performed in the samples. The color (L*, a*, b*) values of the samples used in our study are given in Table 1.

Table 1. Color measurement results of hawthorn vinegar (HV) and propolis added hawthorn vinegar (PHV)

	L*	a*	b*
HV	88.66	0.81	6.68
PHV	52.73	7.10	43.59

The "L" value ranges from 0-100 and indicates the brightness of a foodstuff. According to the results, hawthorn vinegar has a brighter appearance than propolis-added hawthorn vinegar. If the "a" value is "+", it means redness, and according to the results, the redness of propolis added to hawthorn vinegar is higher than hawthorn vinegar. If the "b" value is "+", it means yellowness, and according to the results, the yellowness of propolis added hawthorn vinegar is much higher than that of hawthorn vinegar. As a result, while the L* value of the vinegar sample decreased with the addition of propolis, the a* and b* values increased. In similar studies in the literature, the color values (L*, a*, and b*) of hawthorn vinegar samples were examined and the results are given in Table 2.

Table 2. Color measurement results of hawthorn vinegar samples

References	L*	a*	b*
[25]	31.40	20.48	40.08
[26]	18.10	1.96	10.67
[27]	27.80	1.33	-0.30
[28]	61.3	4.47	33.1

When Table 2 is examined, it has been observed that the color values of the studies made with hawthorn vinegar are in very different ranges. The L* value of the HV sample used in the study is much higher than the results obtained in similar studies. It can be said that the L* value of the PHV sample has a lower value, so its brightness has decreased. The a* value of the HV sample used in the study is lower than the results obtained in similar studies. It can be said that the a* value of the PHV sample has a higher value and is near to the studies in the literature. The b* value of the HV sample used in the study is similar to the studies in the literature. It can be said that the b* value of the PHV sample has a high value and its yellowness is more than the studies in the literature. It can be stated that the reason for this difference in the studies is due to the color of the shell of the hawthorn species [27]. In addition, it is thought that the heat treatment used in the extraction process affects the color properties of the samples.

3.2. Bioactive Properties

3.2.1. Total phenolic content and total flavonoid content

The total phenolic content (TPC) and total flavonoid content (TFC) of hawthorn vinegar (HV) and propolis added hawthorn vinegar (PHV) calculated as a result of the analyzes made in the study are given in Table 3.

Table 3. Total phenolic and total flavonoid content of hawthorn vinegar (HV) and propolis added hawthorn vinegar (PHV)

Analyzed Sample	TPC (mg GAE / L)	TFC (mg QE / L)
HV	56.76 ± 2.57	290.44 ± 4.19
PHV	73.55 ± 2.08	1364.89 ± 3.85

GAE: Gallic acid equivalence, QE: Quercetin equivalence

In a study by Kadaş et al., [25], the total phenolic content of the hawthorn vinegar sample was determined as 50.2 ± 0.23 mg GAE/L, similar to our study. It was determined that the addition of propolis to the vinegar sample increased the total phenolic content (Figure 1). When other studies in the 27ort his27e are examined, the total phenolic content values of hawthorn vinegar samples are 280 ± 1 mg GAE/L [20], 306.80 ± 5.07 mg GAE/L [26], 751.11 ± 15.71 mg GAE/L [27], 647 ± 0.115 mg GAE/L [28], and 24207 ± 0.02 mg GAE/L [29]. The reason 27ort his difference is thought to be related to the geographical region, location, climatic conditions, and soil structure where the hawthorn plant is grown [31].

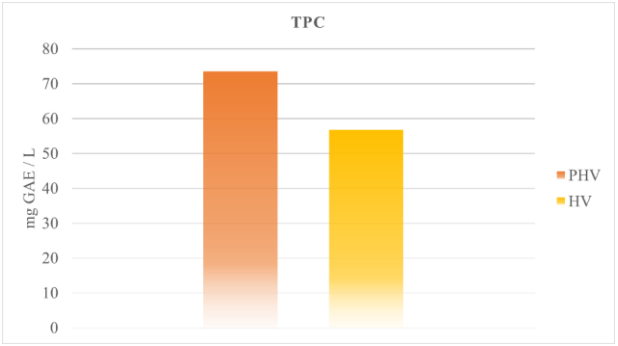

Figure 1. Total phenolic content (TPC) of hawthorn vinegar (HV) and propolis added hawthorn vinegar (PHV)

In a study assessing propolis treated by different extraction methods the propolis/solvent ratio for extraction processes was determined as 1/20 (w/v), similar to our study [32]. In the study, maceration was carried out at room temperature using ethanol (70%) and water as solvent. At the end of the first day, the total phenolic content of propolis was 856 mg GAE/L in an ethanol, and 124 mg GAE/L in water solvent. The total phenolic content of the propolis-vinegar solution in our study was approximately 40% less than the total phenolic content of the water-propolis solution used in the study. It is difficult to compare the results obtained in different solvents. However, it is thought that the use of vinegar as a solvent in propolis extraction has the potential to increase the total phenolic content of propolis with different combinations of temperature and time. This potential may become kinetic in future studies to be.

It was determined that the total flavonoid substance content of the HV sample used in our study (290.44 ± 4.19 mg QE/L) increased approximately 5 times with the addition of propolis (1364.89 ± 3.85 mg QE/L) (Figure 2). The flavonoid content analyses made with hawthorn vinegar in the literature are 50 ± 2 mg CE/L [20], 136.16 ± 0.97 mg CE/L [26], and 261 ±1.45 mg CE/L [28] as catechin equivalence. Comparing the total flavonoid content, calculated as quercetin equivalent, with results calculated with different standards, such as catechin, is quite complex. In the literature, the total flavonoid content of propolis tinctures of Ukrainian origin was evaluated and the results were given as mg QE/L, similar to our study. All propolis tinctures were prepared 1/10 (w/v) ratio and with 70% ethanol. The solutions were first kept in closed containers at 40-50°C for about 3 hours and then extracted at room temperature for 21 hours. The total flavonoid content of propolis tinctures was found to range from 4784.31 mg QE/L ± 8.80% to 6796.51 mg QE/L ± 0.52% [33]. The total flavonoid content of the ethanol-propolis solution used in the study is approximately 3.5 times the total flavonoid content of the propolis-vinegar solution in our study. It is thought that increasing the solution concentration and changing the maceration method parameters in the future experiments to be carried out will change the flavonoid substance content obtained from propolis.

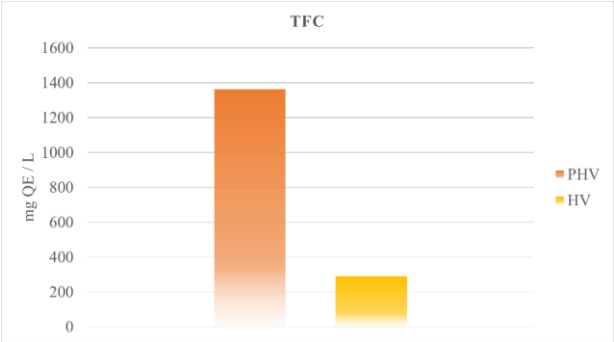

Figure 2. Total flavonoid content (TFC) of hawthorn vinegar (HV) and propolis added hawthorn vinegar (PHV)

More than 60 flavonoid substances, including reduced flavonoid types, namely catechin derivatives, as well as oxidized flavonoid types such as quercetin, rutin, and chrysin, have been isolated from hawthorn fruit [34]. However, it is known that heat treatment applications reduce the total organic acids and total flavonoid content in the samples [35]. For example, Guo et al., [36] in their study with hawthorn fruit, determined that some contents, such as catechin, decreased after 10 minutes of heating. In addition, in the same study, it was determined that heat treatment caused an increase in two main compounds, quercetin, and apigenin. In our study, it is thought that the use of heat treatment in the extraction of the samples contributes to the increase in the total flavonoid substance content, and it is thought that this contribution is largely made by the compound called quercetin. Among the active ingredients for hawthorn, especially the quercetin content increases rapidly during heat treatment and the quercetin content is approximately ten times higher in processed hawthorn than in raw hawthorn [36]; shows that this compound can be used as a chemical quality indicator in the production of propolis-added vinegar. In addition, the quercetin compound plays a role in the prevention of Alzheimer's disease, which is increasing today [37]. For this reason, the necessity of producing innovative and healthy products such as propolis-added vinegar is increasing.

3.2.2. DPPH radical scavenging activity

DPPH radical scavenging activity method was used to determine the antioxidant capacity of the samples. DPPH radical scavenging activities of HV and PHV samples were found to be respectively 53.38% ± 0.6% and 68.33% ± 0.4%. Propolis samples collected from different regions by Akbar et al., [38] were extracted with ethanol at a ratio of 1/10 (w/v) for 24 hours. After the extracts were concentrated with the help of a rotary evaporator, freeze-dried powder extract was obtained. When the DPPH free radical scavenging activities of 8 propolis samples were examined, it was observed that the values varied between 52.79% ± 1.09% and 68.39% ± 1.02%. These results show that the HV and PHV samples used in our study are the same as the DPPH radical scavenging activities. Yıldırım [32] used ethanol (70%) and water as a solvent in the extraction of propolis at room temperature by maceration. At the end of the first day, the antioxidant activity of propolis was determined as 65% in ethanol and 52% in a water solvent. The results are almost the same as the antioxidant activity results obtained with the use of vinegar as a solvent in our study. This suggests that the substitution of vinegar instead of ethanol in propolis extraction will increase day by day.

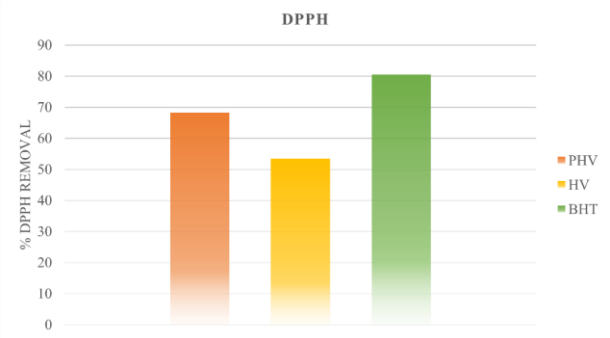

Figure 3. DPPH radical scavenging activities (%) of hawthorn vinegar (HV), propolis added hawthorn vinegar (PHV), and BHT

Only in studies with hawthorn vinegar, DPPH radical scavenging activities were 41% ±1% [20], 55.59% ± 3.86% [26], and 54.9% ± 0.399% [28] which is quite similar to our study. With the addition of propolis to hawthorn vinegar, it is clearly seen in Figure 3 that the DPPH radical scavenging activities of the samples increased and they took a value close to the synthetic antioxidant substance BHT. When Figure 3 is examined, the DPPH radical scavenging activities of the samples with a concentration of 1mg/mL and a synthetic antioxidant with the same concentration, BHT, are given as BHT>PAS>AS. The presence of some side effects of BHT, which is one of the synthetic antioxidants, has led studies to focus on antioxidants of plant origin [39]. In our study, it was determined that the PHV sample scavenged less but close amounts of DPPH radicals compared to BHT. It is thought that this situation will lead to plant-based antioxidant research.

4. DISCUSSION AND CONCLUSION

Propolis collected from trees and plants by honey bees has a very complex content and can't be used crudely. Flavonoids and phenolic compounds in the content of propolis are related to the biological activity of propolis. Since the solvents used in propolis extraction extract different components, the extraction method and solvent selection are very important. It has been concluded that it would be more beneficial to conduct research in living systems with organic solvents. For this reason, the search for different solvents continues in addition to the organic solvents that are frequently used for propolis extraction. In a study conducted for this purpose, the solubility of propolis in vegetable oils was investigated. As a result of the study, the total polyphenol content, flavonoid content, and iron-reducing power of the olive oil extract of propolis; it has been determined that propolis is richer than corn, hazelnut, and sunflower oil extracts [40]. Another type of solvent used in propolis extract besides vegetable oils is vinegar. For this purpose, another study was conducted and the solubility of propolis in various kinds of vinegar was investigated. As a result of the examination, the total polyphenol, flavonoid content, and iron-reducing power of propolis in grape vinegar extract were found to be higher than organic (pomegranate, apple), commercial (pomegranate, apple, grape) and white vinegar extracts of propolis [41].

Vinegar, which has been widely used among people for centuries; is a healthy solution formed by the oxidation of sugar-containing fruits first to ethyl alcohol and then ethyl alcohol to acetic acid. In our study, vinegar was used as a propolis solvent, and hawthorn vinegar was chosen as a vinegar type. The fact that hawthorn fruit is rich in flavonoids and has been used for many years, especially in the field of alternative medicine, played an important role in this selection. In our study, the physicochemical and bioactive properties of hawthorn vinegar (HV) and propolis-added hawthorn vinegar samples (PHV) were examined separately. The data obtained show that propolis changes the physicochemical properties of vinegar. It is also among the results of our study that the bioactive properties of the propolis-added hawthorn vinegar sample increased compared to the hawthorn vinegar sample. It is thought that the heat treatment used especially during propolis extract significantly increases the quercetin content of vinegar. It is also thought that the alcohol formed in the first step of vinegar production and the resulting acetic acid contribute to the solubility of propolis. It is inevitable that vinegar, which has important effects on human health, should be included in the daily diet with the addition of propolis. Modifying the method applied in this study and applying it to different vinegar varieties will shed light on future studies. With the increase in the use of propolis-added vinegar in salads and sauces, a functional food supplement can be used in nutrition.

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