

Investigation of the Total Antioxidant/oxidant Status, Total Phenolic Compounds, and Oxidative Stress Index in Sun-dried Vegetables: Example of Gaziantep, Türkiye

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ABSTRACT

Sun drying is one of the oldest methods of preserving food, including vegetables. This process reduces the moisture content of the vegetables, inhibiting the growth of microorganisms that cause spoilage and rot. This study aims to investigate the total antioxidant/oxidant status (TAS/TOS), phenolic compound, and oxidative stress index (OSI) in sun-dried vegetables (okra, peppers (hot-sweet), zucchini, and eggplant). The vegetables used were chosen randomly. The total antioxidant/oxidant status of the sun-dried vegetables prepared in methanol was analysed using a spectrophotometer and commercially available kits. Total phenolic compounds were analysed using a spectrophotometer based on the Folin-Ciocalteu reagent method. OSI represents the balance between antioxidants and oxidants in the system, with higher OSI values indicating greater oxidative stress. The total antioxidant status and phenolic compounds were highest in sun-dried okra (6.568±0.22 mmol Eq/L, 79.51±0.21 mg/g respectively). The total oxidant status was highest in sun-dried eggplant (14.645±0.24 H₂O₂/L). The oxidative stress index was highest in sun-dried zucchini (0.249±0.00 H₂O₂/L). Consequently, this study highlights the high content of antioxidants and phenols, emphasizing their potential health benefits in reducing oxidative stress. It reaffirms the value of traditional sun-drying methods in preserving the nutritional integrity of vegetables. These findings support the inclusion of sun-dried vegetables in the diet for their high nutritional value and health benefits.

Güneşte Kurutulmuş Sebzelere Total Antioksidan/Oksidan Durumun, Total Fenolik Bileşiklerin ve Oksidatif Stres İndeksinin Araştırılması: Gaziantep/Türkiye Örneği

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Anahtar Kelimeler:

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Total oksidan miktarı

Oksidatif stres indeksi

Total fenolik bileşikler

ÖZ

Güneşte kurutma, sebzeler dahil olmak üzere gıdaları korumanın en eski yöntemlerinden biridir. Bu işlem, sebzelerin nem içeriğini azaltarak bozulma ve çürümeye neden olan mikroorganizmaların büyümesini engeller. Bu çalışma, güneşte kurutulmuş sebzelerde (bıyık, biber (acı-tatlı), kabak ve patlıcan) toplam antioksidan/oksidan seviyesi (TAS/TOS), fenolik bileşik ve oksidatif stres indeksini (OSİ) araştırmayı amaçlamaktadır. Kullanılan sebzeler rastgele seçilmiştir. Metanolde hazırlanan güneşte kurutulmuş sebzelerin toplam antioksidan/oksidan durumu, ticari olarak mevcut kitler ve bir spektrofotometre kullanılarak analiz edilmiştir. Toplam fenolik bileşikler, Folin-Ciocalteu reaktif metodu temel alınarak spektrofotometre yardımıyla

analiz edilmiştir. OSİ, sistemdeki antioksidanlar ve oksidanlar arasındaki dengeyi temsil eder ve daha yüksek OSİ değerleri daha fazla oksidatif stresi gösterir. Toplam antioksidan miktarı ve fenolik bileşik içeriği en yüksek güneşte kurutulmuş bamyada bulunmuştur (sırasıyla 6.568±0.22 mmol Eq/L, 79.51±0.21 mg/g). Toplam oksidan miktarı en yüksek güneşte kurutulmuş patlıcanda bulunmuştur (14.645±0.24 H₂O₂/L). Oksidatif stres indeksi en yüksek güneşte kurutulmuş kabağa aittir (0.249±0.00 H₂O₂/L). Sonuç olarak, bu çalışma, antioksidanların ve fenollerin yüksek miktarını vurgulayarak, oksidatif stresi azaltma potansiyelleri açısından olası sağlık faydalarını ortaya koymaktadır. Geleneksel güneşte kurutma yöntemlerinin sebzelerin besin bütünlüğünü korumadaki değerini yeniden teyit etmektedir. Bu bulgular, yüksek besin değeri ve sağlık faydaları nedeniyle güneşte kurutulmuş sebzelerin diyetlere dahil edilmesini desteklemektedir.

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1. Introduction

Drying has been used since ancient times to preserve fruits and vegetables. The main purpose of drying is to prepare food for consumption by ensuring the availability of food throughout the year. The traditional drying process is quite natural and simple, as it is usually done with solar energy. On the other hand, industrial drying is more complex, as a variety of equipment is used, and drying parameters are carefully studied and optimized at each stage of the process. Drying not only preserves the product but also has a beneficial effect on spices, herbs, bioactive enzymes, and nuts (Szychowski et al., 2018). The drying proceeds as follows: first, water is evaporated from the structure of the food, and then this vapor is removed from the surface of the product. Due to the loss of water in the structure of the food, the water activity decreases below the level at which microorganisms can multiply, preventing spoilage by bacteria, mold, and yeast and prolonging the shelf life of the food. The volume of the food decreases due to drying. The reduced volume of the food makes it easier to transport and store (Alcay et al., 2015). In addition, by changing the flavor and texture characteristics of food, new generations of snacks are produced that can be a healthier alternative to other commercially available products such as candy (Sehrawat et al., 2018).

In recent years, the study of the antigenotoxic effects of natural plant products has increased the importance of natural antioxidants in minimizing the genotoxic effects of physical and chemical substances on DNA. Natural antioxidants are very effective against the adverse effects of mutagenic substances to which humans are exposed (Uluman and Kilicle, 2020). Antioxidants in foods are substances that can reduce the negative effects of oxygen or nitrogen free radicals in the human body. The status of antioxidants in foods and the bioavailability of antioxidants depend on the type of food, the time of harvest, the harvesting method, the environmental conditions in which it is grown, and the storage and preparation conditions (Kasim and Kasim, 2019).

Vegetables and fruits are known as "super antioxidants" thanks to the bioactive compounds they contain (Deveci et al., 2016; Mattioli et al., 2018, Delibaş, 2022). Antioxidants have blood pressure regulating (Amponsah-Offeh, 2023), blood sugar regulating (Rajendiran, 2018), anticarcinogenic (Dixit and Pandit, 2023), autoimmune supporting (Farrokhi, 2021), antibacterial, antifungal, antiviral (Song, 2024),

cholesterol lowering, and antithrombotic effects (Amponsah-Offeh, 2023). Studies have shown that adequate consumption of fruits and vegetables plays an important role in preventing non-communicable diseases such as cardiovascular disease (Ness and Powles, 1997), cancer (Dixit and Pandit, 2023), diabetes (Rajendiran, 2018), Alzheimer's disease, and Parkinson's disease, as well as chronic diseases characterized by age-related decline in body functions (Martin et al., 2002). Vegetables and fruits are consumed fresh in the season in which they are grown, but they are also processed out of season, for example, by drying and freezing (Ulger et al., 2018). They contain bioactive substances that successfully cure a variety of diseases, especially those related to oxidative stress (Deveci et al., 2023).

Phenolic compounds contribute significantly to total water-soluble antioxidant activity are the largest group of secondary metabolites produced by plants. Flavonoids, lignin precursors, and phenolic acids are the most important phenolic compounds (Okmen et al., 2009). These compounds not only have unique sensory properties such as bitterness, astringency, color, flavor, odor, and antioxidant stability, but also various biological activities such as anticancer, anti-inflammatory, antimicrobial activities, degradation of oxidized low-density lipoprotein, and enhancement of plasmatic antioxidant capacity (Ricciutelli et al., 2017; Yu et al., 2021). Carotenoids, such as capsanthin, α - and β -carotene, and zeaxanthin, are the main contributors to these protective effects (Kaur and Kapoor, 2001). Orange sweet peppers are a rich source of zeaxanthin, violaxanthin, and β -carotene; in yellow sweet peppers, violaxanthin, antheraxanthin, lutein, and zeaxanthin are the dominant carotenoids (Pugliese et al., 2014). Fresh vegetables are known to be a natural source of antioxidants and phenolic compounds (Delibaş, 2022). However, the values of antioxidants and phenolic compounds of vegetables consumed in our country by sun drying are not known. Thus, this study aimed to determine the total antioxidant status (TAS), total oxidant status (TOS), total phenolic compounds (TPC), and oxidative stress index (OSI) of sun-dried vegetables in Türkiye.

2. Material and Methods

2.1. Production of dried vegetables

The material used was dried vegetables produced and collected in the Gaziantep province in the Southeast Anatolia region of Turkey in June-July 2023. These dried vegetables were ground into powder. The powdered dried vegetable samples were stored in dark glass bottles protected from light and moisture. According to the sample calculation carried out with the program G-power 3.1.9.2, 8 vegetable varieties were examined in each group, as 8 samples were required to achieve a margin of error of 5 at a confidence level of 95%, with $\alpha=0.05$, power=0.95, and an effect size of 0.8. Thus, 20-25 samples from each vegetable group were used. Samples selected to represent the mass were thoroughly homogenized before analysis. 10 g of homogenized vegetables were used to obtain the extracts to be used in the experiments. The procedure was performed as described by Remberg et al. (2003) and Stangeland et al. (2009).

2.2. Preparation of dried vegetable extracts

Dried vegetable samples were taken from 3 different points and extracted. The extracts obtained from the different vegetables taken from these three points were analysed separately in the spectrophotometer, and the data set was created by taking the average of the results. 10 g of dried vegetable samples (okra, sweet and hot pepper, zucchini, and eggplant) (Figure 1) stored in powder flasks were weighed, 500 mL of methanol (CH₃OH) was added, and Soxhlet extraction was performed at 60 °C for about 18 hours. The methanol was removed from the obtained extract by evaporation at 48 °C in a vacuum evaporator (Heidolph Lavorota 4000 rotary evaporator). The prepared plant extracts were stored at -18 °C until analysis (Heleno et al., 2016). A random sampling technique was employed for sample collection. Accordingly, the selection was made on the assumption that each of the dried vegetables had an equal chance of being included in the study.

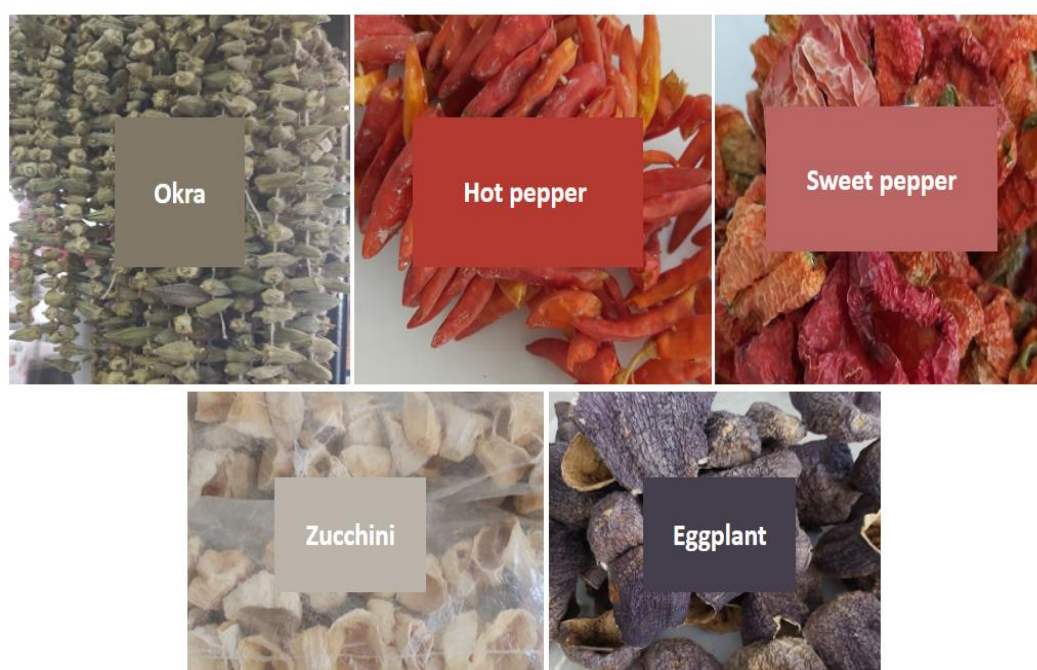


Figure 1. Samples of dried vegetables

2.3. Total antioxidant/oxidant status analysis

The TAS in the extracts was analysed spectrophotometrically using the TAS Kit developed by Erel (2004) (Rel Assay Kit Diagnostics, Turkey). TAS was calculated as mmol Trolox equivalent/L. TOS in the vegetable extracts was analysed spectrophotometrically using the TOS Kit (Rel Assay Kit Diagnostics, Turkey) developed by Erel (2005). TOS was calculated as $\mu\text{mol H}_2\text{O}_2$ equivalent/L. The OSI, expressed as a percentage of the ratio between the TOS and the TAS value, was calculated from the results. To calculate the OSI values, the mmol value was converted from the unit of the TAS result to the unit μmol of the TOS result. The percentage ratio of TOS to TAS yielded the OSI, an indicator of the degree of oxidative stress. To perform the calculation, the unit of TAS, mmol Trolox equivalent/L, was changed to μmol Trolox equivalent/L. The OSI value was calculated according to the following

formula: $OSI = [(TOS, \mu\text{mol H}_2\text{O}_2 \text{ equivalent/L}) / (TAS, \mu\text{mol Trolox equivalent/L}) \times 100]$ (Eren et al., 2015; Delibaş and Kıray, 2023).

2.4. Total phenolic compounds analysis

TPC analysis was performed with a spectrophotometer using a modification of the Folin-Ciocalteu reagent method (Slinkard and Singleton, 1977). This method is because phenolic compounds dissolved in water, or organic solvents form a purple color with Folin-Ciocalteu reagent (47641, Sigma) in an alkaline environment. Since this colored compound has an absorption maximum of 760 nm, it was analysed spectrophotometrically (Epoch, Biotek, USA) at 760 nm. Gallic acid (G7384, Sigma) was used as a standard, and the TPC was calculated as mg gallic acid equivalent (GAE) g⁻¹. TPC analysis was repeated three times, and the results were averaged.

2.5. Statistical analysis

The Windows operating system-based program GraphPad Prism 9.5.0 was used for all statistical analyses of the data obtained in our studies. The distribution of the study groups was examined with the One-sample Kolmogorov-Smirnov test. Data are presented as mean±standard error of the mean (SEM), and our data were considered statistically significant if $p < 0.05$. Comparisons between groups were performed with a one-way analysis of variance (ANOVA), and Tukey's test was used as a post hoc test. Before the analysis, it was checked whether the data fulfilled the assumptions of the ANOVA. For this purpose, the equal distribution of the variances between the groups was examined in relation to the dependent variable with the Levene statistic.

3. Results

In this study, TAS/TOS, TPC content, and OSI were analyzed in a selection of sun-dried vegetables commonly consumed in the Gaziantep region of Turkey. The main objective was to evaluate how the sun-drying process affects different vegetables' antioxidant capacity, oxidant content, and phenolic content. These parameters are crucial for understanding the nutritional value and potential health benefits of sun-dried vegetables.

The results of the one-way ANOVA analysis performed to determine whether there was a statistically significant difference between the means of the measurements obtained in the study by dried vegetables are shown in table 1. It was found that TAS was highest in dried okra (6.568 ± 0.22) and lowest in dried sweet pepper (5.320 ± 0.15). TOS was highest in dried eggplant (14.645 ± 0.24) and lowest in dried sweet pepper (12.239 ± 0.24). TPC is highest in dried okra and lowest in dried hot pepper. The OSI is highest in dried zucchini (0.248 ± 0.005) and lowest in dried okra (0.212 ± 0.01) (Table 1).

Table 1. TAS, TOS, TPC, and OSI properties of drying vegetables

Vegetables	TAS ($\bar{x}\pm\text{SEM}$)	TOS ($\bar{x}\pm\text{SEM}$)	OSI ($\bar{x}\pm\text{SEM}$)	TPC ($\bar{x}\pm\text{SEM}$)
Okra	6.568 \pm 0.22 ^a	13.972 \pm 0.23 ^b	0.213 \pm 0.01 ^a	79.51 \pm 0.21 ^a
Hot pepper	5.856 \pm 0.24	14.219 \pm 0.31 ^b	0.243 \pm 0.00 ^b	68.98 \pm 0.47 ^b
Sweet pepper	5.320 \pm 0.15 ^b	12.239 \pm 0.24 ^a	0.230 \pm 0.00	74.85 \pm 0.24 ^d
Zucchini	5.734 \pm 0.21	14.271 \pm 0.23 ^b	0.249 \pm 0.00 ^b	75.16 \pm 0.32 ^d
Eggplant	6.154 \pm 0.13	14.645 \pm 0.24 ^b	0.238 \pm 0.00	72.73 \pm 0.44 ^c
	F=5.693	F=13.972	F=5.704	F=119.503
	P=0.012	P=0.000	P=0.012	P=0.000

Data are expressed as mean \pm SEM of the assays. Determinations were made in triplicate. The same superscript letters show values with statistically insignificant differences per property ($p>0.05$)

The TAS of dried okra is higher than other vegetables and significantly higher than dried sweet pepper ($p=0.008$) (Figure 2a). The TOS of dried sweet peppers is significantly lower than that of other vegetables (dried okra: $p=0.005$; dried hot pepper: $p=0.005$; dried zucchini: $p=0.001$; dried eggplant: $p=0.000$) (Figure 2b). The TPC of dried okra is significantly higher than that of other dried vegetables (dried hot pepper: $p=0.000$; dried sweet pepper: $p=0.000$; dried zucchini: $p=0.000$; dried eggplant: $p=0.000$). The TPC of dried hot pepper is significantly lower than that of other dried vegetables (dried sweet pepper: $p=0.000$; dried zucchini: $p=0.000$; dried eggplant: $p=0.011$). The TPC of dried eggplant is significantly lower than that of dried sweet pepper and dried zucchini ($p=0.000$; $p=0.004$ respectively) (Figure 2c). The OSI of dried okra is significantly lower than that of dried hot pepper and dried zucchini ($p=0.029$; $p=0.009$ respectively) (Figure 2d).

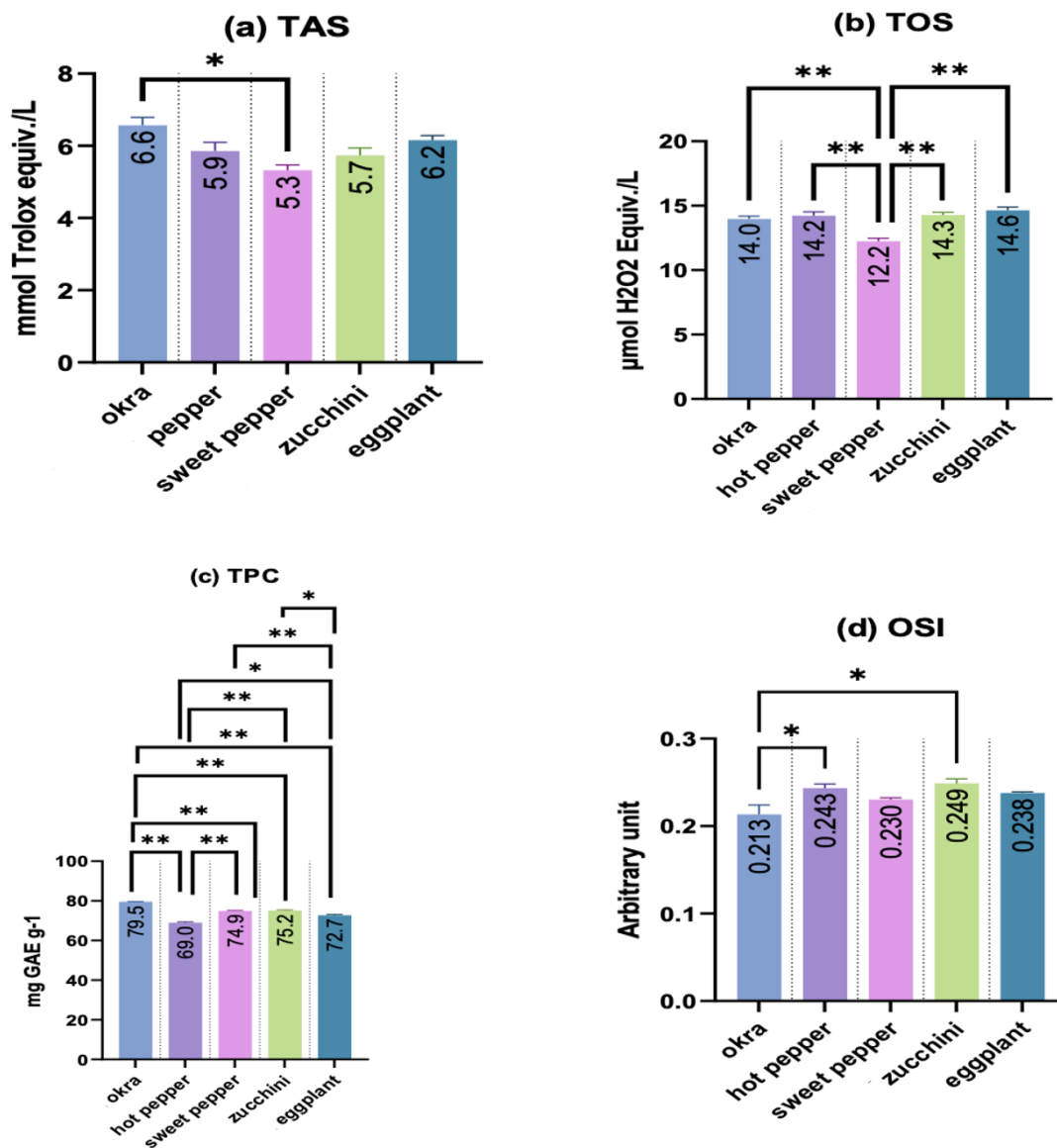


Figure 2. Properties of dried vegetables. **(a)** TAS values of dried vegetables. Data are mean±SEM (*p<0.05). **(b)** TOS values of dried vegetables. Data are mean±SEM (**p<0.01). **(c)** TPC values of dried vegetables. Data are mean±SEM (*p<0.05; **p<0.01). **(d)** OSI values of dried vegetables. Data are mean±SEM (*p<0.05).

4. Discussion

Oxidative reactions generate free radicals, initiating chain reactions that can damage cells. In contrast, antioxidants neutralize these free radicals and stop the chain reactions, thereby slowing down or preventing oxidation (Slinkard and Singleton, 1977). The role of antioxidants in mitigating oxidative damage is well documented, particularly in their potential to counteract diseases such as cancer, diabetes, and cardiovascular disease (Islam, 2019). More recent studies, such as Chaudhary et al. (2023), emphasize the complex interactions between antioxidants and free radicals, highlighting their role in both physiological processes and disease prevention. Fruits and vegetables are important sources of

antioxidants due to their high antioxidant activity and the rich mixture of antioxidant compounds (Landete, 2013). Increasing concern about the potentially carcinogenic effects of synthetic antioxidants in foods has increased interest in natural antioxidants, considered more reliable for health (Koca and Karadeniz, 2003). Antioxidant activity, a frequently studied quality parameter in dried vegetables, is primarily related to the content of vitamins and polyphenols. Although there is extensive research on various dried vegetables, okra, peppers, zucchini, and eggplant have been studied only to a limited extent (Zhang and Hamauzu, 2003; Al-Dabbas et al., 2023; Saha et al., 2023; Thanh et al., 2023). Considering the limited number of studies determining the overall status of antioxidants and oxidants in these vegetables, this study aims to make an important contribution to the literature.

Several studies have already investigated the antioxidant properties of certain vegetables. For example, Chen et al. (2015) investigated the antioxidant capacity of okra and found significant levels of phenolic compounds that contributed to its high antioxidant capacity. Xu et al. (2020) specifically investigated the antioxidant status of okra dried by different methods (freeze-drying, sun-drying, oven-drying, and microwave drying) and found high levels of antioxidants in all samples. In our study, the total antioxidant capacities of sun-dried okra were significantly higher than those of other vegetables, including hot peppers, dried zucchini and dried eggplant. These results are consistent with previous studies suggesting that the release of phenolic compounds by heat treatment could explain the increased antioxidant levels of okra (Llorach et al., 2002; Martinez-Valdivieso et al., 2017). In addition, Gemedé et al. (2015) studied the nutritional quality of okra, highlighting its high content of polyphenols and other bioactive compounds that contribute to its antioxidant effects. In addition, Shen et al. (2020) found significant antioxidant activity in different okra varieties, highlighting how both varietal differences and preservation methods can influence antioxidant content. This research supports the idea that the antioxidant properties of okra make them an important part of the diet to manage oxidative stress and prevent related diseases.

In terms of TOS, dried okra had a lower TOS than dried peppers, dried zucchini, and dried eggplant, although it was still higher than that of dried peppers. This inverse relationship between TAS and TOS is well documented, as antioxidants mitigate oxidative stress by neutralizing free radicals (Islam, 2019). Bell peppers are known for their high content of bioactive phytochemicals, especially carotenoids. Kim et al. (2006) have shown that regular consumption of peppers can reduce the risk of cancer (Dixit and Pandit, 2023), and cardiovascular disease (Ness and Powles, 1997). Our study found that TAS was higher in hot peppers than in sweet peppers, which is consistent with previous research on the high carotenoid content of hot peppers (Pugliese et al., 2014). However, genotype, ripening stage, and drying method have a significant effect on carotenoid content, suggesting that further studies are needed to fully explain the differences observed in our study. TOS content was also higher in hot peppers, which may reflect the parallel relationship between TAS and TOS observed in previous studies (Åsgård et al., 2007).

Zucchini is known for its high content of natural antioxidants, including beta-carotene, phenolic compounds, and vitamin C, all of which contribute to its high TAS value (Guzman et al., 2010). Especially the content of natural antioxidants such as beta-carotene, phenolic compounds, and vitamin C is quite high in green zucchini. However, the shelf life of zucchini is limited to 1 to 2 days because it is a vegetable that spoils quickly after being cut, losing firmness, turning brown, and rotting. Therefore, if zucchini is to be eaten fresh in season, it should be consumed immediately. If it is to be consumed out of season, various drying and freezing methods should be used for preservation (Guzman et al., 2010). The most common method for drying zucchini is sun-drying outdoors. Although this method is the most widely used, there are not enough studies on which preservation method is best in terms of its effects on the nutritional content of zucchini. In a study of fresh zucchini, it was found that both yellow and green zucchini were high in antioxidants. Active antioxidant compounds such as lutein, β -carotene, zeaxanthin, and dehydroascorbic acid in zucchini have a beneficial effect on human nutrition (Eissa et al., 2013). A recent study by Moradi et al. (2019) has also confirmed that the antioxidant properties of zucchinis may play a role in managing oxidative stress and improving blood glucose control in type 2 diabetics.

Although there is no study evaluating the antioxidant or oxidant properties of dried zucchini, our study clearly shows that the TAS of dried zucchini is high. Moreover, zucchini ranked second only to dried eggplant in TAS. It can be concluded that the antioxidant activity of the vegetable correlates with the flavonoid content. The TPC in dried okra, which has the highest antioxidant status among the vegetables studied, was also the highest (Martínez-Valdivieso et al., 2017). The vegetable with the lowest antioxidant activity is sweet pepper, while the vegetable with the lowest TPC is hot pepper. Optimal antioxidant status protects against oxidative stress, which is defined as an imbalance between free radicals and antioxidant protection. Therefore, as the TAS increases, the OSI decreases (Åsgård et al., 2007; Islam, 2019). Our study proved this. The vegetable with the highest TAS value and the lowest OSI value is dried zucchini.

Eggplants, one of the most consumed dried vegetables, are traditionally sun-dried for long-term preservation. Despite their popularity, few studies have investigated the nutritional properties of dried eggplant. Our study fills this gap and shows that eggplant has the second highest TAS value after dried okra. Interestingly, eggplant also ranked first in TOS, further emphasizing the balance between the antioxidant and oxidative properties of this vegetable.

5. Conclusions

In conclusion, this study is one of the few that analysed the biochemical content of dried vegetables. This study investigated the TAS/TOS, TPC and OSI of five different dried vegetables. It was found that all the vegetables studied had antioxidant capacity. While total antioxidant status was highest in dried okra, it was lowest in dried sweet pepper. The flavonoid content of the vegetable correlated closely with its antioxidant activity. Dried vegetables have been shown to contain antioxidants, although not as much

as fresh vegetables. To obtain more accurate results, vegetable groups and analyses must be diversified. It is thought that the promising results of this study will form the basis for further studies.

Conflict of Interest

No potential conflict of interest was reported by the authors.

Author contributions

H.A.D. and A.D. have designed the study and collected the data. F.H.D. executed the experiment with the help by H.A.D. F.H.D. wrote the article, and critically reviewed by H.A.D. All authors accept responsibility for all content.

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