Original Article

Palm oil *versus* hydrogenated soybean oil: effects on serum lipids and plasma haemostatic variables.

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The purpose of this study was to test if replacement of trans fatty acids by palmitic acid in an experimental margarine results in unfavourable effects on serum lipids and haemostatic factors. We have compared the effects of three different margarines, one based on palm oil (PALM-margarine), one based on partially hydrogenated soybean oil (TRANS- margarine) and one with a high content of polyunsaturated fatty acids (PUFA-margarine), on serum lipids in 27 young women. In nine of the participants fasting levels and diurnal postprandial levels of haemostatic variables on the 3 diets were compared. The sum of 12:0, 14:0, 16:0 provided 11% of energy (E%) in the PALM diet, the same as the sum of 12:0, 14:0, 16:0 and trans fatty acids in the TRANS-diet. Oleic acid provided 10-11E% in all three diets, while PUFA provided 5.7, 5.5 and 10.2 E%, respectively. Total fat provided 30-31% and the test margarines 26% of total energy in all three diets. Each of the diets was consumed for 17 days in a crossover design. There were no significant differences in total cholesterol, LDL-cholesterol and apoB between the TRANS- and the PALM-diet. HDL-cholesterol and apoA-I were significantly higher on the PALM-diet compared to the TRANS-diet while the ratio of LDL- to HDL-cholesterol was lower, although not significantly (P = 0.077) on the PALM-diet. Total cholesterol, LDL-cholesterol and apoB were significantly lower on the PUFA-diet compared to the two other diets. HDL-cholesterol was not different on the PALM- and the PUFA-diet while it was significantly lower on the TRANS-diet compared to the PUFA-diet. Triglycerides and Lp(a) were not different among the three diets. The diurnal postprandial state level of tissue plasminogen activator (t-PA) activity was significantly decreased on the TRANS-diet compared to the PALM-diet. t-PA activity was also decreased on the PUFA-diet compared to PALM-diet although not significantly (P=0.07). There were no significant differences in neither fasting levels or in circadian variation of t-PA antigen, PAI-1 activity, PAI-1 antigen, factor VII coagulant activity or fibrinogen between the three diets. Our results suggest that dietary palm oil may have a more favourable effect on the fibrinolytic system compared to partially hydrogenated soybean oil. We conclude that from a nutritional point of view, palmitic acid from palm oil may be a reasonable alternative to trans fatty acids from partially hydrogenated soybean oil in margarine if the aim is to avoid trans fatty acids. A palm oil based margarine is, however, less favourable than one based on a more polyunsaturated vegetable oil.

Key Words: coronary heart disease, *trans* fatty acids, palm oil, hydrogenated soybean oil, fibrinolysis, serum cholesterol, serum lipids, plasma haemostatic variables, palmitic acid

Introduction

Trans fatty acids have unfavourable effects on serum lipids and thus on the risk of coronary heart disease. Replacement of trans fatty acids by less atherogenic fatty acids in margarine and other food products is therefore desirable. Palm oil consists of about 50% saturated and 50% unsaturated fatty acids and has a content of 45-50% palmitic acid and about 10% linoleic acid.[1] From the literature it appears that the effects of palmitic acid and palm oil on blood lipids are controversial. Hegsted *et al.*, reported that palmitic acid was cholesterol increasing, but less so than myristic acid.[2] Nestel *et al.*, found no difference in total- and LDL-cholesterol when an elaidic acid (18:1trans) rich diet was compared to one rich in palmitic acid.[3] *Ng et al.*, found a palm oil diet not to be hypercholesterolemic in Malaysian volunteers,[4] and in a later study found that dietary palmitic and oleic acid exert similar effects on serum cholesterol and lipoprotein profiles in normocholesterolemic men and women.[5] Marzuki *et al.*, found no significant difference in effect on total-, LDL- and HDL-cholesterol when palm oil was compared to soybean oil.[6] Choudhury *et al.*, suggest that 16:0 is not always a cholesterol increasing fatty acid. They found no significant differences between palm-olein and olive oil.[7]

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On the other hand, Temme *et al.,* found a significant rise in total serum cholesterol with a diet rich in palmitic acid compared to one rich in oleic acid.[8] One study in primates also indicated that palmitic acid increases total cholesterol.[9] In a comparison between the effects of elaidic acid (18:1trans) and palmitic acid Sundram *et al.,* found the first one to be far more cholesterol increasing than the latter.[10]

In two previously published studies we have compared the effects of three test margarines on serum lipoproteins and some haemostatic variables.[11,12] One margarine had a high content of partially hydrogenated soybean oil (PHSO) (TRANS-margarine), one of similar hardness consisted of 80% palm oil (PALM-margarine), and one was high in polyunsaturated fatty acids (PUFA-margarine). The aim of the studies was to test whether or not palmitic acid can replace trans fatty acids in margarine without having unfavorable effects on blood lipids and haemostatic factors and how a palm oil based margarine compared nutritionally to one high in PUFA.

Materials and methods

Participants and their baseline characteristics

Thirty young female home economics students were recruited in a strictly controlled dietary study. All participants were in good health with no history of diabetes, anaemia, renal, hepatic or gastrointestinal disturbances, hypertension or intolerance and all had normal dietary habits (checked by a detailed food frequency questionnaire). None were taking any medication known to affect serum lipids except for oral contraceptives. We had no screening criteria with regard to smoking habits, age, physical activity or body weight. All participants were requested to maintain their regular lifestyles and especially their usual extent of physical activities throughout the study. They were not allowed to drink alcohol during the study period.

Two persons withdrew during the study and one participant had not fasted before blood sampling and this person also had problems with compliance and was excluded from the study. Thus, data from 27 subjects with a mean age of 27 years (SD 5.8, range 19-42) were evaluated for effects on fasting serum lipid and haemostatic factors. Seven of the women were taking oral contraceptives and ten were smokers. The average weight of the participants was 77.5kg (SD 13 kg) ranging from 56.4 to 100 kg and the body mass index ranged from 20 to 36 kg/m2 (mean 26.5 kg/m^2, SD 4.1). Four persons had a body mass index (BMI) above 30.

Among the 27 subjects ten entered the study of postprandial diurnal variation of haemostatic variables. None of these participants except for one person who used oral contraceptives were taking any medication known to affect haemostatic variables. One person had problems with compliance and was excluded. Thus, data from nine participants were evaluated. The mean age of these nine was 26 years (SD 6.0 range 19-37) and the average weight was 73.1 kg (SD 12.3 kg, range 56.4- 93 kg) and the mean BMI was 25.1 kg/m^2 (SD 4.3 range 20.9 - 32.8 kg/m^2. Three were smokers. All participants were within 85-120% of desirable BMI (≤ 25 kg/m^2) except for one person (BMI=32.8 kg/m^2).

The protocol and the objective of the study were explained to the participants in detail and they gave informed consent before entry into the study. The study protocol was approved by the Regional Committee for Ethics in Biomedical Research of Norway.

Study design

The study ran during periods of 17 days. Studies have demonstrated new stable levels of serum lipids and lipoproteins within 14 days on a controlled diet.[13-15] Each person received the three diets in a sequence determined by assignment to one of six possible sequences as directed by a Latin-square design. The participants were fed simultaneously and all three diets were fed in the three periods. In this way, variation due to residual effects of the previous diet or to drift of variables over time could be minimized. After the end of the test period, the participants crossed over to the next diet with a wash-out period of one week. Body weight was monitored twice a week. BMI was calculated as weight (kg)/height (m)^2. Dietary compliance was checked daily by interview and diaries.

On the seventeenth day all meals were eaten under supervision at the University College. The subgroup of nine students participating in the postprandial study was served breakfast, lunch, dinner and evening meal at 0800, 1100, 1430 and 1900, respectively. Blood samples were taken one and a half hour after each meal and after overnight fasting.

Test margarines

We wanted to test a hard virtually *trans* free margarine containing palm oil (PALM-margarine) with a hard margarine containing a high amount of *trans* fatty acids from partially hydrogenated soybean oil (PHSO) (*TRANS*-margarine). The margarines were formulated to ensure that the sum of 12:0, 14:0 and 16:0 in the PALM margarine was equal to that of the same fatty acids plus *trans* fatty acids in the TRANS margarine. The PALM-margarine was produced from 80% palm oil, 11% soybean oil and 9% rapeseed oil and the *trans* margarine from 56% partially hydrogenated soybean oil, 34% refined rapeseed oil and 10% refined soybean oil. A third margarine was a commercial soft high polyunsaturated margarine consisting of sunflower oil, rapeseed oil coconut oil, and palm oil (PUFA-margarine). Because of its high content of sunflower oil this margarine contained more vitamin E than the two others (205 mg/kg vs. 81, and 100 in the PALM- and the *trans*-margarine, respectively). The margarines were produced by the addition of water, vitamin A, vitamin D, NaCl, aroma (diacetyl 0.002%), beta-carotene as colour and emulsifier (soybean lecithin 0.3%). The PALM-margarine contained 16% water, the *TRANS*-margarine 15.7% and the PUFA-margarine 18% water. The fatty acid composition of the test margarines and the diets is given under Results.

Experimental test diets

The three diets were based on a 7-day menu and were calculated by using a computer based, nutrient-calculation program. They were designed to have the same nutrient composition except for the fatty acids and for the tocopherol content. The fat from the background diet was

calculated to supply a minimal amount of 6% of energy (E%) while the test fat was planned to amount to 28 E%. The menu for the three experimental diets contained the same basic food items and differed only in the source of test margarine used for spreading, baking and cooking. The test fats were also incorporated into the menus in several foods including bread, buns, porridge and sauces for dinner. Dinner was prepared and served under supervision at the college from Monday through Friday. All meals were prepared at school. Supper and breakfast for the next day were sent home. Each Friday weekend meals were packaged for home consumption. Perishable foods were sent in frozen condition. During the controlled feeding periods no foods other than those in the menu were allowed. If the participants lost weight they were allowed to eat buns with the same fat and energy composition as the rest of the diet. Coffee, tea, and mineral water with artificial sweeteners were allowed ad libitum. Initial daily caloric intake for each individual was calculated from estimates of energy requirements based on weight and height.[16] All foodstuffs were weighed for each individual subject. The subjects were supplied with free food to meet 100% of their mean daily energy requirements. The participants were unaware of the type of fat consumed during the three different dietary periods.

Chemical analysis

Duplicate portions corresponding to a daily energy intake of 8.5 MJ were taken of the three diets. After homogenization and freeze-drying the homogenates corresponding to 7 consecutive days were pooled into one portion and kept frozen at -20°C until analyzed. The content of nitrogen, total fat, cholesterol and metabolizable energy of the diets were determined as described previously.[17] The fatty acids of the respective fat extracts were converted to methyl esters by the BF3 method and analyzed by gas chromatography as described in an earlier study.[17]

Blood sampling and analyses

Blood samples were taken after an overnight fast. Standard enzymatic methods were used to analyze serum cholesterol, HDL-cholesterol (after precipitation of the LDL-fraction with dextran sulfate and magnesium) and serum triglycerides.[18,19] using automated analyzer equipment (Hitachi 737, Hitachi Limited, Tokyo, Japan). LDL-cholesterol was calculated using the Friedewald equation.[20] Serum apolipoprotein A-1 (Orion Diagnostika, Espoo, Finland) and serum apolipoprotein B (Behringwerke Ag, Marburg, Germany) were both quantified immunoturbidometrically using an automated enzyme analyzer (Cobas Fara, Hoffman-La Roche, Basel, Switzerland). Serum lipoprotein (a) was quantified by a commercial kit (TintElize Lp (a), Biopool AB, Umeå, Sweden). The coefficients of variation were the following: total cholesterol 2%, HDL-cholesterol 5%, triglycerides 3%, apoA-I 6.3%, apoB 5.5% and Lp (a) 7.7% at 100 mg/L, and 2.7% at 400 mg/L.

For determination of diurnal variation in haemostatic variables a fasting sample was collected on day 17 in each of the three periods at 0730 followed by four non-fasting samples at 0930, 1230, 1600 and at 2030, each about 1h ½ after a meal. On the next day another fasting blood sample was taken at 0730. All venepunctures were performed in the supine position after at least 15 minutes rest. Blood sampling and treatment and methods for the determination of PAI-1 activity, PAI-1 antigen (free PAI-1 as well as in complex with t-PA), t-PA activity, t-PA antigen (free t-PA as well as in complex with PAI-1), fibrinogen, coagulation FVII and FVII activity were according to published procedures and with the use of commercial kits (for detailed description see ref 12). The coefficients of variation were 8.0% for t-PA activity and 8.4% for t-PA antigen, 4.5% for PAI-1 activity, 9.8% for PAI-1 antigen, 4.8% for fibrinogen and 2.1% for FVII activity. All blood analyses were performed at the Clinical Chemistry Department and the Clinical Research Unit, Ullevaal University Hospital, Oslo.

Statistical methods

Data on fasting variables were analyzed by repeated-measures analysis of variance (ANOVA) for a crossover trial. When the analysis indicated a significant effect of a diet ($P<0.05$), the Bonferroni method was used for a pairwise comparison between the three diet groups. P values < 0.05 were considered significant. All P values are two-tailed. Plasma Lp (a) had a skewed distribution and was log transformed before pairwise comparisons were performed. The diurnal levels data were analyzed by repeated measures analysis of variance for a crossover trial. The comparisons between the sets of observations were based on within subject differences. The number of participants was limited and logarithmic transformation was performed on the individual values of skewed variables before statistical computations and significance testing. The statistical package SPSS 8.0 (SPSS Inc., Chicago, IL) was used for the data analysis.

Results

Dietary compliance

All the 27 participants complied well and only very small deviations from the diet were noted. The fasting body weights were significantly reduced ($P<0.01$) during the first two periods (mean loss 1.9 and 0.47 kg, respectively). The weight loss was almost entirely confined to the four participants with BMI >30 that lost between 1.7 and 7kg during the study period. In order to adhere to the "intention to treat" principle the data from all 27 participants are given. The data were also calculated after exclusion of the four subjects with BMI>30. This caused negligible changes in the blood parameters and did not significantly alter any of the conclusions (see below). The mean fasting body weights of the nine participants that took part in the study of postprandial haemostatic variables were not significantly different at the end of the three dietary periods.

Test diets

The energy and protein contents were found to be identical in the three diets (Table 1). Fat provided 30-31% (slightly less than the calculated 34%), and the test margarines 26% of total energy in all three diets. The PALM-diet contained 79.8g fat per 10MJ, the *TRANS*-diet 79.2g fat, and the PUFA-diet 79.2g fat per 10MJ. All three diets were low in cholesterol (Table 1).

Table 1. Content of energy and nutrients of duplicate portions of the test diets[a]

	PALM-diet	*TRANS*-diet	PUFA-diet
Energy, MJ	8.4	8.35	8.48
Protein, % of energy	17.7	17.1	17.2
Fat, % of energy	31	30.1	30.4
Carbohydrate, % of energy	51.3	52.8	52.4
Cholesterol, mg	86	56	80

[a]Portions corresponding to an estimated intake of 8.5 MJ/d were analyzed

The composition of fatty acids in the test margarines and in the duplicate portions of the diets is shown in Table 2. The PALM- diet contained 0.3% *trans* fatty acids, and the *TRANS*-diet 23.1% *trans* fatty acids corresponding to 7 E%. The sum of the cholesterol-increasing saturated fatty acids (12:0 + 14:0 + 16:0) in the PALM-diet was 36% corresponding to 11,2 E%. This was the same as the sum of *trans* fatty acids 23,1% (7E%) and 12:0 + 14:0 + 16:0, 12,5% (3,8E%) in the *TRANS*-diet. The PUFA-diet had a content of 21% of 12:0, 14:0, 16:0 saturated fatty acids (6.1E %). The sum of *cis* 18:2 and 18:3 in the PALM-diet and the *TRANS*-diet were almost identical. The content of oleic acid was almost the same in the three diets.

Serum lipids and apolipoproteins

Table 3 shows concentrations of total-, LDL-, and HDL-cholesterol, apolipoprotein B (apo-B), apolipoprotein A-1 (apoA-1) and triglycerides at baseline and after the three different test diets. There were no significant differences in total cholesterol, LDL-cholesterol and apoB between the *TRANS*- and the PALM-diets. HDL-cholesterol and apo A-I were significantly higher on the PALM-diet compared to the *TRANS*-diet while the ratio of LDL-cholesterol to HDL-cholesterol was lower on the PALM-diet although this difference failed to reach statistical significance (P=0.077).Total cholesterol, LDL-cholesterol and apoB were sigificantly lower on the PUFA-diet compared to the two other diets. HDL-cholesterol was not different on the PALM- and the PUFA-diets, but was significantly lower on the *TRANS*-diet compared to the PUFA diet. The ratio of LDL- to HDL-cholesterol was

Table 2. Fatty acid composition of the test margarines and the corresponding diets

Fatty acid	PALM-marg	PALM-diet	*TRANS*-marg	*TRANS*-diet	PUFA-marg	PUFA-diet
12:00	0.2	0.5	0.2	0.5	3.2	2.6
14:00	0.9	1.6	0.2	1	1.4	1.8
16:00	37.2	33.9	8.7	11	12.5	15.8
16:1c	0.1	0.4	0.2	0.4	0.1	0.4
17:00		0.1	0.2	0.1	0.1	0.1
18:00	4	5.3	8.6	8.8	3	4.4
18:1t		0.1	27	22.6		0.2
18:1c	38.6	37.9	34.8	35.2	39.8	38.6
18:2t/c[a]		0.1	1	0.4	0.2	0.3
18:2c	15.6	16	13.6	13.5	30.8	27.3
18:3c	1.9	2.4	4.2	4.7	6.1	6.1
18:3t/c[b]	0.2		0.4		0.6	
20:00	0.4	0.4	0.4	0.3	0.4	0.2
20:1t		0.1		0.1		0.2
20:1c	0.4		0.6		0.7	
22:00	0.1	0.2	0.3	0.3	0.6	0.3
22:1c	0.1		0.1		0.2	
Sum *trans* fatty acids	0.2	0.3	28.4	23.1	0.8	0.5
Sum 12:0, 14:0, 16:0 and *trans* fatty acids	38.5	36.3	37.5	35.6	17.9	20.7
E% from 12:0, 14:0, 6:0		11.2		3.8		6.1
E% from *trans* fatty acids		0.1		7		0.2
E% from *cis* MUFA		11.8		10.6		11.7
% of energy from PUFA		5.7		5.5		10.2

[a]include *trans*, *cis* and *cis*, *trans;* [b] include *trans*, *cis*,*cis* and *cis*, *cis*, *trans*

Table 3. Serum lipid and lipoprotein levels at the end of the three dietary test periods[1]

	PALM-diet	*TRANS*-diet	PUFA-diet
Total cholesterol, mmol/L	4.74 ± 0.66[a]	4.61 ± 0.70[b]	4.45 ± 0.64[ab]
LDL-cholesterol	2.90 ± 0.75[a]	2.88 ± 0.70[b]	2.61 ± 0.65[ab]
HDL-cholesterol	1.47 ± 0.32[a]	1.32 ± 0,29[ab]	1.43 ± 0.28[b]
LDL/HDL cholesterol	2.09 ± 0.73	2.26 ± 0.73[a]	1.88 ± 0.65[a]
Triglycerides mmol/l	0.90 ± 0.42	0.92 ± 0.39	0.89 ± 0.36
ApoB, g/L	1.11 ± 0.22[a]	1.11 ± 0.22[b]	1.04 ± 0.23[ab]
ApoA-1, g/L	1.78 ± 0.27[a]	1.70 ± 0.26[a]	1.75 ± 0.25
Lp (a) mg/L	309 ± 300	340 ± 339	326 ± 355

[1]Values given as mean ± SD (*N*=27). Numbers with identical superscript indicate significant difference, all *P*<0.01

higher on both the PALM- and the *TRANS*-diet compared to the PUFA-diet although the difference between the PALM- and the PUFA-diet was significant only after exclusion of the four subjects with BMI>30 (*P* =0.063 for *N*=27 and 0.001 for *N*=23). ApoA-I was not different on the PALM- and the *TRANS*-diets compared to the PUFA-diet. Triglycerides and Lp (a) were not significantly different among the three diets.

Levels of diurnal haemostatic variables, triacylglycerols and cholesterol

Of the three diets the PALM-diet had the most favourable effect on t-PA activity (Table 4, Fig.1A). The diurnal levels of t-PA activity were significantly decreased on the *TRANS*-diet compared to the PALM-diet (*P* = 0.05) but not different from those on the PUFA-diet. The decreased diurnal levels of t-PA activity on the PUFA-diet compared to the PALM-diet did not reach statistical significance (*P* = 0.07 with Bonferroni correction) (Table 4, Fig.1A). t-PA activity was lowest in the morning while t-PA antigen and PAI-1 activity and PAI-1 antigen were highest in the morning (Fig.1 A-D). There were no significant differences in fasting t-PA activity between the three diets (data not shown). No significant differences were observed in the levels of t-PA antigen, PAI-1 activity and PAI-1 antigen, factor VIIc activity and fibrinogen (Table 4, Fig.1B-F). No significant differences in postprandial diurnal levels of serum triacylglycerols and total cholesterol (*N* = 9) were observed between the three diets possibly due to lack of statistical test power.

Discussion

In this study we wanted to test whether palm oil could replace PHSO in margarine without causing unfavorable effects on serum lipids and haemostatic variables. *Trans* fatty acids in the *TRANS*-margarine were replaced by an equal amount of saturated fatty acids, mostly palmitic acid, in the PALM-margarine. The sum of 12:0, 14:0 and 16:0 saturated fatty acids was equal to the sum of the same fatty acids and *trans* fatty acids in the *TRANS*-margarine. The contents of *cis* polyunsaturated and *cis* monounsaturated fatty acids were approximately the same in the PALM- and the *TRANS*-diets. The results show that the PALM-diet increases HDL-cholesterol compared to the *TRANS*-diet without having any significant effect on LDL-cholesterol leading to a slightly more favourable ratio of LDL-cholesterol to HDL-cholesterol. Also the

Table 4. Differences in the grand mean levels of diurnal variation of haemostatic variables between the dietary test periods (*N* = 9, six comparisons)[a]

	Mean Differences	*P* value	95% Confidence Interval
Fibrinogen, g/l			
PALM-*TRANS*	0.013 (1.030)	0.595	-0.04, 0.06, (-1.10, 1.15)
PALM-PUFA	-0.003 (-1.007)	0.891	-0.05, 0.05, (-1.12, 1.12)
TRANS-PUFA	-0.016 (-1.038)	0.505	-0.06, 0.03, (-1.15, 1.07)
Factor VII, (%)			
PALM-*TRANS*	-0.004 (-1.009)	0.928	-0.08, 0.08, (-1.20, 1.20)
PALM-PUFA	-0.006 (-1.014)	0.878	-0.09, 0.08, (-1.23, 1.20)
TRANS-PUFA	-0.002 (-1.014)	0.949	-0.08, 0.08, (-1.20, 1.2
PAI-1 antigen, ng/ml			
PALM-*TRANS*	-0.051 (-1.124)	0.599	-0.23, 0.13, (-1.70, 1.35)
PALM-PUFA	-0.050 (-1.122)	0.599	-0.23, 0.13, (-1.70, 1.35)
TRANS-PUFA	0.000 (1.000)	0.999	-0.18, 0.18, (-1.51, 1.51)
PAI-1 activity, (IU/ml)			
PALM-*TRANS*	-0.006 (-1.014)	0.945	-0.17, 0.16, (-1.48, 1.45)
PALM-PUFA	0.061 (1.151)	0.457	-0.11, 0.23, (-1.29, 1.70)
TRANS-PUFA	0.067 (1.167)	0.417	-0.10, 0.23, (-1.26, 1.70)
tPA antigen, ng/ml			
PALM-*TRANS*	-0.000 (-1.000)	0.993	-0.10, 0.10, (-1.26, 1.26)
PALM-PUFA	-0.010 (-1.023)	0.834	-0.09, 0.11, (-1.23, 1.29)
TRANS-PUFA	0.011 (1.026)	0.826	-0.09, 0.11, (-1.23, 1.29)
tPA-activity, (IU/ml)			
PALM-*TRANS*	0.106 (1.276)	0.048[b]	0.00, 0.21, (1.00, 1.62)
PALM-PUFA	0.098 (1.253)	0.072[b]	-0.01, 0.20, (-1.02, 1.58)
TRANS-PUFA	-0.0073 (-1.017)	1.00[b]	-0.11, 0.10, (-1.29, 1.26)

[a] All data are logtransformed during the variance analysis. Untransformed data are given in parentheses.
[b]Adjustment for multiple comparisons: Bonferroni.

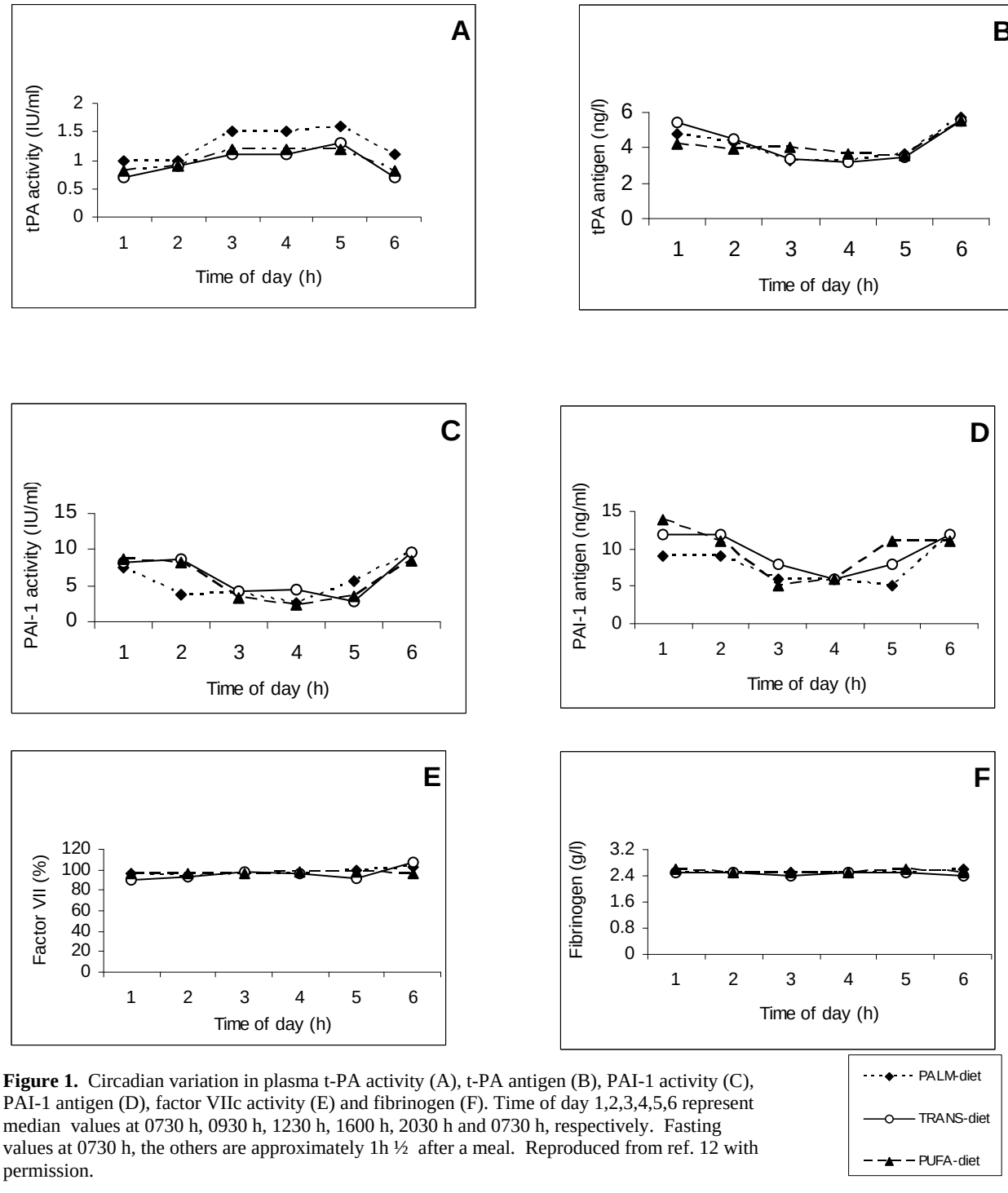

Figure 1. Circadian variation in plasma t-PA activity (A), t-PA antigen (B), PAI-1 activity (C), PAI-1 antigen (D), factor VIIc activity (E) and fibrinogen (F). Time of day 1,2,3,4,5,6 represent median values at 0730 h, 0930 h, 1230 h, 1600 h, 2030 h and 0730 h, respectively. Fasting values at 0730 h, the others are approximately 1h ½ after a meal. Reproduced from ref. 12 with permission.

palm oil rich diet had a more favourable effect on postprandial t-PA activity than one rich in *trans* fatty acids. The results indicate that from a nutritional point of view palm oil may be used as a substitute for PHSO in margarine, which could contribute to a reduced content of *trans* fatty acids in food products. The sensory properties of the two margarines were not identical, however. When changing from PHSO to palm oil in the margarine industry it is essential to obtain satisfactory consistency and taste. As expected, a margarine where saturated fatty acids corresponding to 20% of total fatty acids were replaced by polyunsaturated fatty acids (the PUFA-margarine) had more favourable effects on blood lipids than both of the two other test margarines.

We did not find any significant difference between the PALM-diet and the *TRANS*-diet on the effect on LDL-cholesterol. This result is in accordance with those reported by Nestel *et al.*[3] In that study no significant difference in total- nor in LDL-cholesterol was found with an elaidic acid rich diet (18:1*trans*, 7E%) in 27 mildly hypercholesterolemic men compared to a palmitic acid diet.[3] Our PALM-diet, like that of Nestel *et al.*, contained very little myristic acid which is known to be the most potent cholesterol increasing fatty acid.[4,21,22]

Also, Wood *et al.*, did not find a difference in total- or LDL-cholesterol between crude palm oil and a *trans* fatty acid-rich margarine.[23] The difference in palmitic acid corresponded approximately to that of *trans* fatty acids in

the 2 diets. The diets differed, however, both in oleic and linoleic acid and their results[23] can therefore not be directly compared to ours. Judd *et al*., observed significantly higher total-cholesterol and also higher, but not significantly, LDL-cholesterol on a saturated fat diet (12:0, 14:0 and 16:0) compared to a high *trans* (6.6 E%) diet.[8] Mensink and Katan found a significant increase in total- and LDL-cholesterol on a saturated fat diet compared to *trans*-fatty acid diet.[25] The saturated fat diets in both of these two studies contained higher amounts of both lauric, myristic and palmitic acid than the *trans* diets. When *trans* fatty acids (mostly elaidic acid) were compared directly to palmitic acid Sundram *et al.*, found *trans* fatty acids to be far more total- and LDL cholesterol increasing than palmitic acid.[10] Based on results from 4 dietary studies including those descried here, differing in fatty acid composition we have included saturated and *trans* fatty acids into previously published predictive equations for serum cholesterol by constrained regression analysis.[22] According to these equations the effects of palmitic acid on total and LDL-cholesterol were slightly higher than for *trans* fatty acids. Our analysis failed to obtain a credible predictive equation for HDL-cholesterol. However, here we found the highest HDL-cholesterol on the PALM-diet (Table 3). Also others have found that HDL-cholesterol may be significantly increased with intake of palmitic acid compared to oleic- or elaidic rich diet.[3,23] Even if palmitic acid may be slightly more LDL cholesterol increasing this effect may thus be counteracted by the HDL cholesterol increasing effect of palmitic acid compared to that of *trans* fatty acids. In fact several studies have shown that 18:1 *trans* fatty acids decrease HDL-cholesterol compared to saturated fat or to oleic acid.[25-27] This HDL-cholesterol lowering effect was also observed in this study in that the lowest HDL-cholesterol and apoA-I were observed on the *TRANS*-diet (Table 3). The higher LDL-cholesterol to HDL-cholesterol ratio observed on the *TRANS*-diet was mainly due to this effect on HDL-cholesterol. Also others have noted the increasing effect of *trans* fatty acids on the LDL- to HDL-cholesterol ratio.[25,27] When comparing stick margarines (high in *trans*) to palm oil it was also found that the latter had a slightly more favourable effect on total over HDL cholesterol.[28] Palm oil may thus be preferable to partially hydrogenated soybean oil when considering the effects on the LDL/HDL ratio. In accordance with the results of previous studies total- and LDL-cholesterol were significantly lower on the PUFA-diet than on the PALM-diet[29,30] and also compared to the *TRANS*-diet, the PUFA-diet reduced total- and LDL- cholesterol.[25,26,31]

On the basis of available evidence[32-34] it may be assumed that the combined LDL-cholesterol increasing and HDL-cholesterol lowering effect of *trans* fatty acids contribute to increased risk for CHD. Several epidemiological studies have shown a connection between intake of *trans* fatty acids and atherosclerotic coronary disease.[35-37] In a recent large prospective study it was found that intake of *trans* fatty acids was more strongly associated with CHD than that of saturated fatty acids.[49] This increased risk is, however, greater than would be expected from the effects on serum lipids.[35] To what extent effects on coagulation and fibrinolysis contribute to this increase in risk is unknown. The stronger effect of the PALM-diet on the circadian variation in plasma t-PA activity compared to the *TRANS*-diet may indicate a more favourable effect on the fibrinolytic system.

The relation of haemostatic variables to cardiovascular disease (CVD) is controversial, however[38-44] even if several epidemiological studies show that plasma levels of t-PA activity,[45,46] t-PA antigen[39,47,48] and PAI-I activity[43,46,48,49] may be associated with risk of myocardial infarction. It has been suggested that only for fibrinogen is there significant, strong and consistent evidence of a causal association to CVD.[50] In the Northwick Park Heart Study, which was a prospective epidemiological study, the blood fibrinolytic capacity was evaluated by measuring clot lysis from diluted blood. An increased risk of myocardial infarction was observed according to low activity of lysis of clots.[44] This could again point to the importance of fibrinolysis in atherosclerotic coronary disease.

Up to now mostly fasting values of haemostatic variables have been studied and no differences have been found in fasting haemostatic variables when *trans* 18:1 has been compared to 18:0[51], oleate or carbohydrate.[52] Presumably may postprandial changes in haemostatic variables which fluctuate during the day and night be more physiologically relevant than only fasting values because postprandial changes is usually more long lasting than the fasting condition.

In conclusion, the results from our studies indicate that from a nutritional point of view palm oil may be a reasonable alternative to partially hydrogenated soybean oil in margarine production if the aim is to avoid *trans* fatty acids while maintaining same degree of hardness. A palm oil based margarine is, however, less favourable than one based on a more polyunsaturated vegetable oil.

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Palm oil *versus* hydrogenated soybean oil: effects on serum lipids and plasma haemostatic variables.
棕榈油与氢化大豆油对血清脂质和血浆止血因子影响的比较

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本研究的目的是检验以棕榈酸取代实验性人造黄油中反式脂肪酸是否对血清脂质和止血因子产生不利影响。我们比较三种不同人造黄油对 27 个年轻女性血清脂质的影响，这三种人造黄油一种是基于棕榈油的人造黄油（棕榈油型人造黄油），一种是基于部分氢化的大豆油的人造黄油(反式脂肪酸型人造黄油)，一种为多不饱和脂肪酸（PUFA）含量高的人造黄油（PUFA 型人造黄油）。我们还比较了其中 9 个参与者摄取三种不同膳食后空腹和日间餐后止血因子的水平。棕榈油型膳食中 12:0, 14:0 和 16:0 脂肪酸共提供了 11%的能量，与反式脂肪酸型膳食 12:0, 14:0, 16:0 和反式脂肪酸相加所提供的能量相当。油酸为三种膳食各提供 10-11%的能量，而 PUFA 分别为三种不同膳食提供 5.7, 5.5 和 10.2 %的能量。总的脂肪各为三种不同膳食提供 30-31%的能量，而人造黄油为三种不同膳食分别提供 26%的能量。在交叉设计的实验中，受试者摄取每种膳食各 17 天。反式脂肪酸型膳食与棕榈油型膳食组间总的胆固醇，LDL-胆固醇和 apoB 不存在显著的差异。棕榈油型膳食组 HDL-胆固醇和 apoA-I 显著高于反式脂肪酸型膳食组，而棕榈油型膳食组 LDL-胆固醇与 HDL-胆固醇的比例低于反式脂肪酸型膳食组，尽管差异不是显著性的（P=0.077）。与其他两种膳食组相比，PUFA 型膳食组总的胆固醇，LDL-胆固醇和 apoB 显著降低。棕榈油型膳食组和 PUFA 型膳食组 HDL-胆固醇之间不存在差异，而反式脂肪酸型膳食组 HDL-胆固醇显著低于 PUFA 型膳食组。三种膳食组甘油三酯和 Lp(a)不存在差异。与棕榈油型膳食组相比，反式脂肪酸型膳食组日间餐后组织血纤维蛋白溶酶原激活剂(t-PA)活性水平显著下降。与棕榈油型膳食组相比，PUFA 型膳食组 t-PA 活性也下降，尽管不是显著性的下降(P=0.07)。t-PA 抗原，PAI-1 活性，PAI-1 抗原，VII 因子凝结剂活性，纤维蛋白原等空腹水平或者昼夜变化在三种膳食组间不存在差异。我们的结果表明与部分氢化的大豆油相比，膳食棕榈油可能对血纤蛋白溶解系统有更为有利的作用。我们从营养角度推断假如为了避免反式脂肪酸，棕榈油中的棕榈酸可能是对人造中黄油部分氢化大豆油反式脂肪酸的一种比较合理的替代物。然而，基于棕榈油的人造黄油较基于含多不饱和脂肪酸植物油的人造黄油为差。

关键词：冠状动脉性心脏病，反式脂肪酸，棕榈油，氢化大豆油，纤维蛋白溶解，血清胆固醇，血清脂质，血浆止血因子，棕榈酸