

# Active ingredient for cholesterol: Vitasterol® S-80 Esterified IP produced by VITAE CAPS, S.A.

**H**igh cholesterol is a leading cause of heart disease and stroke, because cholesterol clogs arteries, causing arteriosclerosis.

Recent research from the World Health Organization highlights the importance of raised blood cholesterol as a risk factor for cardiovascular diseases. The World Health Report 2002 estimates that around 8% of all disease burden in developed countries is caused by raised blood cholesterol, and that over 50% of cardiovascular disease in developed countries is due to blood cholesterol levels in excess of the theoretical minimum (3.8 mmol/l).

More recently the INTERHEART case-control study estimated that 45% of heart attacks in Western Europe and 35% of heart attacks in Central and Eastern Europe are due to abnormal blood lipids, and that those with abnormal lipids are at over three times the risk of a heart attack compared to those with normal lipids.

Trend data from MONICA Project (Geneva) show that the prevalence of cholesterol of 6.5 mmol/l and above varies substantially across the populations sampled. These data were collected using standardized methods between 1989 and 1997 for the 35-64 years age range in 36 populations in European countries, New Zealand, Australia, United States, Canada, China and Russia. (Graphic 1)

**People could prevent or reduce high cholesterol with physical activity and healthy diet, but it is also reduced taking something in diet such as sterol esters and sterols.**

## Sterols definition

Sterols and sterol esters are present in our daily diet, they are natural components of vegetable oil such as soy and sunflower. They belong to the same family as cholesterol with similar characteristics, including  $\beta$ -Sitosterol, Campesterol and Stigmasterol. The efficacy of these plants to reduce cholesterol levels in blood are based on their ability to inhibit the absorption of cholesterol produced in the small intestine either internally or through the diet.

Since first works written by Pollak in 50s, free sterols lowering effect of cholesterol in humans has been demonstrated. At a later stage, it has found that sterols esterified with fatty acids have higher effectiveness because they are more soluble and can be hydrolysed in the normal digestive process.

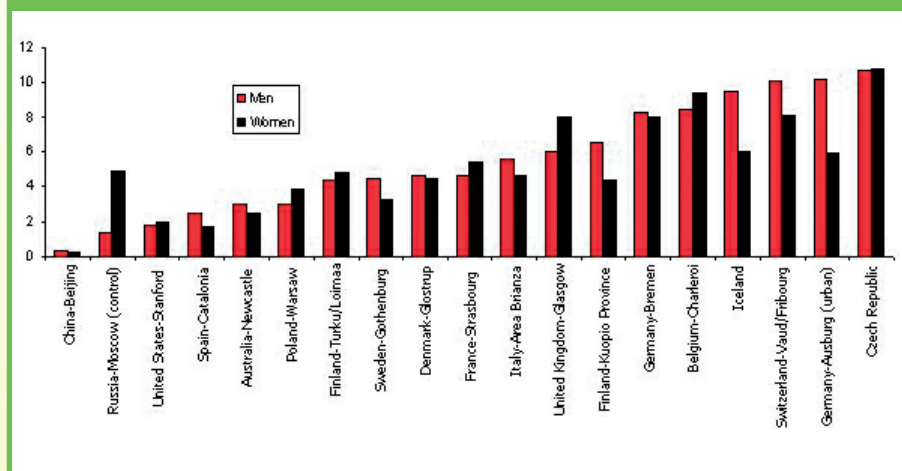
To date, over 40 clinical trials have been conducted involving over 2000 individuals at study sites in Europe, North America, Latin America,

Africa, Asia and Australia. These studies have shown that consuming sterols and sterol esters decrease cholesterol levels by an average of

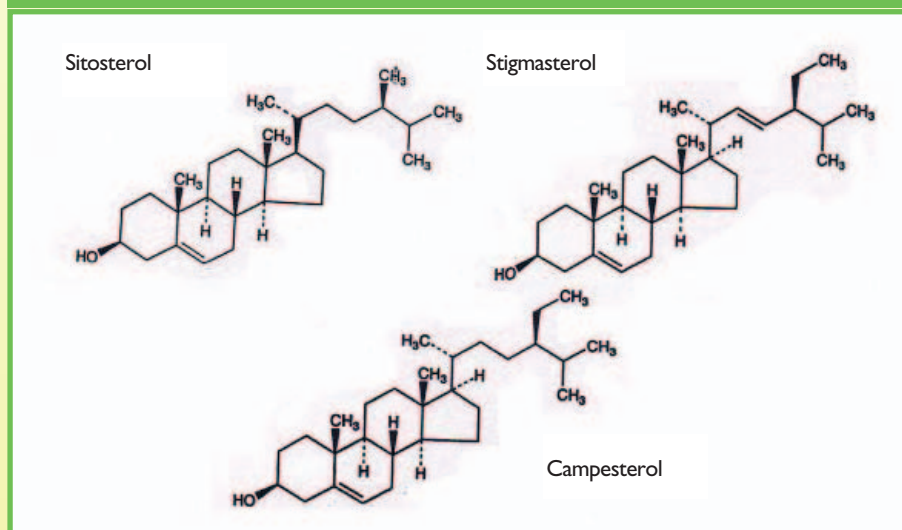
5-15% and LDL cholesterol by an average of 10-20%.

In July 2000, the European Parliament and

**Graphic 1: Percentage of adults aged 35-64 with blood cholesterol levels > 7.8 mmol/l, latest available date, MONICA Project Populations**



## Structures of sterols



EU approved food categories	Other applications
Yoghurt	Juices
Milk	Breakfast cereals
Cheese	Bakery products
Soy drinks	Supplements
Rye bread	Snack bars
Spicy Sauces	
Yellow fat spreads	
Salad dressings	
Milk based fruit beverages	

Vitasterol® S-80 IP Composition (Listed as Free Sterols)	Free Sterols (wt %)
$\beta$ -Sitosterol	45
Campesterol	25
Stigmasterol	23
Brassicasterol	1
Campestanol	0
Sitostanol	0
Other sterols and stanols	3
<b>Total</b>	<b>97</b>

the Council authorised the placing on the market of "yellow fat spreads with added sterol esters" as a novel food under Regulation (EC) No 258/97 of the European Parliament and of the Council. After that, many applications have done at European Commission to use sterol and sterol esters. Vitae Caps have EU authorisation in most of EU food approved categories.

**Vitasterol® S-80 esterified IP is the result of VITAE CAPS, S.A. high investment efforts to launch ingredients which enhance people's health. Other ingredients produced at VITAE CAPS, S.A. are natural source Vitamin E IP (Vitapherole®) and soy IP isoflavones (Vitaflavon®).**

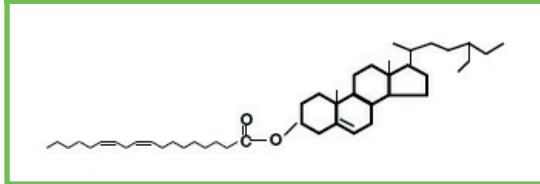
**Vitasterol® S-80 IP - Characteristics**

**Vitasterol® S-80 IP** is a concentrated form of sterols from IP seeds and contains mainly: B-Sitosterol, Campesterol and Stigmasterol.

**Vitasterol® S-80 esterified IP**

**Vitasterol® S-80 esterified IP** is obtained by esterification of free plant sterols with fatty acids from vegetable oil. The esterification process achieves that sterols will be more soluble in oils, being a better vehicle for the absorption of sterols to the small intestine.

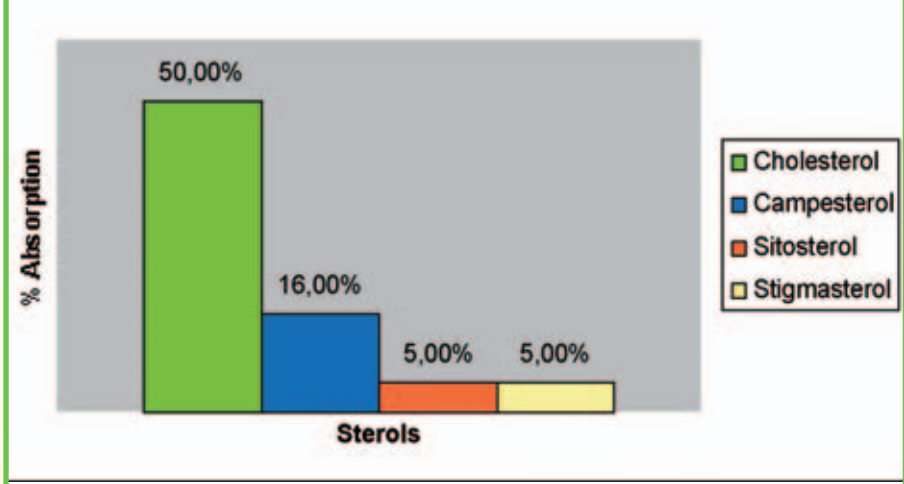
**Structures of sitosterol sterol ester**



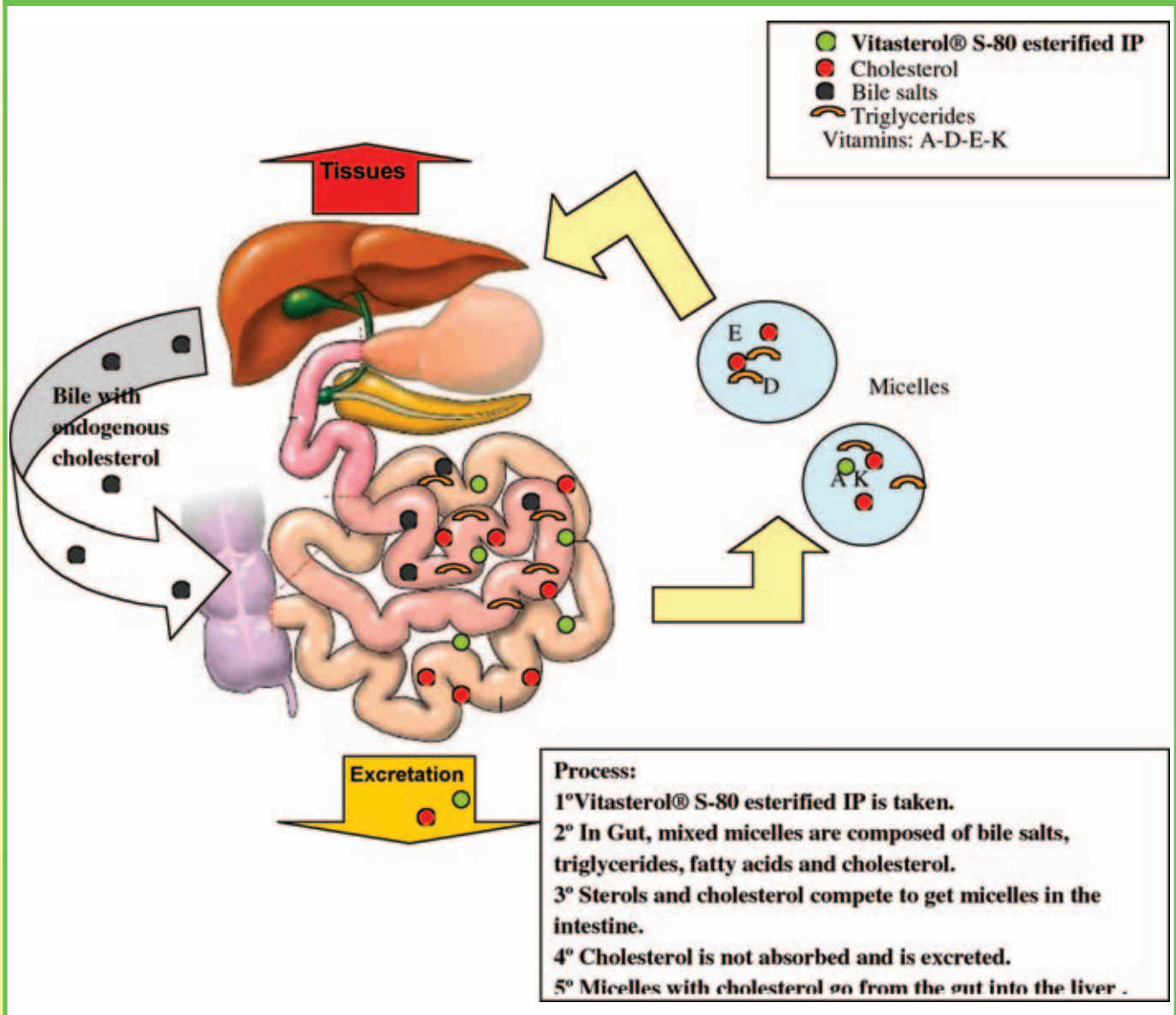
**Vitasterol®, S-80 Esterified IP Properties**

Sterol esters possess a lot of properties: anti-inflammatory, anti-cancer, anti-atherogenicity, and anti-oxidation activities apart from the most recognized one, lowering low density lipoprotein (LDL) cholesterol

**Cholesterol and Sterol Absorption Efficiency**



**Cholesterol and Sterol Absorption Efficiency**



and maintaining good heart health.

People usually consume sterol esters every day, however the amount is not enough to have a relevant effect on blood cholesterol, in order to reduce cholesterol, human should take 1,3 g/day of plant sterols, human should take at least 1,3 g/day and at the most 3 g/day of plant sterols.

The main sterol existing on human cells is cholesterol. In addition to be an important structural component in cells membranes, it is necessary to synthesize hormonal steroids, bile and vitamin D. Even though it is indispensable, an excessive amount in blood is harmful. It accumulates in wall arteries inducing their narrowing and reducing the nutrition's contribution to tissues.

Scientific researches have proven that sterols prevent endogenous cholesterol absorption produced by our body as well as consumed in diet. This property come from the resemblance of cholesterol and sterol esters structure. When bile salt liver's, containing cholesterol, try to get micelles before being inhibited by the intestine. If there are sterol ester in these salts, they compete, as they are more soluble, they displace cholesterol and it is secreted in the faeces.

**How can Vitasterol® S-80 esterified IP work to decrease blood cholesterol?**

Cholesterol could come from the bile

(endogenous) or from food (exogenous), when Vitasterol® S-80 esterified IP is taken in food, sterols are delivered to the small intestine where they meet with endogenous cholesterol transported by bile salts and exogenous one. In the gut, they try to compete to get micelles, however as sterol esters are more soluble, cholesterol is not absorbed and is excreted. Unlike cholesterol, sterol esters are not soaked up in the blood. Even if intake of sterol esters is increased, they will be faster eliminated than cholesterol through the bile.

Vitasterol® S-80 esterified IP must be taken by people with cholesterol problems, but not by children younger than five years of age, lactating women and pregnant women.

No secondary effects have been found, except the reduction of serum carotene levels in some of the studies. Nonetheless it would not happen, if people take a diet enriched with

vegetables and fruits.

**Vitasterol® S-80 esterified IP applications**

A wide range of food applications for sterols and sterol esters have been approved by the European Commission: yoghurt, milk, cheese, soy drinks, spicy sauces, yellow fat spreads and rye bread. However US Food and Drug Administration goes ahead allowing more uses in everyday consumer products such as health drinks, health bars, pasta and noodles, salty snacks, processed soups, puddings, soy milk, ice cream and cream substitutes, adult confections, vegetarian meat analogs, cheese and cream, edible vegetable oil, adult ready-to-eat breakfast cereals, baked goods and fruit/vegetable juices.

An example of applications is the following:

Product	Sterol ester content	Free sterol content	Product Quantity
Milk type products	1,6 g	1 g	250 ml
Yellow fat spreads	1 g	1 g	12,5 g
Yoghurt type products	1,2 g	1 g	100 g-150g

**Table 2: Some examples of recent clinical trials are summarized**

Food type	Number of subject duration	Diary dose	Reference	
Phytosterols-enriched margarines	85 subjects with 2 diabetic patients 12 weeks	2x10 g/day with or without 8% phytosterol-esters	Randomised, placebo-controlled, double-blind clinical trial in two parallel groups over 12 weeks. After 4 weeks, total and LDL cholesterol were significantly in the phytosterol group by 5.2% and 6.8%, respectively, compared to baseline (p<0.05). After 8 and 12 weeks, these reductions became smaller and were no significant any more compared to baseline or between the groups, but a repeated measurement analysis demonstrated a significant difference for both variables between the two groups (each p<0.05). HDL cholesterol was significantly increased in the phytosterol group compared to the placebo group after 8 and 12 weeks, but there was no overall difference in the repeated measurement analysis between the two groups.	Lee et al, 2003
Margarine enriched with plant sterols	42 healthy male students	30 g/day in 2 servings	Randomised double groups The subjects consuming margarine with sterols showed a significant (11%) decrease in LDL-C (P<0.001). After the consumption of margarine with sterols, the adhesion and aggregation time of blood platelets was significantly prolonged after collagen-epinephrine activation.	Kozłowska-Wojciechowska et al, 2003
Plant sterol-enriched spreads	42 healthy adults 4 weeks	Sterol content of the enriched spread 8.3% 25 g/day	Randomised, placebo-controlled, double-blind crossed over study. Serum total and LDL-cholesterol concentrations lowered by 7% (18 mg/dl) and 10% (16 mg/dl), respectively, with the plant sterol enriched compared to the control spread. Serum HDL-cholesterol concentrations lowered did not significantly differ between the two spreads. Apolipoprotein B concentrations lowered by 8% (0.08 g/day) with the plant sterol enriched spread, while concentrations of apolipoprotein A-I did not change.	Temme et al, 2002
Plant sterol-enriched spreads	50 healthy subjects 11 weeks	20 g/day butter, butter with 25 g polyunsaturated spread, 25 g with 2 g of plant sterols	Parallel butter phase, double-blind, randomised, crossover polyunsaturated spread phases. Replacing butter with a standard polyunsaturated fat spread reduced mean plasma total cholesterol concentrations by 4.6% and low density lipoprotein cholesterol by 5.5%. Replacing butter with a polyunsaturated spread containing plant sterols reduce plasma total cholesterol by 8.9% and low density lipoprotein cholesterol by 12.3%	Cleghorn et al, 2003
Stanol and sterol esters enriched margarines	34 hypercholesterolaemic subjects 4 weeks each margarine	Placebo, 2.01-2.04 g plant sterols and esters	Randomised double-blind with three test margarines. Stanol esters and Sterol esters margarines reduced significantly and equally serum total and LDL cholesterol concentrations as part of a low-fat diet.	Hallikainen et al, 2000

Food type	Number of subject duration	Diary dose	Reference	
Phytosterol ester-enriched low-fat foods: bread, breakfast cereal, milk and yoghurt	58 Healthy man and women 3 weeks each product	1.6 g/day of phytosterols as sterol esters	Randomised, incomplete crossover, single-blind study. Serum total and LDL cholesterol levels were significantly lowered by consumption of phytosterol-enriched foods: milk (8,7% and 15,9%) and yoghurt (5,6% and 8,6%). Serum LDL cholesterol levels fell significantly by 6,5% with bread and 5.4% with cereal. They were both significantly less efficacious than sterol-enriched milk (P<0.001). Plasma sitosterol increased by 17-23% and campesterol by 48-52% with phytosterol-enriched milk and bread. Lipid-adjusted beta-carotene was lowered by 5-10% by sterols in bread and milk respectively. Plant sterols in low-fat milk was almost three time more effective than in bread and cereal.	Clifton PM et al 2004
Low-fat milk-based beverage with plant sterols	26 Healthy men 1 week for each product	Placebo, 2.2 g plant sterols, 2.2 g plant sterol esters	Double-blind, randomised, crossover study. Both plant sterols reduced beta-carotene and alpha tocopherol bioavailability and cholesterol absorption in normocholesterolemic men.	Richelle et al. 2004
Low-fat milk-based with sterol esters, low-fat yoghurt with sterol and stanol esters	39 Healthy subjects 4 phases of 3 weeks 40 Healthy subjects 3 phases of 3 weeks	300 ml/d of placebo and 300 ml/d with 2 g sterol esters 2 portions of 150 g of placebo, 1,8 g sterol esters yoghurt and 1.7 g plant stanol	2 Studies: one single blind crossover and another one fully randomised, double blind. In study one, sterol ester milk and spread were equally efficacious in lowering total and LDL-cholesterol as cholesterol as compared to placebo by 6%-8% and 8%-10%, respectively. Sterol ester enriched milk an the combination of sterol ester enriched milk plus spread both lowered lipid-adjusted serum beta-carotene concentration by 10%-14% (P<0,02). In study two, the sterol ester and stanol ester enriched yoghurts reduced LDL-cholesterol significantly compare to placebo by 0,27 +/-0,05 mmol (6%) and 0,23 +/-0,05 mmol/l (5%), respectively.	Noakes et al. 2005
Salad dressings enriched with plant sterol esters	53 Men and women 3 weeks	Placebo, 3.6 g plant sterol esters	Randomised parallel trial. Plasma LDL cholesterol (LDLC), HDL cholesterol (HDCL), and triglycerides (TG) responses of men and women to the study diets were similar and not affected by the type of salad dressing they consumed. Switching from a self-selected baseline diet to a controlled diet with no plant sterol esters resulted in a significant reduction of LDLC of 7.9%, HDLC of 3,1%, and TG of 9.2%. Daily consumption of 3.6 g plant sterol esters resulted in further decreased in LDLC of 9.7% and TG of 7.3% with no effect on HDLC.	Judd et al. 2002
Orange juice	72 midly hypercholesterolemic subjects 8 weeks	Placebo orange juice and plant sterol fortified orange juice (2 g/d)	Placebo-controlled, double-blind, randomised study. Sterol orange juice supplementation significantly decreased total (72%), LDL (12,4%), and non-high-density lipoprotein (HDL) cholesterol (7.8%) compared with baseline and compared with placebo orange juice (P<0.01). Apolipoprotein B levels were significantly decreased (9.5%) with sterol orange juice. There were no significant changes in HDL cholesterol or triglycerides with the sterol orange juice	Devaraj et al. 2004
Bread, meat products and jam enriched with sterols	78 subjects from 25-75 years 15 weeks	Placebo food items, bread, meat products, jam with 1,25 to 5 g/day	Randomised, double-blind, placebo-controlled feeding. Reduction in serum total cholesterol was 8% in the active treatment group and 3% in the placebo group and that of LDL cholesterol was 13% in the active treatment group and 5% in the placebo group.	Tikkanen MJ et al 2001

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