## DETERMINATION OF FOLIC ACID LEVEL IN SOME FOODSTUFFS

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Folic acid, which is a B group vitamin, is usually found in the tissues of plants and animals as conjugates containing one or more forms [1]. It is either synthesized by microorganisms together with pteridins in the digestive system from paraaminobenzoic acid taken with foods, or directly taken from foods [2, 3].

UDC 547.915

Folic acid plays a role in biosynthesis of methionine, purine, and pyrimidine [3, 4] and is required for conversion of serine and glycine in amino acid metabolism [2, 5]. Folate deficiency increases plasma homocysteine concentrations and promotes oxidative stress in rat liver [6]. Supplementation with high-dose folate significantly reduces plasma homocysteine in patients with and without atherosclerosis [7].

Folic acid supplementation may help to prevent ischemic heart disease and strokes, and possibly several cancers (cervical, bronchial, colon, breast) [8–11]. It is also essential for efficient DNA synthesis and repair [12].

A sufficient level of folic acid is needed in early pregnancy to prevent neural tube defects [13]. So the present study was aimed at determining the levels of free folic acid in some foods.

Free folic acid (FFA) levels for various vegetables, fruits, and animal foods are given in Table 1.

Assuming that only 1/4 of folate derivates is taken as FFA per day, according to the consumption standard, it is stated that 400  $\mu$ g folic acid per day is sufficient for a mature person [14]. This value is stated as 800  $\mu$ g during pregnancy and 500  $\mu$ g during lactation [15]. The minimum requirement for folic acid during higher growing stages is stated as 500  $\mu$ g per kilogram [14].

As seen in Table 1, potato has the lowest free folic acid in the examined vegetables. The free folic acid level in other examined fresh vegetables ranges from  $140\pm20$  ng/g to  $3900\pm205$  ng/g. The amounts of FFA are found about  $133\pm17$  ng/g in dried onion and  $7300\pm270$  ng/g in dried mint. The reason for the higher folic acid level in dried mint as compared to fresh mint is the higher water loss than folic acid during the drying process.

Apples have the lowest level of FFA ( $25\pm6$  ng/g) among the examined fresh fruits. The FFA level in all other examined fresh fruits ranges from  $75\pm11$  ng/g to  $432\pm39$  ng/g. In dried fruits it ranges between  $510\pm32$  and  $1290\pm165$  ng/g, while for animal foods, it ranges between  $16\pm4$  ng/g and  $2750\pm184$  ng/g. In general, the FFA levels in our samples are in compliance with the results of Muller [16] and Baysal [17]. The differences in folic acid levels of some samples examined are caused by different ecological and packing conditions for these foods. FFA is usually present as a monoglutamate in animal foods, and as a polyglutamate in vegetable foods. Both forms are absorbed in the small intestines after hydrolysis by conjugase enzyme [17]. However, FFA can be directly absorbed by the small intentines. Folic acid deficiency can happen in some intestinal diseases because it is not hydrolyzed. In addition, folic acid requirement increases during infancy, childhood, pregnancy, and lactation. It become very important to eat foods rich with folic acid, in view of the association of folic acid with some diseases as discussed previously.

Dried mint is also rather rich in folic acid and is recommended as a source. Most vegetables, fruits, and animal products have enough folic acid to supply the daily requirement if consumed as a balanced diet. The consumption of raw vegetables as salads is advisable, since folic acid is heat sensitive. In addition, it is desirable to consume the water used in cooking because most of folic acid passes into it during the cooking process.

In conclusion, it is recommended to consume salad with fresh mint in summer and dried mint tea in winter. The best way to avoid folic acid deficiency is to consume a balanced diet of various foods, especially green vegetables. Our results also show why some plants are important for the treatment of some diseases.

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TABLE 1. Free Folic Acid Levels of Various Vegetables, Fruits, and Some Animal Foods

Foods	Folic acid, ng/g	Foods	Folic acid, ng/g	Foods	Folic acid, ng/g	Foods	Folic acid, ng/g
Potato*	108±15.0	Marrow**	235±25.0	Spinach*	658±44.0	Hazelnut**	780±48.0
Dried onion*	133±17.0	Orange**	260±27.0	Fresh mint*	3900±205	Walnut**	970±57.0
Carrot*	140±20.0	Cucumber**	280±30.0	Dried mint*	7300±270.0	Peanut**	1100±98.0
Green onion leaf*	205±26.0	Lemon**	305±33.0	Apple**	25±6.0	Lentil**	1170±145.0
Cabbage*	297±32.0	Strawberry**	360±34.0	Apricot**	75±11.0	Dry bean**	1290±165.0
Lettuce*	395±34.0	Green bean**	432±39.0	Plum**	80±13.0	Milk***	16±4.0
Celery*	460±37.0	Wheat**	510±32.0	Banana**	100±15.0	Yogurt***	160±24.0
Cauliflower*	560±40.0	Corn**	580±35.0	Tomato**	$140{\pm}17.0$	Cheese***	193±16.0
Rhubarb*	587±43.0	Almond**	610±41.0	Green pepper**	155±19.0	Egg yolk***	645±50.0
Parsley*	640±40.0	Chick pea**	630±40.0	Aubergine**	165±22.0	Liver***	2750±184.0

\*Vegetable; \*\*Fruit; \*\*\*Animal product.

Results are mean values  $\pm$  SD of three replicated analyses.

In this study, various vegetables, fruits, and animal products were used as material (See Table 1). Fresh food materials were immediately analyzed. For analyses, 1.5-3 g of food samples was taken in a PVC tube and 0.5 mL 1 M HClO<sub>4</sub> and 0.1 mL 0.5% ascorbic acid mixtures were added in each tube for protecting folic acid coenzyme against oxidation [2]. Then 6–7 mL of double distilled water was added and the whole was heated at 80°C for 10 min. The samples were cooled to room temperature (24–25°C). Then they were made up to 10 mL with double distilled water and centrifuged at 4000 rpm for 10 min, and then a 20 µL sample was taken from the tubes and injected into the HPLC column. Folic acid levels in samples were determined by using the mobile phase consisting of mixing of 0.5 M ammonium acetate and acetonitrile (4:1; v/v) with 1 mL/min; flow speed in C18 column (25 cm, 3.9 mm ID, 0.25 inch OD, Techopak); 280 nm wavelength [16, 18, 19].

Results are mean values  $\pm$  SD of three replicated analyses.

All the chemical reagents used in the analysis were analytical grade and obtained from Merck (Darmstadt, Germany). Double distilled water was used throughout the work. HPLC was performed with the CECIL-1100 and UV detector (Cambridge, England).

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