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Analysis of Fatty Acid and Some Lipophilic Vitamins Found in the Fruits of the *Ficus carica* Variety Picked from the Adiyaman District

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Abstract: Fig is a fibrous plant, which contains organic acids, provitamin A and vitamin C. It is widely consumed in both fresh and dried form in Turkey. In this study, we aimed to establish the fatty acid and lipophilic vitamin components of the fruits of the *Ficus carica* variety found in the province of Adiyaman, Turkey. For the analysis of the fig fruit of the *Ficus carica* variety, the whole fruit, the skin and the flesh were analyzed separately using High Performance Liquid Chromatography (HPLC) and Gas Chromatography (GC). As a result of the analysis, on examining the fig fruit as a whole, several vitamins with lipophilic properties were identified. Upon analyzing different sections of the fruit, it was observed that the skin and the flesh had different vitamin and fatty acid contents. The fig fruit was found to be rich in terms of fatty acids as well as vitamin content. Primarily, it was identified that in addition to the non-essential fatty acids such as palmitic (16:0), stearic (18:0) and oleic (18:1 n-9) fatty acids, it also contained a significant level of essential fatty acids such as linoleic acid (18:2 n-6) and linolenic acid (18:3 n-3). In conclusion, the fig fruit was confirmed to have important nutrients in terms of human nutrition rather than just a fibrous food.

Key words: *Ficus carica*, fatty acids, lipophilic vitamins, fruits, HPLC, adiyaman district

INTRODUCTION

Turkey accounts for 30% of the world fig fruit production (Tous and Ferguson, 1996; Doymaz, 2005). The fig fruit is consumed fresh in summer, whilst the dried form is always available in other seasons. Fig belongs to the Moraceae family and has >750 varieties; its natural range is Western Asia and the Mediterranean region and it grows in countries with a mild climate. In Turkey, fig trees are grown in the Aegean region and in the areas where the Mediterranean climate prevails (Bellakhdar *et al*., 1991; Ensminger *et al*., 1994; Dominguez *et al*., 1996). Fresh fig can be consumed within 7-10 days of picking if kept under suitable conditions (Veberic *et al*., 2008).

In recent years, the nutritional value of the fig fruit as well as its pharmacological and various others were investigated. These studies have shown that consumption of fig fruit prevents coronary occlusion, as a laxative effect due to its fibrous structure (Rubnov, 2001) has sore throat soothing, laxative, stimulant and expectorant effects (Bellakhdar *et al*., 1991; Guarrera *et al*., 2003) and is reported to be used in hemorrhoid treatment (Baytop, 1984). Furthermore, it has been claimed that its sap has certain chemicals/nutrients, which prevents the growth of cancerous cells (Rubnov *et al*., 2001) that the boiled fig leaves have hypoglycemic effect in type-1 diabetic rats (Serraclara *et al*., 1998) that it contains A, B_1, B_2 and C vitamins and minerals (Doymaz, 2005) that together with high levels of primarily glutamine and aspartic acid in its structure, its amino acid content is high that due to 28% of the fibers contained in the fig being water soluble, fig helps regulate blood sugar levels and blood cholesterol (Solomon *et al*., 2006; Lianju *et al*., 2003) and that it leads to a significant increase in the antioxidant capacity in the plasma within 4 h of consumption (Vinson *et al*., 2005). The nutritional values of 100 g fresh fig is as it follows (Morton, 1987) (Table 1).

Table 1: Lipophilic vitamins and fatty acids of the fig fruit in terms of its nutritional values

Nutritional values	Fresh	Dried
Calories	80	274
Moisture	77.5-86.8g	23.0g
Protein	1.2-1.3g	4.3g
Fat	0.14-0.30g	1.3g
Carbohydrates	17.1-20.3g	69.1g
Fiber	1.2-2.2g	5.6g
Ash	0.48-0.85g	2.3g
Calcium	35-78.2mg	126mg
Phosphorus	22-32.9mg	77mg
Iron	0.6-4.09mg	3.0mg
Sodium	2.0mg	34mg
Potassium	194mg	640mg
Carotene	0.013-0.195mg	-
As Vitamin A	20-270I.U.	80I.U.
Thiamine	0.034-0.06mg	0.10mg
Riboflavin	0.053-0.079mg	0.10mg
Niacin	0.32-0.412mg	0.7mg
Ascorbic acid	12.2-17.6mg	0mg
Citric acid	0.10-0.44mg	

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The present study analyzed the lipophilic vitamins and fatty acids of the fig fruit in terms of its nutritional values obtained/(grown in) from the district of Adiyaman. The reasons being that fig is a fruit, which helps cell regeneration due to its high protein, vitamin and mineral content and the scientific studies conducted to date have not fully established the fatty acid and lipophilic vitamin contents besides the mineral substance content of different sections of this fruit. As a result of this study, it was identified that fig, which plays an important role in human nutrition intake is not only a fibrous fruit but also an important source of fatty acid and lipophilic vitamins.

MATERIALS AND METHODS

Fig samples: Fig samples were taken from the fig trees grown in Adiyaman province at the end of August beginning of September when they well developed. Fig samples Stored in -25°C until being used for biochemical analyses. We grouped samples as skin, white inner part, red inner part and whole fig.

Extraction of lipids: Fig samples whose wet weights were determined were homogenized with 3/2 (v v^{-1}) Hexane-isopropanol mixture (Hara and Radin, 1978). The homogenate was centrifuged at 5000 rpm for 5 min at 4°C and parts of fig remnants were precipitated. The supernatant part was used in the ADEK vitamin and fatty acid analysis.

Preparation of fatty acid methyl esters: An aliquot was taken from the supernatant part of the fig samples and 5 mL of 2% methanolic sulfuric acid was added. The mixture was vortexed and then kept at 50°C for 12 h. Then, after being cooled to room temperature, 5 mL of 5% sodium chloride was added and then it was vortexed. Fatty acid methyl esters were extracted with 2×5 mL hexane. Fatty acid methyl esters were treated with 5 mL 2% $KHCO_3$ solution and then the hexane phase was evaporated by the nitrogen flow and then by dissolving in 0.5 mL fresh hexane (Christie, 1992), they were taken to auto sampler vials.

Gas chromatographic analysis of fatty acid methyl esters: Methyl esters were analyzed with the SHIMADZU GC 17 Ver. 3 gas chromatography (Kyoto, Japan). For this analysis, 25 m of long Machery-Nagel (Germany) capillary colon with an inner diameter of 0.25 μm and a thickness of 25 micron film was used. During the analysis, the colon temperature was kept at 120-220°C, injection temperature was kept at 240°C and the detector temperature was kept at 280°C. The colon temperature program was adjusted from 120-220°C and the temperature increase was determined to be 5°C min^{-1}, until 200°C and 4°C min^{-1} from 200-220°C. It was kept at 220°C for 8 min and the total duration was set as 35 min and nitrogen gas was used as the carrier gas. During the analysis, before the analysis of fatty acid methyl esters, mixtures of standard fatty acid methyl esters were injected and the residence time of each fatty acid was determined. After this process, the necessary programming was made and the fatty acid methyl esters mixtures of the samples were analyzed (Christie, 1992).

HPLC analysis of adek vitamins: Five mL supernatant was taken to 25 mL tubes with caps and 5% KOH solution was added. After it was vortexed, it was kept at 85°C for 15 min. The tubes were then taken and cooled to room temperature and 5 mL of pure water was added and mixed. Lypophlic molecules that did not saponify were extracted with 2×5 mL hexane. The Hexane phase was evaporated with nitrogen flow. It was dissolved in 1 mL (50 + 50%, v v^{-1}) acetonitril/methanol mixture and then was taken to auto sampler vials and was analyzed.

The analysis was made with the Shimadzu brand HPLC device. In the device as the pump LC-10 ADVP UV-visible, as the detector SPD-10AVP, as column oven CTO-10ASVP, as auto sampler SIL-10ADVP, as degasser unit DGU-14A and Class VP software (Shimadzu, Kyoto Japan) was used and during the mobile phase the acetonitril/methanol (60 + 40% v v^{-1}) mixture was used. The mobile phase flow rate was determined to be 1 mL A UV detector was used for the analysis and as a column the Supelcosil LC 18 (15×4.6 cm, 5 μm; Sigma, USA) column was used. For vitamin E, 202 nm (Katsanidis and Addis, 1999) and for vitamin D and K, 265 nm was used.

RESULTS AND DISCUSSION

The results of some lipophilic vitamins found in different sections of the fruit used in the present study belonging to the *Ficus carica* variety from Adiyaman province are shown in Table 2 and the fatty acid amounts in Table 3.

The results of the present study established that all of the fig fruit used as material in this study contained γ-Tocopherol, δ-Tocopherol, vitamin D_2, vitamin D_3, α-Tocopherol, α-Tocopherol acetate and vitamin K_1. The content values of these vitamins vary in different sections of the fruit. It was identified that the skin and the white sections of the fruit contained high amounts of γ-Tocopherol whilst this value was not found in the red core section of the fruit. Whereas, δ-Tocopherol was

Table 2: Lipophilic vitamin contents of whole fig fruit and different sections of fig fruit

Some lipophilic vitamins	Whole fig fruit	Skin	Red part	White part
γ-Tocoferol	0.90±0.03	14.60±2.80	0.67±0.05	12.95±8.55
δ-Tocopherol	0.20±0.01	0.70±0.05	0.35±0.05	0.30±0.01
D_2	0.20±0.05	0.32±0.22	0.15±0.05	2.16±0.02
D_3	3.57±0.97	0.60±0.05	11.30±1.31	0.40±0.10
α-Tocopherol	0.35±0.08	1.04±0.32	0.75±0.43	1.55±0.60
α-Tocopherol asetat	0.37±0.10	0.36±0.12	1.00±0.33	3.33±1.43
K_1	4.05±0.73	0.56±0.13	4.05±0.75	3.77±0.73

Table 3: Fatty acid compositions of the whole fig fruit and different sections of fig fruit

Fatty acids	Whole fig fruit	Skin	Red part	White part
16:0	17.52±1.39	24.87±1.32	12.37±2.04	29.69±1.51
16:1, n-7	2.69±0.45	3.12±0.31	0.67±0.12	0.40±0.04
16:1, n-9	1.27±0.56	1.44±0.12	0.70±0.24	0.20±0.04
18:0	6.22±0.72	7.59±0.95	3.14±0.09	8.53±2.14
18:1, n-9	19.72±1.07	24.61±1.38	16.12±1.98	28.31±1.48
18:1, n-7	2.54±0.18	3.05±0.60	14.50±2.85	2.35±0.46
18:2, n-6	23.04±0.48	23.10±2.41	24.38±0.82	19.35±1.61
18:3, n-6	1.46±0.06	1.67±0.42	1.39±0.07	0.43±0.02
18:3, n-3	23.87±6.27	6.57±2.00	39.32±2.45	13.97±2.16
18:4, n-3	1.71±0.58	0.71±0.08	2.34±0.23	0.41±0.05
20:0	1.28±0.47	3.58±2.93	0.23±0.07	0.33±0.01
20:3, n-6	6.44±0.95	5.95±1.29	1.29±1.16	7.53±0.83
24:1	0.75±0.15	2.29±1.16	0.89±0.16	4.80±1.28

found in all sections of the fruit in varying amounts especially in the skin it was found to be slightly higher. Vitamin D_2 was also found in all sections of the fruit but the amount of this vitamin was found to be higher in the white flesh compared to the other sections. Despite being found in all sections of the fruit, the amount of vitamin D_3 was found to be much higher in the red flesh. α-Tocopherol, which has antioxidant properties (Wang and Quinn, 1999; Baydas *et al*., 2002; Celik *et al*., 2002) identified to have uniform amounts in all sections of the fruit. α-Tocopherol acetate is also found in all sections of the fruit and it was observed that this value was higher in the white flesh compared to the other sections. It was identified that among all the vitamins, present in fig vitamin K_1 is found to be the highest in the fruit but the least in the skin.

In a study conducted using fig leaves, it was stated that its natural and chloroform-treated forms reduced diabetes induced hyperglycemia and vitamin E levels (Perez *et al*., 2003). Konyalioglu *et al.* (2005) reported that the fig leaves contain 10 times more vitamin E compared to the soya bean, which is used as an industrial source of α-tocopherol (vitamin E).

A similar study conducted by Jeong and Lachance (2001) analyzed the fatty acid composition of the skin, whole fruit and the flesh of the mission variety of the *Ficus carica* fruit by GC and the sterol content by Gas Chromatography_Mass Spectrometry (GC_MS). They found that the whole fruit contained high amounts of sitosterol. Furthermore, plant sterols such as campesterol, stigmasterol and fucosterol were also detected in the whole fruit. They stated that the fatty acid composition of the variety under investigation contained myristic acid (14:0), palmitic acid (16:0), stearic acid (18:0), oleic acid (18:1 n-9), linoleic acid (18:2 n-6) and linolenic acid (18:3 n-3).

The study carried out and found that the *Ficus carica* latex inhibited the DNA synthesis of cancerous cells and that the *Ficus carica* latex has a strong anti-proliferation effect by causing apoptic cell death.

Perez *et al*. (2000) stated that streptozotocin induced diabetic rats, which were given boiled fig leave extracts for three weeks; as a result the boiled fig leaves exhibited a hypoglycemic effect.

The analyses conducted indicated that the fig fruits used in the present study had fatty acid components in varying amounts in different sections of the fruit. The fatty acids analysis of the fruit showed, significant levels of non-essential fatty acids such as palmitic (16:0), stearic (18:0), oleic (18:1 n9) fatty acids and essential fatty acids such as linoleic acid (18:2 n6) and linolenic acid (18:3 n3).

Essential fatty acids need to be obtained from food externally since they cannot be synthesized by the human body (Jiang *et al*., 1998; Youdim *et al*., 2000; Yilmaz *et al.*, 2008). Since, linoleic acid (18:2 n6), which is essential nutrient for humans, is found at high levels especially in the skin of fig, it is important to consume the fruit as a whole without peeling the skin to maximize linoleic acid intake. It was established that among these fatty acids, palmitic, oleic and stearic acids are found most in the white flesh of the fruit. Whereas, linoleic and linolenic acid concentrations were found to be highest in the red flesh. Whilst other fatty acid components analyzed were observed to occur in lesser quantities. As a result of a similar study conducted by Kolesnik *et al.* (1987), the basic fatty acid components in the fig mostly consisted of linoleic, linolenic, oleic and palmitic acids.

CONCLUSION

In conclusion, the scientific studies to date have not focused on the fatty acid component amounts in different sections of the fig. This study demonstrated that in terms of human nutrition, fig is a nutritious fruit, which contains important molecules rather than just a fibrous food.

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