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Phytosterols

Review Article

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Abstract

Phytosterols are typically plant origin and cannot be synthesized by human beings, therefore their source in human body originates only from diet. Phytosterols are structurally very similar to cholesterol except that they always contain some substitutions at the C24 position on the sterol side chain. Phytostanol and phytosterol esters are chemically stable, fat type materials and having comparable chemical and physical properties to edible fats and oils. Phytosterols are insoluble in water but are soluble in non-polar solvents, such as hexane, iso-octane and 2-propanol. Level of Plasma phytosterol in mammalian tissues are normally very low due to poor absorption from the intestine and faster excretion from the liver compared to cholesterol. It can be metabolized in the liver of mammals into C21 bile acids instead of the normal C24 bile acids. The health benefits of Phytosterols in animals/humans such as reduction of cholesterol levels with decreased risk of coronary heart diseases, anti-inflammatory activities, induction of apoptosis in cancer cells, disease prevention and treatment. Instead of having beneficial effects on human health they have some drawbacks, such as the food or food products that are fortified with phytosterols are not given to pregnant women because of their safety has not been yet studied. Food authorities of different countries has specified the fortification level of phytosterols in food products. Plant sterol, stanols and their esters has been given GRAS status in United States.

1. Introduction

The term "Cholesterol" was derived from the Greek word "Chole"–meaning bile and "Stereos"–meaning solid, followed by the chemical suffix "–ol" for an alcohol. It is an organic molecule, which is a sterol (or a modified sterol) and comes under the class of lipid molecules (Rapheal J. Ogbe, 2015). They are structural components of the cell membrane; their function includes regulating membrane fluidity and permeability as well as membrane-associated metabolic processes. The term phytosterols refers to more than 200 different compounds and are found in various plants and marine sources (Lindsay D.G, 2000).

Sources of phytosterols include fat-soluble fractions of seeds, roots, stems, branches, leaves and blossoms. They are constituents of both edible and ornamental plants, including herbs, shrubs and trees (Priyanka R. Chaudhari, 2002). Commercially, phytosterols are extracted from vegetable oils, such as soybean oil, rapeseed (canola) oil, sunflower oil or corn oil, or from so-called "tall oil", a by-product of the manufacture of wood pulp. Hydrogenation of phytosterols can result to form phytostanols. Phytosterols and phytostanols are high melting powders (Ferrari R.A., 1997). Both are chemically stable materials, having comparable chemical and physical properties to edible fats and oils. These substances are insoluble in water, but soluble in non-polar solvents, such as hexane, iso-octane and 2-propanol. The esters are also soluble in vegetable fats and oils (Richard Cantril, 2008).

Phytosterols and phytostanols are nonnutritive. Phytosterol has same basic functions in plants as cholesterol in animals. They were first reported to play a regulatory role in serum cholesterol concentration in 1951 and were followed by the numbers of clinical trials to establish that 2.0-2.5g/day (Williams C.L., 1999). In addition to cholesterol lowering effect of phytosterols have several health benefits such as prevention against certain types of cancer like colon, breast and prostate stimulation of immunity and protection of skin. (Jessica J.A Ferguson, 2016).

2. Types of Phytosterols

Phytosterols have been classified into two classes:

1. Sterols, with double bond in the sterol ring, are unsaturated compounds.

2. Stanols, without double bond in the sterol ring, are saturated molecules. Phytosterols which are abundantly present in plants and human diets are sitosterols and campesterols. Stanols are present in plants, but they form only 10% of total dietary phytosterols (Raphael J. Ogbel, 2015).

3. Occurrence of Phytosterols

The richest naturally occurring sources of phytosterols are vegetable oils & their products. They can be present either in the free form or as esters of fatty acid/cinnamic acid or as glycosides. Its bound form is usually hydrolyzed in the small intestine by pancreatic enzymes. Nuts, are rich in phytosterols, are often eaten in smaller amounts, but they have significant contribution to total phytosterol intake (Valsta L.M., 2007) Cereal products, vegetables, fruits and berries, usually are not as rich in phytosterols, may also be significant sources of phytosterols due to their higher intakes (Sudhop T.,2005). Thus, the main source of phytosterols is vegetables oils but smaller amounts are also present in nuts, legumes, grains, cereals, wood pulp and leaves. It has been reported that phytosterols are found in all plant foods but the highest concentrations are found in unrefined plant oils, including vegetables, nuts and olive oils. The human beings are not able to synthesis phytosterols, therefore all phytosterols in human blood and tissues are derived from diet, whereas cholesterol in human blood and tissues is derived from the diet and endogenous cholesterols synthesis. The other sources of phytosterols include the Seeds, whole grains and legumes (Andersson S.W., 2007).

Though sterols and stanols are ubiquitous in the plants world but they are most effective when taken with food, so they are now produced commercially to be added to food. Phytosterol are available under various trade names such as Benecol® and Flora pro.active®. Plant sterols or stanols are esterified by creating an ester bond between a fatty acid and the sterol or stanol are known as plant sterol or stanol esters. The process of esterification makes plant sterols and stanols more fat-soluble, so they can easily be incorporated into fat-containing foods, including margarines and salad dressing (Awed A.B., 2001). The most common phytosterols in human diet are β-Sitosterol, Campesterol and Stigmasterol, which account for about 65%, 30% and 3% of diet contents respectively. The most commonly found plant stanols in the human diet are Sitostanol and Campestanol, which combines to make up about 5% of dietary phytosterol (Raphael J. Ogbe1, 2015).

4. Physical and Chemical Characteristics of Phytosterols

Phytosterols and Phytostanols are the largest group of compounds that are found exclusively in plants. They are structurally like cholesterol but differ from cholesterol in the structure of the side chain. They have total 27-30 carbon atoms. They consist of a steroid skeleton with a hydroxyl group attached to the C-3 atom of the A-ring and an aliphatic side chain attached to the C-17 atom of the D-ring. Sterols contain a double bond, typically between C-5 and C-6 of the sterol moiety. They are obtained from hydroxylated polycyclic isopentenoids having a 1, 2-cyclopentanophenthrene structure. Phytosterols exists in different forms and available in wide sources (Gupta A.K., 2012). Phytostanol and phytosterol esters are chemically stable, fat type materials and having comparable chemical and physical properties to edible fats and oils. Phytosterols are insoluble in water but are soluble in non-polar solvents, such as hexane, iso-octane and 2-propanol. The esters 0f phytosterols are also soluble in vegetable fats and oils. Stability of heat of phytosterol esters is comparable to or even better than that of the parent vegetable oil or oil blend from which the fatty acids were derived Salta F.N., 2008). As a pure material or in a product, phytosterol esters can produce similar decomposition products to those of edible oils and fats as oxidation of the fatty acid moiety is the major cause of the quality deterioration and formation of off-flavors in oils and fats. The color of isolated phytosterols in general will be a whitish solid and/or a pale-yellow color. These phytosterols are soluble in organic solvents but insoluble in water which reveals that they have hydrophobic nature. The melting point is -25.7 to 38.8 °C (Wadikar D.D, 2017).

5. Stability and Textural Characteristics of Phytosterols

The factors affecting phytosterol oxidation include temperature, heating time and the composition of the lipid matrix. Phytosterol esters are likely to be more susceptible to oxidation at elevated temperatures than free phytosterols (Bouic P.J.D., 1999). The phytosterols and their fatty acid esters are very stable compounds and undergo only limited degradation during oil processing. Only under harsh conditions at temperatures greater than 100°C and in the presence of oxygen, oxidation of the phytosterol moiety may occur, in the same way as that for cholesterol. As phytosterols have only one double bond in the B-ring like the fatty acids but are much more stable than the monounsaturated fatty acids (e.g. oleic acid), because of steric hindrance by the ring structure. Therefore, under severe conditions sterol oxidation products are formed slowly and under conditions like when used for shallow frying (temperatures range 160-200 °C, 5-10 minutes of frying) the level of oxidation of sitosterol products were seen (2.5 and 5.1%) using free sterols instead of esters in the same levels are respectively, during a pan-frying phytosterol oxides were formed at a very low level (Ferrari R.A., 1997).Phytosterols like cholesterol undergo oxidation during the period of storage. The presence of the tertiary carbon atoms in the structure of phytosterols makes them more susceptible to degradation and formation of variety of oxidized products may occur. The textural characteristics of products that are fortified with phytosterols are significantly influenced by the nature of phytosterol and when phytosterols are added to products, the firmness of the products was increased. Addition of phytosterol powders at 3 and 4% level in cheese spreads have shown significant increase in firmness of the cheese spread. The increased firmness of the cheese spread might be due to addition of free phytosterols in powder form which can occupy the free space present in the cheese spread (Wadikar D.D, 2017).

6. Absorption and Metabolism

Although various diets contain similar amounts of phytosterols and cholesterol however, serum phytosterol concentrations are usually several hundred times lower than serum cholesterol levels in humans (Svoboda J.A., 1985). Likely cholesterol phytosterols are incorporated into mixed micelles before they are taken up by enterocytes. In the enterocytes, phytosterols are not as readily esterified as cholesterol, so they are incorporated into chylomicrons at very low concentrations. Those phytosterols, that are incorporated into chylomicrons enter blood circulation and are absorbed by the liver. Inside the liver, phytosterols are metabolized into cholesterol and other metabolites, due to the action of several enzymes and a key enzyme called cholesterol 7∝-hydroxylase into bile acids, and rapidly secreted into bile, this enzyme is a regulatory enzyme in bile acids biosynthesis. Even though cholesterol may be secreted into bile, the rate of phytosterol secretion in bile is greater than cholesterol secretion. So, if there are low serum concentrations of phytosterols compared to cholesterol this could be explained by decreased intestinal absorption and increased excretion of phytosterols into bile (Dickson O. Ochalefu, 2017).

7. Biological Activities of Phytosterols

a) Effects on Cholesterol Absorption and Lipoprotein Metabolism

The blood cholesterol reducing effect of phytosterols was given 50 years ago. It was not until the 1990 s that significant advancement has been made in the understanding of this field and serious thought was given to their use, as a functional ingredient (J. Quillez, 2003). It was marketed as a pharmaceutical product under the name cytellin, and has been used for treatment of elevated cholesterol from 1954-1982. Now a day, it is well established that high intakes of plant sterols or stanols can lower serum total and LDL-cholesterol concentrations in humans (Nissinen M., 2002). In the intestinal lumen, phytosterols replace the cholesterol from mixed micelles and inhibit cholesterol absorption in blood. In human beings, the consumption of 1.5-1.8 g/day of plant sterols or stanols can reduced cholesterol absorption by 30-40 %. In recent studies in animals it has been observed that intake of phytosterols esterified with n-3 fatty acids not only leads to a lower in cholesterol and triglycerides but also has other cardiovascular advantages (such as beneficial effects on endothelial and vascular smooth muscle cell function) associated with the consumption of n-3 fatty acids (Ewart H.S., 2002). Tissue LDL–receptor expression has been increased in response to decreased cholesterol absorption and resulting in increased clearance of circulating LDL. Decreased cholesterol absorption is also related with increased cholesterol synthesis, and increasing phytosterol intake has been shown to increase endogenous cholesterol synthesis in humans. Instead the increase in cholesterol synthesis induced by increasing phytosterol intake, the net result is a reduction in serum LDL cholesterol concentration (Ratnayake W.M., 2003). Health Canada reviewed the evidence of 84 randomized controlled trials that have been published between 1994-2007 involving phytosterol supplementation. An average of 8.8% reduction in

LDL-cholesterol level was observed at a mean intake of 2 grams of phytosterols per day. Therefore, Health Canada concluded that the sufficient scientific evidence exists to support a relationship between phytosterol consumption and blood cholesterol lowering (Raphael J. Ogbe, 2015).

b) Anti-cancer effect of phytosterol

In past years, a great deal of interest has been given to the role of phytosterol in the protection from cancer. Raicht et al. described that dietary SIT may offer protection from chemically induced colon cancer (Raicht et al., 1980). More recently, specific food components have been identified which are uniquely providing a beneficial effect in lowering the risk of specific cancer subtypes. Plant sterols are well known for their effects on blood cholesterol levels, however research into their potential role in lowering cancer risk remains in its infancy (Willett, W.C., 1995). The dietary contribution of phytosterol may be one nutritional factor that affecting the distribution of this disease (Bruce J., Grattan Jr., 2013). Several mechanisms such as inhibition of carcinogen production, stimulation of apoptosis, and induction of the sphingomyelin cell cycle have been suggested as accounting for the anti-carcinogenic effects of phytosterols. However, cancer therapy using phytosterol formulations has yet to be designed, largely due to the gap in the literature with regards to method of action (Vanu Ramkumar Ramprasath., 2015).

c) Alterations in Cell Membrane Properties

Cholesterol is the important structural component of mammalian cell membranes, so replacement of cholesterol with phytosterols has been found to alter the physical properties of cell membranes in vitro, which could potentially affect signal transduction or membrane-bound enzyme activity (Raphael J. Ogbe, 2015). Limited evidence from animal model of hemorrhagic stroke suggested that very high intakes of plant sterols or stanols replaced cholesterol in red blood cell membranes, resulting in increased deformability and potentially increased versatility. However, daily phytosterols supplementation (1g/1000kcal) for four weeks did not alter red blood cell versatility in human beings (Simon G. Mafulul, 2015).

d) Alteration in Testosterone Metabolism

Evidence from animal studies suggests that very high phytosterol intakes can result in alteration of testosterone metabolism by inhibiting 5-alpha-reductase, a membrane-bound enzyme that converts testosterone to dihydro-testosterone, a more active metabolite (Jones P.J., 1999). Thus, phytosterols may reduce the activity of enzymes involve in testosterone metabolism in male animals. A recent study has been shown that there are no significant changes in free or total serum testosterone concentrations were observed in men who take 1.6 g/day of plant sterol esters for one year (Ling W.H., 1995).

e) Anti-inflammatory Effects

Limited data from cell culture and animal studies has been shown that phytosterols may be attenuating the inflammatory activity of immune cells, including macrophages and neutrophils. So, they have anti-inflammatory activities in the living systems (Howard B.V., 1997).

f) Disease Treatment

Benign Prostatic Hyperplasia (BPH) is the term that is using to describe a non-cancerous enlargement of the prostate. The enlarged prostate may exert pressure on the urethra and resulting in difficulty urinating. In the six-month study of 200 men who have symptomatic BPH, 60 mg/d of a beta-sitosterol preparation improved symptoms, increased peak urinary flow, and decreased post-void residual urine volume compared to placebbo. However, relatively a few controlled studies have been examined to show the efficacy of phytosterol supplements in men with symptomatic BPH (Raphael J. Ogbe, 2015).

g) Adverse Effect

Phytosterols are usually well tolerated; however nausea, indigestion, diarrhea and constipation have occasionally been reported. So far, a few adverse effects have been linked with the regular consumption of plant sterols or stanols for up to one year (Thanh T.T., 2006).

h) Pregnancy and Lactation

Phytosterols or stanols added to foods or supplements generally are not recommended for pregnant or breastfeeding women because their safety has not been studied. At present, there is little or no evidence that high dietary intakes of naturally occurring phytosterols, such as consumed by vegetarian women, could adversely affect pregnant or lactating women (Raphael J. Ogbe, 2015).

i) Phytosterol-Nutrient Interactions

I. Fat-Soluble Vitamins (Vitamins A, D, E & K)

The effect of plant sterols and stanols at the fat-soluble vitamins status has also been studied in clinical trials because they are known to lower the cholesterol absorption and serum LDL cholesterol concentrations. Plasma vitamin A (retinol) concentrations have not been affected by plant stanol or sterol esters consumption for up to one year. Most of the studies found that there are no changes in plasma vitamin D (25-hydroxyvitaminD3) concentrations, however, one placebo-controlled study in individuals consuming 1.6 g/day of sterol esters for one year observed a small (7%) but statistically significant decrease in plasma 25–hydroxyvitamin D3 concentration. Here is very evidence that plant sterol or stanol consumption adversely affects vitamin K status. Consumption of 1.6 g/day of sterol esters for six months has been associated with a non-significant (14 %) decrease in plasma vitamin K concentrations but carboxylated osteocalcin, a functional indicator of vitamin K status, remained unaffected.

II. Carotenoids

Dietary carotenoids are fat soluble phytochemicals so, they circulate in lipoproteins. 10–20% reductions in plasma carotenoids have been reported after short term and long-term consumption of plant sterol or stanol enriched foods. It is not obvious whether reductions in plasma carotenoids concentrations could cause any health risks but several studies have found that increasing intakes of carotenoid rich fruits and vegetables can prevent phytosterol-induced decrease in plasma carotenoids (Simon G. Mafulul, 2015).

8 Legislation

Phytosterols have been used for reducing plasma cholesterol levels for half a century now, and so far, no marked adverse effects have been reported. It has been found that the plant sterols, within the safe range, that causes desirable reduction in blood levels of total cholesterol and LDL-cholesterol, are referred clinically safe. Plant sterols or stanols added to foods or supplements are not suggested for pregnant or breast-feeding women because their safety has not been studied.

Plant sterol, stanols and their esters has been given GRAS status in United States. Based on this recognition, the US Food and Drug administration (FDA) approved fat spreads which containing up to 20% of either steryl or stanyl esters. The EU Scientific Committee on Food approved the use of the phytosterol esters in yellow fat-spreads (Maximum level of 8% of free phytosterols) in 2000. The European Food Safety Authority suggested that plant sterol containing foodstuffs should not be consumed in amounts resulting in total phytosterols intakes exceeding 3 g/day.

In 2004 the EU commission published regulation for labeling of foods and food ingredients with added phytosterols and/ or phytosterol esters, here is need for such products to be labeled with additional information including the words “With added plant sterols/ plant stanols”. A committee from Food Safety and Standards of India (FSSAI) stated that under the fortification category of food product standards and the food additives, Phyto or plant stanol esters and sterols may be added to many food products fat spreads, milk products, milk based fruit drinks, fermented milk products, soy and rice drinks, cheese products, yoghurt products, spice sauces, salad dressings, juices and nectars (Trautwein EA., 2007).

Conclusion

Phytosterols are naturally occurring compounds, found in plants and they include sitosterol and campesterol, and their saturated counterparts sitostanol and campestanol. Phytosterols and their derivatives have several biological activities which promote the health of animals, humans and micro-organisms. These health benefits include, lowering of plasma total and LDL-cholesterol levels, which decrease the risk of cardiovascular diseases, anti-inflammatory activities and prevention of colon, breast and prostate cancers, and treatment of benign prostatic hyperplasia. Therefore, regular consumption of plant sterols and stanols in natural foods should not exceeding 3 g/day is considered healthy to man and animals. Plant sterol, stanols and their esters has been given GRAS status by several legal authorities. Fortification of the food with Plant sterol and stanols could be an important public health policy if new technology can lower the cost to enable a greater demand to be satisfied.

References

- Andersson S.W., Skinner J., Ellegard L., Welch A.A., Binghan S., Mulligan A., Andersson H, Khaw KT.2004 Eur J Clin Nutr, 58, 1, 1378 – 85.
- Awad AB, Fink CS, Williams H, Kim U, Eur J Cancer Prev, 2001, 10, 6, 507 – 513.
- Bouic PJD and Lamprecht JH. “Plant Sterols and Sterolins: A Review of Their Immune-Modulating Properties”. Alternative Medical Review 4.3 (1999): 170-177.16.
- Bruce J., Grattan Jr.2013. Plant Sterols as Anticancer Nutrients: Evidence for Their Role in Breast Cancer. Nutrients (5) 359-387.
- Ewart H S, Cole L K, Kralovec J, Layton H, Curtis J M, Wright J L C, Murphy M G.2002. Fish oil containing phytosterol esters alters blood lipid profiles and left ventricle generation of thromboxane A2 in adult guinea pigs. J Nutr 2002; 132: 1149–1152.

- Ferrari R.A., Esteves W., Mukherjee K.D., and Schulte E., 1997. Alterations of sterols and steryl esters in vegetable oil during industrial refining. J. Agric. Food Chem., 45, 4753-4757.
- Ferrari RA., et al. "Alteration of sterols and steryl esters in vegetable oils during industrial refining". Journal of Agricultural and Food Chemistry 45.12 (1997): 4753-4757
- Gupta A K., et al. "Role of phytosterols in lipid-lowering: current perspectives". Q J Med 104.4(2011): 301-308.6. Aparna Kuna and Poshadri A. "Phytosterol- A functional ingredient for Indian food industry". Indian Food Industry 30.3 (2012): 21-28.7.
- Howard BV, Kritchevsky D: Phytochemicals and cardiovascular disease: a statement for healthcare professionals from the American Heart Association. Circulation 95:2591–2593, 1997.
- J. Quiłlez, P. Garciła-lorda, Y. J. Salas-salvadoł Y. 2003. Potential uses and benefits of phytosterols in diet: present situation and future directions. Clinical Nutrition 22(4):
- Jessica J.A. Ferguson, Elizabeth stojanovski, 2016. Fat type in phytosterol product influence their cholesterol lowering potential. Progress in lipid research 64, 19-20.
- Jones PJ, Ntanios FY, Raeini-Sarjaz M, Vanstone CA: Cholesterol-lowering efficacy of a sitostanol-containing phytosterol mixture with a prudent diet in hyperlipidemic men. Am J Clin Nutr 69: 1144–1150, 1999.
- Lee EY, Shim YH, Chitwood DJ, Hwang SB, Lee J, Paik YH, Biochem Biophys Res Commun, 2005, 328, 929936.
- Ling WH, Jones PJ: Dietary phytosterols: a review of metabolism, benefits and side effects. Life Sci 57:195–207, 1995.
- Nissinen M, Gylling H, Vuoristo M, Miettinen TA, Am J Physiol Gastrointest liver physiol, 2002, 282, 6, G1009 – 1015.
- Nouroos zadeh J, Appleqvist L.1992. Isolation and quantitative determination of sterols and sterol oxides in plant based foods: Soybean oil and wheat flour". Journal of American oil chemistry society. 69.3:288; 293.

- PIIRONEN V, LINDSAY DG, MIETTINEN TA, TOIVO J, LAMPI AM. (2000). Plantsterols: biosynthesis, biological function and their importance to human nutrition. J Sci Food Agric 80: 939-66.
- Priyanka R. Chandhari, Amit Kumar Jain, and K. D. Aparnathi.2008. Phytosterol: A Functional Ingredient in Food. Lipid health dise, 1-19.
- Raicht, R. F., Cohen, L. I., Fazzini, E. P., Sarwal, A. N. & Takahashi, M. (1980) Protective effect of plant sterols against chemically induced colon tumors in rats. Cancer Res. 40: 403–405.
- Raphael J. Ogbe1, Dickson O. Ochalefu, Simon G. Mafulul and Olumide B. Olaniru.2015. review on dietary phytosterols: Their occurrence, metabolism and health benefits. Asian Journal of Plant Science and Research 5(4), 10-11.
- Raphael J. Ogbe1, Dickson O. Ochalefu, Simon G. Mafulul and Olumide B. Olaniru.2015. review on dietary phytosterols: Their occurrence, metabolism and health benefits. Asian Journal of Plant Science and Research 5(4), 11-12.
- Raphael J. Ogbe1, Dickson O. Ochalefu, Simon G. Mafulul and Olumide B. Olaniru.2015. A review on dietary phytosterols: Their occurrence, metabolism and health benefits 5(4), 16-19.
- Ratnayake WM, Plouffe L, L'Abbe MR, Trick K, Mueller R. Hayward S, Lipids, 2003, 38, 12, 1237 – 1347.
- Richard Cantril, Yoko Kawamura.2008. Phytosterols, phytostanols and their esters. Lipid science technology, 107-110. Wadikar DD, Lakshmi Iyer and Patki PE.2017. Phytosterols: An Appraisal of Present Scenario. Acta scientific nutritional health, 26-27.
- Salta FN., et al. "Distribution and retention of phytosterols in frying oils and fried potatoes during repeated deep and pan frying". European Food Research and Technology 227.2 (2008): 391-400.
- Sudhop T., Lutjohann D., von Bergmann K.2005.Pharmacol Ther.105, 3, 333– 341.
- Svoboda J.A., Thompson MJ, In: Kerkut GA and Gilbert LI (Eds), Comprehensive Insect Physiology, Biochemistry and Pharmacology, Pergamon, Oxford, London, 1985, pp 137.

- Thanh T.T., Vergnes M.F., Kaloustian J., El-Moselhy T.F., Amiot-Carlin M.J., and Portugal H., 2006. Effect of storage and heating on phytosterol concentrations in vegetable oils determined by GC/MS. J. Sci. Food Agric. 86, 220-225.
- Trautwein EA and Demonty I (2007). Phytosterols: natural compounds with established and emerging health benefits. Oilseeds fats Crops Lipids, 225-226.
- Valsta L.M., Lemstrom A, Ovaskainen M.L, Lampi A.M, Toivo J, Korhonen T, Piironen V, Brit J.2007, nutr 92, 4, 671-8.
- Wadikar DD, Lakshmi Iyer and Patki PE.2017. Phytosterols: An Appraisal of Present Scenario. Acta scientific nutritional health 1(1), 26-27.
- Willett, W.C. Diet, nutrition, and avoidable cancer. Environ. Health Perspect. 1995, 103, 165–170.
- Williams CL, Bollella MC, Strobino BA, Boccia L, Campanaro L: Health effects of plant stanol ester and bran fiber on lipids, stool weight and stool frequency in young children. J Am Coll Nutr 6:572–581, 1999.
- Zawitowski J., Kitts D.D., Stoynov N., and Du K., 2003. Thermal oxidation behaviour of phytosterol mixture in different oil systems. 94th AOCS Annual Meeting, Kansas City, KS, USA.