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Comparison of rice bran oil margarine with Flora margarine  
and Flora pro-activ® margarine for lowering cholesterol

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Sarah Louise Eady

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## Abstract

Phytosterols have been shown to be effective in reducing serum cholesterol levels in numerous human clinical studies and regular consumption is recommended as part of therapeutic lifestyle changes aimed at reducing low density lipoprotein (LDL-C) in the treatment of hyperlipidaemia, a risk factor for cardiovascular disease. Fat based spreads have been shown to be a very successful vehicle for delivery of plant sterols, readily accepted by consumers and efficacious in reducing cholesterol levels. Alfa One™ Rice Bran Oil (RBO) spread is a new product entering into the market place. It is derived from rice bran oil and contains high levels of unsaponifiable material rich in phytosterols, triterpene alcohols, ferulic acid esters ( $\gamma$ -oryzanol) and vitamin E isomers. As such it may have the potential to lower serum cholesterol levels when consumed on a daily basis.

In order to establish the effectiveness of Alfa One™ Rice Bran Oil (RBO) spread compared with Flora pro-activ® margarine, a well established brand of plant sterol margarine already proven to lower cholesterol, a randomised double blind cross-over human clinical trial over 12 weeks was conducted. The study was divided into two treatment arms. The first arm of the study was to determine whether Alfa One™ RBO spread (containing 1.5% plant sterols) could lower total and LDL cholesterol levels to a greater extent than standard Flora margarine (containing no plant sterols) or Flora Pro-activ® margarine (containing 8% plant sterols). The second study arm tested the proposition that daily consumption of Alfa One™ Rice Bran Oil (RBO) spread in conjunction with rice bran oil (containing 0.5% plant sterols) would lower total and LDL cholesterol to a greater extent than Alfa One™ RBO spread in isolation and more than Flora margarine in conjunction with sunflower oil.

Eighty mildly hypercholesterolaemic individuals (total cholesterol  $\geq 5$  mmol/L and  $\leq 7.5$  mmol/L) were recruited and randomised into two groups of forty. Participants were asked to continue with their normal dietary pattern but to

replace any margarine/butter/fat consumption with the trial products. One group of 40 were then assigned to the first treatment arm of the study (margarine-only group) and were randomised to consume 20 g (4 teaspoons) Alfa One™ RBO spread daily for 4 weeks, or 20 g Flora margarine daily for 4 weeks, or 20 Flora pro-activ® daily for 4 weeks. Phytosterol levels delivered in these amounts were: RBO margarine: 118mg phytosterol and 14 mg  $\gamma$ -oryzanol; Flora proactiv® 1600 mg phytosterol; Flora margarine 0mg phytosterol. The second group of 40 were allocated to the second arm of the trial (margarine and oil group) and consumed 20 g Alfa One™ RBO spread and 30 ml rice bran oil (RBO) daily for 4 weeks, or 20 g Flora margarine and 30 ml sunflower oil daily for 4 weeks, or 20 g Alfa One™ RBO spread daily for 4 weeks, changing treatment at the end of each 4-week period. Phytosterol amounts delivered in these amounts were: RBO margarine: 118 mg phytosterol and 14 mg  $\gamma$  oryzanol; RBO 222mg mg phytosterol, 150 mg  $\gamma$  oryzanol. Each participant consumed all three treatments in a random order over a 12 week period. At baseline and following each 4 week intervention period, measurements were made of weight and blood pressure. Venous blood samples were collected for analysis of total cholesterol, low density lipoprotein cholesterol (LDL-C), high density lipoprotein cholesterol (HDL-C), total cholesterol: HDL-C, triglycerides and plasma phytosterols. Three-day diet records from each individual were also collected for analysis of normal dietary intake.

Results showed that compared to a standard Flora margarine, Alfa One™ RBO spread significantly reduced total cholesterol by 2.2% ( $P=0.045$ ), total cholesterol:HDL by 4.1% ( $P=0.005$ ) and LDL-C by 3.5% ( $P=0.016$ ), but was not as effective overall as Flora Pro-activ® which reduced total cholesterol by 4.4% ( $P=0.001$ ), total cholesterol:HDL by 3.4% ( $P=0.014$ ) and LDL-C by 5.6% ( $P=0.001$ ). Consumption of Flora margarine alone produced no significant decrease from baseline figures in any of the cholesterol parameters measured. Surprisingly, in group two, the addition of rice bran oil to the Alfa One™ RBO spread produced no differences in cholesterol levels. The reason for this unexpected result is being explored further.

These results confirm that Alfa One™ RBO spread is effective in lowering serum cholesterol levels when consumed as part of a normal diet. Studies have shown that a 1% reduction in LDL-C can equate to a 2% decrease in coronary heart disease (CHD) risk thus suggesting that the 3.5% reduction demonstrated by Alfa One™ RBO spread in this study could be effective in reducing CHD risk as much as 6% in a mildly hypercholesterolaemic population.

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## Abbreviations

ABC	adenosine triphosphate binding cassette
Acetyl Co A	acetyl coenzyme A
AGE	advanced glycation end products
ALA	alpha linoleic acid
ALP	atherogenic lipoprotein phenotype
Apo	apolipoprotein
ATP	adenosine triphosphate
bHLH	basic helix-loop helix
BMI	body mass index
BP	blood pressure
CE	cholesterol ester
CETP	cholesterol ester transfer protein
CHD	coronary heart disease
CHO	carbohydrate
CMR	chylomicron remnant
CTD	C-terminal domain
DHA	docosahexanoic acid
DNA	deoxyribonucleic acid
EPA	eicosapentaenoic acid
FDA	food and drug administration
FFA	free fatty acids
FH	familial hypercholesterolaemia
FPG	fasting plasma glucose
GI	glycaemic index
GL	glycaemic load
HDL-C	high density lipoprotein
HIV	human immunodeficiency virus
HMG-CoA	3-hydroxy-3-methylglutaryl-CoA
HsCRP	C-reactive protein
ICAM-1	intracellular adhesion molecule -1
IDL	intermediate density lipoprotein
Insig-1	insulin induced gene 1
IL	interleukin
IR	insulin resistance
LCAT	lecithin: cholesterol acyltransferase
LDL-C	low density lipoprotein cholesterol
LDLr	low density lipoprotein receptor
Lp (a)	lipoprotein (a)
LPL	lipoprotein lipase
LRP	LDL receptor related protein
LXR	liver X receptor
MCP-1	monocyte chemo attractant protein-1
M-CSF	macrophage colony stimulating factor
MetS	metabolic syndrome
MOH	Ministry of Health
MUFA	monounsaturated fatty acids
NCEP	National cholesterol education programme
NFκβ	nuclear factor kappa beta

NHLBI	National Heart, Blood and Lung Institute
NIDDM	non insulin dependent diabetes mellitus
NO	nitric oxide
NOS	nitric oxide synthase
NZ	New Zealand
PAI-1	Plasminogen activator inhibitor 1
PPAR	peroxisome proliferator activated receptor
PPG	post prandial glucose
PUFA	polyunsaturated fatty acids
RAGE	receptor of advanced glycation end products
RAS	rennin angiotensin system
RBO	rice bran oil
SCAP	SREBP cleavage activating protein
SFA	saturated fatty acids
SREBP'S	sterol regulatory element binding proteins
S1P	site 1 protease
S2P	site 2 protease
TG	triglyceride
TLC	therapeutic lifestyle changes
TNF- $\alpha$	tissue necrosis factor alpha
TRF	tocotrienol rich fraction
US	United States of America
VCAM-1	vascular cell adhesion molecule-1
VLDL-C	very low density lipoprotein cholesterol
WHO	World Health Organisation