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Determination and Comparison of Total Polyphenol and Vitamin C Contents of Natural Fresh and Commercial Fruit Juices

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Abstract: Owing to interest in the role of antioxidants in human health which has prompted research to assess fruit and their products antioxidants, such as vitamin C and phenolics and also because of increment in commercial fruit juices consumption, the objectives of present study were to determine and compare the total polyphenol and vitamin C contents of natural fresh and commercial fruit juices. Folin-Ciocalteu assay and Spectrophotometric method were used to measure the total polyphenol and vitamin C. The concentrations of total polyphenol were ranging from 23.75±1.6 to 420.69±4.9 mg/100 ml for fresh and 18.57±3 to 381.9±3.4 mg/100 ml for commercial juices with lowest amount in apricot and the highest amount in pomegranate juices in both sources. Significant differences ($p<0.05$) were observed in the polyphenol contents of peach, pomegranate, grapefruit and orange between fresh and commercial juices. The vitamin C levels of fresh juices were ranging from 14.31 for apricot to 24.51 mg/100 ml for orange juices and those of commercial were from 13 for mango to 17.29 mg/100 ml for pomegranate juices. The mean vitamin C contents of fresh juices were higher than those of commercial but these differences were not significant. Although various juices differed markedly in the quantity of polyphenol and vitamin C, they can be considered as a good source of these components.

Key words: Fruit juice, total polyphenol, vitamin C, health

INTRODUCTION

Current recommendation of health experts is to increase the consumption of fruits since there is convincing evidences linking a diet rich in fruits with reduced incidence of coronary heart disease, cancer and various age relating chronic diseases (Margetts and Buttriss, 2003). These protective effects are hypothesized to owe, at least in part, to antioxidant and anti-proliferative effects of various polyphenols and vitamins such as vitamin C that present in fruits and their products (Collins, 1999; Strain and Benzie, 1999).

Polyphenols are a group of secondary metabolites widely distributed in the medicinal plants, vegetables, fruit and a variety of beverages such as tea, wine and fruit juices. These metabolites are the most abundant antioxidants in human diets (Scalbert *et al*., 2005) and recently receiving increasing interest from consumers, manufacturers and food marketing for their health benefits (Scalbert and Williamson, 2000).

Vitamin C is an essential phytonutrients for the metabolism of living cells that occurs in different concentrations in natural foods especially fruits and their products. It is considered as the major antioxidant in the diet. Gardner *et al*. (2000) have revealed that Vitamin C accounted for 65-100% of antioxidant capacity of citrus juices.

Juices besides fruits are suitable food products in term of ingestion of health protective phytochemicals (Netzel *et al*., 2002). The bioactive components may even be better absorbed from juices than from plant tissues (Bitscha *et al*., 2001) and also the consumption of fruit juices help fulfil the recommended fruit servings (Dennison, 1996). Currently the American Academy of Pediatrics issued recommendations for natural fresh fruit juice consumption for children and adolescents (American Academy of Pediatrics, 2001) for achieving recommended intakes of important nutrients such as vitamin C, folate and magnesium (Ballew *et al*., 2000). In recent years the consumption of fruit juices increased at very quick rates (Kabasakalis *et al*., 2000) but rapid growth in the commercial fruit juices and fruit drinks production propel public to consume these types of juices instead of fresh and natural juices. Rampersaud *et al*. (2003) showed that at around age seven, children's consumption of natural real juice flat-lines, while intake of fruit-flavoured beverages increases.

Due to the great importance of polyphenols and vitamin C in human health and also because of the growth in commercial fruit juices production and consumption, this study was carried out with aim of determination and comparison of the polyphenol and vitamin C contents of natural fresh and commercial fruit juices.

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MATERIALS AND METHODS

Sample preparation: In Iran, fruit juices are generally consumed in two types of natural fresh and commercial packaged fruit juices. To achieve the objectives of present study, three bottles of 33 (total = 99) commercial fruit juices from 3 different brands and 11 different type of fruit juices were collected from local stores in Tabriz-Iran between July and October 2008. Three different samples of 11 different types of fresh fruits (total = 33) were prepared by squeezing juice out of the fruits. All fruits were thoroughly washed with tap water and any fruit with signs of defect and immaturity sorted out in the laboratory. The juices were extracted from fruits by food processor. Fruit juices having fibre content were filtered before the measurements.

All reagents and solvents used were of analytical grade and were used without further purification. Doubly distilled water was used throughout. Terbium (III) chloride hexa-hydrate ($TbCl_3$-$6H_2O$) was from Acros Organics, USA. Quercetin was obtained from Sigma. All other materials purchased from Merck.

Total polyphenol assay: The total polyphenol contents of fruit juices were determined by using Folin-Ciocalteu method (Singleton *et al.*, 1965). Briefly 1 ml of extract or standard solution of gallic acid (20, 40, 60, 80 and 100 mg/l) was decanted in 25 ml volumetric flask which containing 9 ml of distilled deionized water. 1 ml of Folin-Ciocalteu reagent was added to the mixture and shaken. After 5 min, 10 ml of 7% Na_2CO_3 solution was added and the solution was dilute to volume with distilled deionized H_2O and mixed. After incubation for 90 min at room temperature, the absorbance against prepared reagent blank (distilled deionized H_2O) was measures at wavelength of 750 nm. Total polyphenol contents of fruit juices were reported as mg Gallic Acid Equivalent (GAE)/100 ml.

Vitamin C assay: Vitamin C contents of fruit juices were determined spectrophotometrically by metaphosphoric acid extraction of 2,6-dichlorophenol indophenol dye (Robinson and Stotz, 1945) using a Cecil spectrophotometer (model 8000) in wavelength of 500 nm with a 1 cm quartz cell. The vitamin C contents of fruit juices were reported as mg/100 ml.

Statistical analysis: Parameters were expressed as mean and standard deviation. Significant differences between the group of commercial and natural fresh fruit juices were calculated by Independent t-test. The ANOVA test with Tukey post Hoc test was used to compare the total polyphenol and vitamin C contents of different natural fresh and commercial fruit juices. P-values of less than 0.05 were considered statistically significant.

RESULTS

Total polyphenol contents of fresh fruit juices presented in Table 1. The mean polyphenol contents of fresh juices (90.49±0.2 mg GAE/100 ml) were significantly higher than commercial fruit juices (79.82±0.3 mg GAE/100 ml) ($p<0.001$).

The total polyphenol contents of fresh fruit juices were ranging from 23.75±0.1 (apricot juices) to 421.42±0.5 (pomegranate juice) mg GAE/100 ml while those of commercial fruit juices were ranging from 18.57±0.4 (apricot juice) to 381.91±0.4 mg GAE/100 ml (pomegranate juice). Considering the wide variation in the total polyphenol contents of both fresh and commercial fruit juices, they were divided into two groups of high polyphenol contents (>100 mg GAE 100 ml) including pomegranate and red grape juices and low polyphenol contents (<100 mg GAE/100 ml) including all other fruit juices.

Table 2 shows the vitamin C contents of different natural fresh and commercial fruit juices. The mean vitamin C contents of fresh fruit juices (17.44±1.06 mg/100 ml) were reasonably higher than those of commercial (15.01±1.2 mg/100 ml) ones ($p<0.05$). The vitamin C contents of natural fresh fruit juices were higher than those of commercials except for peach and apricot juices however the differences between the vitamin C contents of peach and pineapple juices from natural and commercial sources were not statistically significant. It has been suggested that pasteurization can decrease the vitamin C content of juices. Klopotek *et al.* showed that the vitamin C contents of strawberry juices decrease 35% by pasteurization. In another study, Taoukis et al. revealed that in the case of pineapple juices, the loss of vitamin C in 45^{o} C, was up to 25%.

In the case of fresh juices, the vitamin C contents were ranging from 14.31±0.2 (apricot juice) to 24.51±0.15 (orange juice) mg/100 ml. The statistical analysis indicated that the vitamin C contents of orange juice was significantly higher than all other fruit juices ($p = 0.000$).

DISCUSSION

All fresh fruit juices had higher polyphenol content than commercial ones and only in the case of pineapple the difference were not statistically significant. Different factors such as processing techniques, clarification and pasteurization can affect polyphenol contents of commercial juices. Hertog *et al.* (1992) and Shadidi and Nazck (1995) had shown that polyphenol contents could be affected by different processing techniques. According to Ritter *et al.* (1992) and Karadeniz and Eksi (2001) reports, clarification also could decrease the polyphenolic contents of commercial fruit juices, furthermore klopotek and collegeuse revealed that pasteurization had influence (-27% decrease) on the polyphenol contents.

Table 1: Mean±SD of polyphenol contents of different types of natural fresh and commercial fruit juices

	PC (mg GAE. 100 ml^{-1})		
Fruit juice	Natural fresh juices	Commercial juices	p-value
Total mean	90.49±0.2	79.82±0.3	<0.001
Pomegranate	421.42±0.51	381.73±0.43	<0.001
Red grape	144.56±0.64	135.2±0.36	<0.001
Sour cherry	67.29±0.26	64.28±0.51	0.001
Peach	58.29±0.39	50.05±0.32	<0.001
Mango	56.72±0.25	28.57±0.23	<0.001
Orange	54.28±0.33	42.85±0.14	<0.001
White grape	37.69±0.38	33.71±0.18	<0.001
Pineapple	36.16±0.50	35.74±0.32	0.28
Red grape fruit	49.4±0.33	41.81±0.34	<0.001
Apple	45.38±0.41	42.81±0.25	<0.001
Apricot	23.75±0.11	18.57±0.41	<0.001

All analytical data are the mean of triplicate measurements of three independent samples ±SD. *p-value of independent t-test.
PC = Polyphenol content (mg GAE.100 ml^{-1})

Table 2: Mean±SD of vitamin C contents of different types of natural fresh and commercial fruit juices

	Vitamin C content (mg. 100 ml^{-1})		
Fruit juice	Natural fresh juices	Commercial juices	p-value
Total mean	17.44±1.06	15.01±1.2	0.05
Orange	24.51±0.15	15.86±0.62	<0.001
Red grape fruit	23.2±0.23	14.81±0.31	<0.001
Pomegranate	19.01±0.15	17.34±0.23	<0.001
Apple	17.45±0.15	13.4±0.77	<0.001
Sour cherry	17.36±0.13	16.44±0.42	0.005
Red grape	16.75±0.12	15.18±0.76	0.005
Pineapple	15.46±0.17	13.6±0.21	<0.001
White grape	14.79±0.06	14.13±1.31	0.63
Mango	14.65±0.15	12.57±0.02	<0.001
Peach	14.42±0.33	15.63±0.56	0.005
Apricot	14.31±0.21	16.2±0.15	<0.001

All analytical data are the mean of triplicate measurements of three independent samples ±SD. *p-value of independent t-test

In the case of polyphenol contents of fresh fruit juices, our findings were partly similar to the results of Loots *et al.* (2006) who showed that the total polyphenol contents of orange and grape juices were 36.4±9 and 26.9±1.4 mg GAE/100 ml respectively. As reported by other researchers, the polyphenol contents of red grape was higher than those of white grapes that may be due to high concentration of anthocyanin in dark fruits (93,97). Comparison of our results with Gaedener *et al.* (2000) report had revealed that the polyphenol contents of orange and grapefruit juices in our study were lower while in the case of apple and pineapple juices, it was higher than mentioned study's results. The observed variations between the total polyphenol contents of different fruit juices from different studies may be due to differences in varieties, climate, ripeness, extraction method, analytical procedure employed, etc (Vasco *et al.*, 2008). For example in white coloured fruits the polyphenol contents decreases constantly with the progress of the ripening, while in red coloured varieties it increases during the last ripening stage due to the maximal accumulation of anthocyanidines and flavonols (Marinova *et al.*, 2005).

Comparison of total polyphenol contents of commercial fruit juices in present study with the results of Lugasi and Hovari (2003) from hungry showed that the polyphenol content of red grape juice in present study was higher (135.2±0.3 vs 68-98 mg/l00ml) while for pineapple juice it was lower (35.74±0.3 vs. 67.4 mg GAE/100 ml).

The polyphenol contents of commercial fruit juices in the case of pineapple, orange and mango juices in present study were higher than those of Thai beverages, reported by Abdullakasim *et al.* (2007).

The variations between the polyphenol contents of commercial fruit juices in different studies may be due to various factors such as different variety of fruits that used for juice production, the percent of pure juices in final product, squeezing and pasteurization techniques. Various processing techniques in commercial field and the methods of analysis as well may affect the polyphenol content.

Because of the lake of comprehensive data on vitamin C contents of fresh fruit juices, we compared our results with the vitamin C contents of fresh fruits. Comparison of our results with United States Department of Agriculture (USDA) database for standard references (USDA, 1998) clarify that the vitamin C contents of natural fresh fruit juices in our study were less than fruits from USA.

The vitamin C contents of our fruit juices in the case of pomegranate and grapefruit were more than and in the case of orange was less than Pakistani fruits (Iqbal *et al.*, 2006). The observed differences in the contents of vitamin C may be as a result of differences in maturity stage and regional varieties of fruits. It has been reported that the amount of vitamin could even vary between different samples of the same species (Davidson, 1979). Different techniques of measuring and squeezing process may also affect the vitamin C contents of fruit juices (Gil-Izquierdo *et al.*, 2002).

The vitamin C contents of commercial fruit juices were ranging from 12.57±0.02 mg/100 ml to 17.34±0.2 mg/100 ml. The lowest amount of vitamin C was in mango and the highest amount was in pomegranate juices.

Kabasakalis *et al.* (2000) reported that the ascorbic acid contents of orange juice were 42.4 mg/100 ml and grapefruit juice was 43.4 mg/100 ml. The vitamin C contents of commercial pure orange juices immediately after production in Klimczak *et al.* (2007) study was in the range of 36.15-40.85 mg/100 m. Massaioli and Haddad (1981) also have reported the vitamin C contents of commercial orange juices in the range of 27.5-73.11

mg/100 ml. Comparison of our data with these studies indicated that the vitamin C contents of orange juice in these studies were higher than those of our commercial juices. it seems that synthetically added vitamin C to the commercial fruit juices is the major determinant of this vitamin in these juices however it must imply that natural vitamin C is superior to synthetic one.

Conclusion: Although the mean total polyphenol and vitamin C contents of fresh fruit juices were higher than those of commercial ones, as a good source of biologically active compounds such as fibre and total polyphenol compounds, both fresh and commercial fruit juices are so effective in enhancing health status and could be considered as a good replacement to carbonated soft drinks. It seems that until the active components of fruits and their products are clearly established, measuring their total polyphenol and vitamin C contents may be useful in planning diets for health promotion.

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