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Review article

FUNCTIONAL FOOD: RARE HERBS, SEEDS AND VEGETABLE OILS AS SOURCES OF FLAVORS AND PHYTOSTEROLS

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Abstract. Two plant species of the genus *Anthriscus (A. sylvestris* and *A. cerefolium) and Laserpitium latifolium* L. are described as a good source of new aroma constituents and phytosterols. These plants are the herbs widely distributed in Serbia and possess significant medicinal value. They are widely applied in traditional medicine, but they are not used as functional food or in food technology. Two well-known domestic aromatic plant species: *Mentha piperita* L. and *Thymus vulgaris* L. and the lesser known *Alliaria officinalis* Andrz. are described as a good sources of highly prized essential oils in several wild-growing herbs. They are traditionally used in cooking.

Quinoa seeds considered as multipurpose agro-industrial crop and the seeds may be utilized for human food and in flour products and in animal feed stocks because of its high nutritive value. Furthermore, watermelon meal possesses good nutritional quality, which may be commercially used as a new feed in the human or in the cattle nutrition. Seed oils, from *Rubus ideaus*, *Ribes nigrum* and walnut are also presented. The chemical composition on the sterols and essential fatty acids content, of these seed oils are shown that some of them can be successfully used in human nutrition, under specific conditions.

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Finally, developing new cholesterol-reducing products, such as phytosterols and their esters, which are marked as cholesterol–reducing food ingredient as well as the information on its incorporation into foods, are also included.

Key words: phytosterols, herbs, seed oils, sources, and health benefits.

Introduction

Because of the importance of functional food factors for the nutrition of humans and animals, an increasing scientific interest has emerged in health related effects occurring as a consequence of their content in diet. Therefore, in this paper we tried to present the state of the art concerning nutritional aspects, as well as health benefits of the some edible seed oils and rare medicinal herbs. Furthermore, we reviewed alternative sources of sterols as well as their occurrence in domestic well-known and rare seeds, seed oils, and medicinal herbs.

To achieve commercial success, the foremost task will be needed to develop an integrated approach to optimize the collection and cultivation of the raw plant materials. Also, there will be a need to develop process technologies for the very effective utilization of the lesser-known plants in edible and industrial sector.

Sources of phytosterols in edible vegetable oils

Phytosterols, the class of sterols are found in the fat-soluble membrane extract of the plants, including algae. Phytosterols are not synthesized endogenously in humans. Common known sources are vegetables, wood and vegetable oils. Over 250 naturally occurring sterols have been found in the higher plants. As many as 60 sterols have been described, in corn. Despite this diversity, the most frequently occurring phytosterols in nature and in human diets include β-sitosterol, campesterol, stigmasterol and avenasterols (Yankah, V.V., 2001-c). The function of phytosterols in plants is primarily related to their ability to affect membrane fluidity and water permeability. They also act as plant hormones and hormonal precursors.

The difference in composition and content of sterols in edible plant oils can be useful for the quality identification. In the following table 1 the phytosterols content are listed.

Tab. 1. – Sterols composition in an edible oil (mg/kg) *

Phytosterol	Sunflower seed oil	Rape seed Oil (Canola)	Soybean oil	Corn-germ oil	Arashid oil	Palm oil
Cholesterol	≤ 7	0.5-1.3	0.6-1.4	0.2-0.6	< 3.8	2.6-6.7
Brassicasterol	< 0.2	5.0-13.0	< 0.3	< 0.2	< 0.2	/
Campesterol	7.4-12.9	24.7-38.6	15.8-24.2	18.6-24.1	12.0-19.8	18.7-27.5
Stigmasterol	8.0-11.5	< 0.7	14.9-19.1	4.3-7.7	5.4-13.2	8.5-13.9
β-sitosterol	56.2-65.0	45.1-57.9	51.7-57.6	54.8-66.6	47.4-64.7	50.2-62.1
Δ-5-avenasterol	< 6.9	3.1-6.6	54.8-66.6	4.2-8.2	8.3-18.8	< 2.8
Δ-7-stigmasterol	7.0-24.0	< 1.3	4.2-8.2	1.0-4.2	< 5.1	0.2-2.4
Δ-7-avenasterol	3.1-6.5	< 0.8	1.0-4.2	0.7-2.7	< 5.5	< 5.1
The rest (others)	< 5.3	< 4.2	0.7-2.7	< 2.4	< 1.4	/
Total sterols (mg/kg of oil) content	24.37-4545	4824-11276	1837-4089	7950-22150	901-2954	376-617

* Sources: Official gazette SRJ No.54/1999. Regulations of quality and demand for edible oils, lards, margarine and others lipid products.

On the market, there are a lot of food products which are enriched by plant sterols, especially margarines and the other fat products (Law, M. 2000).

Domestic plants as new sources of phytosterols Herbs

Two well-known domestic aromatic plant species: *Mentha piperita* L. and *Thymus vulgaris* L. and the lesser known *Alliaria officinalis* Andrz. are described as a good sources of wide spread phytosterols, and highly prized essential oils in several wild-growing herbs. They are traditionally used in cooking. In addition, plant origin *Alliaria officinalis* Andrz. is presented as a significant garlic flavor vegetable. Plant species of the genus *Anthriscus (A. sylvestris* and *A. cerefolium)* and *Lasepitium latifolim* of the family Apiaceae, are presented as a sources of new aroma constituent, phenolics and phytosterols. These plants are the medicinal herbs widely distributed in Serbia and possess significant medicinal value. They are widely applied in traditional medicine, but they are not used as food additive or in food technology as new herbs or vegetables.

A.sylvestris **(L.) Hoffm., (family Apiaceae),** is a wild growing plant in Serbia. The essential oil obtained from the herb is widely used in flavoring foods, beverages and in perfumery. In traditional medicine water extracts of

the flower are used as diuretics and tonics, the wet nurses have used water extracts of the whole plant and powder has been used as a spice in salad dressing (Josifovic, M., 1975). The chemistry of *A. sylvestris* has been extensively studied. The occurrences of constituents, not previously identified in *A. sylvestris* are reported, such as flavonoids: apigenin, quercetin and its glucoside namely rutin. Finally, monocyclic compounds: *p*-cymene and eugenol (2-methoxy-4-allyl-phenol), pentacyclic triterpene namely quinovic acid, among the other constituents such as cresols and stigmasterols, in a high amounts are also observed (Milovanovic, M., 1996). The results indicate that eugenol is probably the predominant flavor of the plant, which is widely used as a spice in food as good substitute for the clove aroma.

Anthriscus cerfolium **(L.) Hoffm.** is a culinary herb, which is used as a newly commercialized vegetable in China, as for instance dietary fiber in human diets (Kurosava, S. et al., 1992). The Serbian plant has also been used in folk medicine as diuretic, tonic and as a spice as well as salad dressing (Josifovic, M., 1975). Our studies indicate that the specimen does not contain photoactive furocoumarine compounds. Instead, the plant tissue contains eugenol, stigmasterols, β-sitosterol and well-known flavones: luteolin and quercetin. These facts suggest that plant species *A. cerefolium* is a good source of phytosterols, as well as newly aroma constituents, which may be useful in human nutrition (Milovanovic, M., 1997).

Laserpitium Latifolium L. **(family Apiaceae)**, collected in Serbia has identical chemical constituents of the sesquiterpene lactones and phytosteroles as the Czech plant species (Djermanovic, et al., 1995). As active principles, sesquiterpene lactones possess a broad spectrum of pharmaceutical activities, including cardiotonic, anestetic, antitumor and other useful biological properties. Because of its wide application in folk medicine, as diuretic, and a perfect remedy against headache, also the chemistry of these herbs has been extensively studied. There is no data about using as food additive in human nutrition.

Thymus vulgaris **L. (Lamiaceae)**. There are a lot of references by which this herb is described. Because of its wide applications in folk medicine and as spice in food industry, the chemistry of this plant species has been extensively studied for many years ago. We reported the antioxidant potential of domestic plant issue and describe the compounds, which are responsible for the significant activity (Picuric-Jovanovic, K., et al., 1995). Thus, the more thought investigation of this plant seemed to be appropriate.

***Mentha piperita* L. (Lamiaceae)**, as far as traditional medicine is concerned it has been used as a stimulant and spasmolitic and as a spice for flavoring in food technology and especially in cosmetic industry. We have investigated the chemical composition and variation of the sterols and essential oils constituents of the several herbs, obtained from the Russia and Serbia regions, and compare with the Pharmacopoeia demands. It was evidence that the quality and amount of the obtained oils are in direct function of the locality and that there is no morphological difference between the samples (Picuric-Jovanovic, K., et al. 1997).

Plant species ***Alliaria officinalis* Andrz. (Brassiaceae)**, syn Erysimum alliaria L. is used as food additive. It is known, however, that the highest concentration and quality of essential oils in the seeds and the oil obtained from it is widely used for flavoring as substitute for *Sinapis alba* L. (Brassiaceae) in mustard making (Tucakov, J., 1996) This herb has a respectively and strong garlic aroma and showed mild antioxidant activity against the lipid oxidation (Milovanovic, M. et al., 1999).

Seeds ad seed oils

Quinoa seeds (*Chenopodium quinoa* Wild.). Quinoa is highly nutritious product, being cultivated for several thousands years in South America. Quinoa seeds posses a great nutritional importance, as its protein content, making it a healthy choice for vegetarians. Unlike wheat or rice, the quinoa seeds contain a balanced set of essential amino acids for humans, making it an unusually complete protein source. Other positive aspects are the saponins found in the seed hull and the lack of gluten. Saponins may be interesting as potential insecticides, fungicides, and to the pharmaceutical industry as a mediator intestinal permeability, which could aid the absorption of specific drugs, and for reducing the level of cholesterol. The main uses of quinoa are for cooking, baking, etc.; various product for people allergic to gluten, animal feed, green fodder and pellets, modified food products as breakfast cereals and cookies. (Jacobsen, S.E., 2003)

Quinoa is considered a multipurpose agro-industrial crop (Galwey, N.W., 1993), and has been selected by FAO as one of the crops destined to offer food security in the next years (F A O, 1998).

(***Citrullus colocynthis* L**), with traditional name watermelon, is very popular fruit in Serbia. Melon seeds or meal are source of high quality proteins and edible oil used in cooking in human and cattle nutrition in some African and Middle Eastern American countries. The meal from melon

seeds can be made into patties and served as a meat substitute (Akobundu, E. et al., 1982). Unfortunately, according to the literature data there is no any information of domestic sample, about medicinal values of the seeds or seed oil applied for cooking and frying or as useful product with good nutritional value. Recently, it was reported that watermelons are a good sources of lycopene, which is considered to be a powerful antioxidant. It was found that some varieties have as much or more lycopene than the raw tomatoes. The meal, of the domestic melon seeds, contains a high percentage of crude proteins and oil, e.g. 21% and 22% respectively. The melon kernels had especially high content 38% of protein and 52% of oil. The seed oil contains linolenic acid, as a major fatty acid in 62%. The seed oil composition of the domestic watermelon may help in the selection of melon meal for future commercial production in human diet and also for animal and poultry feed or protein production (Milovanovic et al., 2006).

The ***Rubus idaeus* L.** fruit, family **Rosaceae**, with traditional name raspberry, is a native floral element of central and of eastern European regions, but in some cases can be also found in the East Asia and North America. Indeed, *Rubus idaeus* qualitative fruit is extensively used for preparing of juices and syrups, namely Sirupus rubi idaei. Especially, the fruit generally have high content of C-vitamine (12-15mg/%), polyphenolcs, sterols and organic acids, i.e., citric and malic acids (Tucakov, J., 1996). The fruit juice of *R. idaeus* is used in a traditional medicine because of its cardio-tonic properties, for the improvement of immune injury system and in the treatment against of the constipation. In addition, leaves tea is also taken as anti diaroic, as well as astringent for inflammation in the treatment for chest colds and bronchial problems (Serbian Academy of Sciences and Arts, 1989). Moreover, there are a lot chemical constituent studies, which have been made on the carbohydrates, organic acids, vitamins, sterols and essential oils from the fruit and the tannins and flavonoids content of the leaves.

No medicinal or folk use has been reported for this seed oil. *Rubus idaeus* seed oil, of domestic fruit contains 26.4 % α-linoleic acid (ALA). This oil probably has tendency to oxidize because of high content of polyunsaturated fatty acids (PUFA), thus it must be stored under cold oxygen-free and light-free conditions. The total tocopherol content of *Rubus ideaus* seed oil had especially high value, and γ-tocopherol was the major constituent detected in the seed oil sample. Obviously, the high content of the total tocopherols, especially the yield of γ-tocopherol, probably may

protect the seed oil from expected oxidation according to the high degree of PUFA. However *R. idaeus* can not use as a commodity seed in the production of edible oil, but can be useful as a medicinal or pharmaceutical product in consideration that this oil is a rich, relatively pure source of ALA (Picuric-Jovanovic, K., 2000).

The ***Ribes nigrum*** **L**. plant, family **Grossulariaceae**, is a native of east Europe and wide spread in the Mediterranean region. In folk medicine the fruit juice of *R.nigrum* is used in the treatment of indigestion, coughs, colds and as a diuretic and especially the fruit is valued for its dietary fibber content in human diets. This fruits so-called blueberry, on the market is highly prized as a source of γ-linolenic (GLA) acid. Because of its high contents of γ-linolenic acid and total tocopherols, *R.nigrum* seed oil is used as a basic component of creams for acne and eczema in cosmetic industry. GLA is a pre-essential fatty acid in humans and other mammals. Thus, GLA has been applied directly to treat certain diseases, such as eczema, multiple sclerosis and rheumatoid arthritis. Chemical constituent studies also have been made on the lignin, carbohydrates, vitamins, sterols (stigmasterol, campesterol, avenasterols and so on), suberins, prodelphinidins of fruit and leaves.

Thus, the seed oil composition of domestic *R. nigrum* L. and its oxidative stability were described (Picuric-Jovanovic, K., et al. 2002). Studies on the physical and chemical characteristics of the oil from *Ribes nigrum* indicated that the seeds contained from 8-15% of oil. Results of the analyses of the tested oil for the content and composition of fatty acids showed high level of PUFA, especially GLA (12,6%), and tocopherols, with significance amounts of carotene and chlorophyll. The oil was stable to oxidation.

If this oil is to be used in the cosmetic or pharmaceutical industry alone or as a partial component, then it is imperative that the oxidative stability be maximize. Therefore, *Ribes nigrum* seeds, which are considered as waste samples, may be used as a good and new source of oil, rich in GLA content. However *R. nigrums* seeds can be use as a commodity seed in the production of edible oil and it can be useful as salad dressing, or valuable aroma additive for cooking. Because of the potential medicinal values and the widespread distribution of this plant species the seed oil may be very useful.

Walnut oil. Recently, the physical and chemical characteristics of the several walnut cultivars have been observed as well as their influence on

oxidative stability and nutritive value of the walnut oil (Rabrenovic, B., 2006).

Total oil content, crude protein, carbohydrates, ash and moisture are evaluated. Dominant fatty acids in walnut oil are essential fatty acids as linoleic, linolenic and oleic acids. The most common tocopherol is γ-tocopherol and the dominant phytosterols are β-sitosterol and Δ-5-avenasterol. Oxidative stability of the walnut oil confirmed that this oil is relatively unstable when compared with other common plant oils. The existing essential fatty acids following the tocopherol and sterols, have an influence on the high nutritive value of the oil. For this reason, usage of walnuts oil has an essential role in the modification of lipoproteins profile, as well as protective role in cardiovascular diseases, cancer and in the other similar diseases.

Health benefits of phytosterols

Phytosterols – as reducing agents in cholesterol levels. The role of cholesterol in the body appears to be as a constituent of membranes, and also is the precursor of synthesis for steroids and bile acids. Hyperlipidemia and associated disease states of obesity, arteriosclerosis and hypertension are considered to be risk factors in the development of cardiovascular disease. The primary focus on the health is on low-density lipoprotein, cholesterol levels. The contribution of the low-density cholesterol, deposition in the arterial wall is well established. Phytosterols are the molecules structurally similar to cholesterol. The effect of diet, on modifying the risk of the certain diseases has been known for long time. Phytosterols in foods were identified as natural cholesterol-lowering agents in the middle of the last century. However, the development of using pharmaceutical drugs preferable as cholesterol-lowering therapies compared to phytosterols. Through scientific research, the understanding the relationships between the foods, physiological function and disease has progressed over past 20 years. Diets containing phytosterols at levels of 2-3g/day can reduced total cholesterol and low-density cholesterol levels to about 10 -15%, respectively.

Marketing agreements set for phytosterols products. Agreements reached in United States and in Europe should expand the number of foods containing cholesterol–reducing phytosterols and their esters available to consumers. A biotechnology company, which formulates novel dietary components, has developed the technology that will allow phytosterols -

which have been shown to reduce cholesterol levels - to be added to more foods. This new technology will allow incorporation of phytosterol into food products such as beverages, yogurts, and low-fat foods according to USDA (US Department of Agriculture, Food and Nutrition Service; www.fns.usda.gov/fns). In Europe, some food processing companies have agreed to cooperate in marketing and developing new cholesterol-reducing products, such as phytosterol Benecol (phytosterols from wood pulp) and its stanol ester, which is marketed as cholesterol–reducing food ingredient as well as the information on **its** incorporation into foods. Phytrol, a mixture of unesterified phytosterols, isolated from coniferous trees is exclusively licensed for use as functional food ingredient and dietary supplement (Y a n k a h, V. 2001-a). On the contrary, FDA (Food and Drug Administration) did not include unesterified sterols and stanols in the agency's final rule on a health claim for the cholesterol-lowering effects of sterols and stanols in foods and did not allow the statement that such products might lower the risk of coronary artery disease when integrated into diet that is low in saturated fat and cholesterol.

Low cholesterol in causes of cancer. The connection links low cholesterol to higher risk of developing and dying from cancer. Is it good for us? The official line on low cholesterol is that cancer causes the cholesterol drop. It may be that whatever genes cause low cholesterol also lead to the higher risk of cancer. It is possible that people who have low cholesterol get less heart disease and, therefore, is more likely to get cancer (the explanation comes from the Dept. of Nutrition and Food Science at Wayne State University in Detroit, Michigan). Epidemiological studies on membrane structure and function on tumor host tissue, programmed cell death, immune function and cholesterol metabolism suggest that dietary phytosterols may offer protection from some common cancers, including colon, breast, and prostate cancer (A w a d, A.B. 2000).

Phytosterols and other diseases. Phytosterols and especially sterols from the kernels oils: such as olive and nuts (walnut), wheat, pumpkin are investigated in reduction of risks in the cardiovascular disease and might lower the risk of coronary artery disease when integrated into diet that is low in saturated fat and cholesterol. Mechanisms of action of plant sterols of inhibition of cholesterol absorption are observed (J o n e s, P.J.H et al., 2000, and H e i n e m a n n, T. et al., 1991). Low plant sterols diet also is implicated in a host of emotional and behavioral problems, including depression.

The consumption of the low energy lipid products such as different margarine is useful in the hipercholeolemic therapy (Maki, K.C., et al., 2001).

Skin protection. Plant oil produced from the seeds, fruit kernel and vegetable, are rich source of fatty acids, which provide an excellent skin protection from drying effects of wind and sun, an eczema, skin eruption and so on. In addition to these materials one finds the other compounds perhaps after more than simple protection. These compounds are phytosterols, and are included: β-sitosterol, campesterol, stigmasterol, brassicasterol, Δ-5-avenasterol and other unidentified sterols (Leung, A.Y., 1980, and Dweck, C.A., 2003). Furthermore, the naturally occurring phytosterols, a tremendous similarity to synthetic compounds; corticosterone and hydrocortisone, traditionally used in allopathic medicine for their anti-inflammatory effects and particularly for the skin eruption, eczema and other erythemic conditions (Dweck, C.A., 2003).

Nutritional studies. The amounts of the plant sterols present in diets are recommended on the treatment of hypercholesterolemia . In order to use the claim, the food product must contain at least 0.65 grams of vegetable oil sterol esters per serving for a total of 1.3 g/day (at two servings), or 1.7 grams of plant stanol esters per serving for a daily total of 3.4 grams (Watkins, C., 2002). Earlier studies on the therapeutic use of phytosterols suggested cholesterol-lowering effect only at high doses of 10-20g/day. In addition the dose-response effect has been reported with the concentrations demonstrating after levels higher than 2.5 grams were consumed. All nutritional reports indicated, that reduction of the cholesterol absorption, is more effective at lower levels, about 1.5-3.0g/day in suspension with the margarines or the other oils (Yankah, V.V., P.H. Jones, 2001-b). For example, 0.74g/day soybean phytosterols mixed in butter (36% fat, 0.4g/day) lowered total and low cholesterol by 10 and 15%, respectively males after 4 weeks. In hypercholesterolemic males 0.6-0.8g/day of phytosterols have been shown to reduce the same components by 4 and 7%, respectively, over 9 weeks.

Conclusion

Flora of the Serbia is very reach in number of plant issues of various botanical families. Only a few of them are cultivated and included for potential exploitation as alternative sources of flavors and sterols with regard to their terpene composition, such as *Mentha piperita* and *Thymus*

vulgaris. It would appear that Serbia enjoys a unique and impressive position in the field of lesser-known herbs. The various kinds of lesser-known plants can help fill Serbian requirement for edible and different healthy food. The two plant species of genus *Anthriscus* are good and new sources of the clove aroma substitute, namely eugenol and different phytosterol compounds, in very high amounts.

Melon seeds and quinoa seeds as meals are both considered to have a bright future in human and animal feeds. Quinoa has the potential to develop as a new major crop in Balkan region due to its strong resistance to a poor environment and desirable nutritional characteristic of seeds, which are high in protein content, rich in essential amino acids and a high content of a range of essential fatty acids, vitamins and minerals. The seed oil composition of the domestic watermelon may help in the selection of melon meal for future commercial production in human diet and also for animal and poultry feed or protein production. *Ribes nigrum* seeds, which are considered as waste samples, may be used as a good and new source of oil, rich in GLA content. However *R. nigrums* seeds can be use as a commodity seed in the production of edible oil and it can be useful as salad dressing, or valuable aroma additive for cooking. Furthermore, *R. idaeus,* as seed may not use in the production of edible oil, but can be useful as a medicinal or pharmaceutical product in consideration that this oil is a rich, relatively pure source of ALA. Walnut oil is rich in tocopherols and sterols content, but not stable to the oxidation, while the oil is not useful and unpredictable in food production.

Health benefits of phytosterols and nutritional values are described. Phytosterols–as reducing agents in cholesterol levels and other diseases is also reported. Diets containing phytosterols at levels of 2-3 g/day can reduced total cholesterol and low-density cholesterol levels to about 10 - 15%, respectively.

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ADITIVI U HRANI: SEME, BILJNA ULJA I RETKE LEKOVITE BILJKE KAO IZVORI FITOSTEROLA I AROMA

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R e z i m e

Opisane su dve biljne vrste roda *Anthriscus (A. sylvestris* and *A. cerefolium)* i biljka *Laserpitium latifolium* L. kao novi izvori specifičnih aroma i fitosterola. Ove lekovite biljke poseduju medicinske vrednosti i veoma su rasprostranjene u flori Srbije. Koriste se u tradicionalnoj medicini, ali ne i kao aditivi u hrani. Dalje, dve dobro poznate biljke *Mentha piperita* L. (nana) i *Thymus vulgaris* L. (majčina dušica), kao i manje poznata *Alliaria officinalis* Andrz. su prikazane kao izvori visoko cenjenih esencijalnih ulja, u nekoliko domaćih divljih sorti. Ove biljke se koriste u kulinarstvu.

Seme kvinoje, predstavlja mnogostruko vrednu agrokulturu, poseduje visoke nutritivne vrednosti, a koristi se kao brašno u humanoj ishrani i kao stočna hrana. Potom, seme lubenice, kao sačma, poseduje dobre nutritvne kvalitete, pa se preporučuje kao nova hrana u humanoj i stočnoj ishrani. Prikazan je sastav retkih biljnih ulja semena maline, crne borovnice i oraha, odredjen je sadržaj sterola i esencijalnih masnih kiselina, kao i uslovi za njihovu primenu u kulinarstvu. Prikazani su zdravstveni efekti prisustva fitosterola u hrani, jer smanjuju nivo slabo rastvornog holesterola, pa se smanjenje rizik od kardiovaskularnih obolenja. Na kraju, dat je i nutritivni značaj i potrebe za dnevnim unosima namirnica bogatih u sadržaju biljnih sterola.

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