

Original Article

Effect of palm olein oil in a moderate-fat diet on low-density lipoprotein composition in non-human primates

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Plasma low-density lipoprotein cholesterol (LDL-C) concentrations in vervet monkeys (*Cercopithecus aethiops*) can be modulated by the type and amount of fat in the diet. There is, however, a paucity of information on the effect of different types and quantity of dietary fat on the plasma LDL composition in vervets. The objective of this study was to determine the effect of different sources of dietary fat on the concentrations and composition of circulating plasma LDL in vervets consuming moderate-fat diets containing either animal fat, sunflower oil or palm olein. Fifty adult male vervets, never exposed to a Western-type atherogenic diet, were randomly assigned to two groups. For 6 weeks 30 vervets were fed a moderate-fat (28%E) moderate-cholesterol (26 mg cholesterol/1000 kJ) diet (MFD) with a polyunsaturated to saturated fatty acid ratio (P/S) of 0.4; 20 vervets were fed a high-fat (34%E) high-cholesterol (98 mg cholesterol/1000 kJ) diet (HFD) with a P/S ratio of 0.6. Fasting blood samples were collected from all 50 vervets for plasma lipid measurements. The 30 vervets receiving the MFD were stratified into three comparable experimental groups of 10 each according to their LDL-C and high-density lipoprotein cholesterol (HDL-C) concentrations and bodyweight. One group continued with the MFD, in which 11%E was derived from lard (MFD-AF); in the other two groups the lard was substituted isocalorically with either sunflower oil (SO) (MFD-SO) or palm olein oil (PO) (MFD-PO). The three groups were fed the respective experimental diets for 24 months and LDL component concentrations and composition were assessed at 6-monthly intervals. In the long-term study the MFD-AF, MFD-SO and MFD-PO groups showed no significant time-specific group differences at 6, 12, 18 or 24 months with regard to the LDL component concentrations, composition, as well as the LDL molecular weight. As expected, after 6 weeks of dietary exposure the HFD group had significantly higher plasma and lipoprotein total cholesterol, LDL component and apolipoprotein AI concentrations, as well as a higher LDL-C : HDL-C ratio compared to the MFD group ($P \leq 0.0005$). LDL particle size was not significantly different between the HFD and MFD groups, but the HFD group had significantly fewer triacylglycerol and significantly more unesterified cholesterol molecules per LDL particle compared to the MFD group ($P \leq 0.0018$). PO in a MFD is no different from AF or SO in its effect on LDL component concentrations, composition or particle size. The increased LDL-C concentration seen with the HFD could be accounted for by a more than two-fold increase in the number of circulating LDL particles and not as a result of enrichment of particles with cholesterol.

Key words: high-fat high-cholesterol diet, lard, low-density lipoprotein composition, moderate-fat moderate-cholesterol diet, palm olein oil, sunflower oil.

Introduction

Plasma low-density lipoprotein cholesterol (LDL-C) concentrations in African Green (vervet) monkeys (*Cercopithecus aethiops*) can be modulated by the type and amount of fat in the diet.¹ There is evidence that under these conditions changes in plasma LDL-C concentrations can be accounted for mainly by an increase in the number of circulating LDL particles rather than changes in the composition of the LDL particle.^{2,3} These observations are in contrast with the findings of others who reported enrichment of LDL particles with cholesterol ester in non-human primates receiving semisynthetic diets loaded with extra cholesterol.^{4–6}

There is, however, a paucity of information on the effect of different types and quantity of dietary fat on the plasma LDL composition in vervet monkeys consuming a diet consisting entirely of natural cooked foods generally used by Westernised people. The purpose of this study, therefore,

was to determine the effect of different sources of dietary fat on the concentrations and composition of circulating plasma LDL in vervets consuming moderate fat diets (28%E) containing either animal fat (lard), sunflower oil (SO) or palm olein oil (PO).

Materials and methods

Non-human primates

Thirty adult male African Green monkeys (*Cercopithecus aethiops*), also called vervets, were used in this study. Males

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were chosen because they model hypercholesterolaemia better than females.^{7–10}

The vervets were acquired as described in detail previously.³ Fifteen vervets, never exposed to a Western-type atherogenic diet, were recruited from the Medical Research Council (MRC) Primate Unit's in-house breeding colony and 15 were caught with permission from Nature Conservation. The wild-caught males were first quarantined and then conditioned for 3.5 months to their new environment before the study commenced. The average bodyweight of the 30 vervets at baseline was 5.39 ± 0.92 kg (range 3.60–7.00). The vervets were housed permanently in a room within a closed unit. Individuals were kept in stainless steel cages and had access to a large exercise cage for 24 h/week. Olfactory, auditory and visual contacts with peers were not restricted.

Experimental design

The experimental design is shown in Fig. 1. The 30 vervets were stabilised for 6 weeks on a moderate-fat (28%E; 11%E from lard) moderate-cholesterol (26 mg cholesterol/1000 kJ) diet (MFD) with a polyunsaturated to saturated fatty acid ratio (P/S) of 0.4. At the same time, 20 adult male vervets, also never exposed to a Western-type atherogenic diet, were fed a high-fat (34%E) high-cholesterol (98 mg cholesterol/1000 kJ) diet (HFD) with a P/S ratio of 0.6 for 6 weeks. Fasting blood samples were collected from all 50 vervets for plasma lipid measurements. The 30 vervets receiving the MFD were stratified into three comparable experimental groups of 10 each according to their LDL and high-density lipoprotein (HDL) cholesterol concentrations, bodyweight and origin (colony-bred or wild-caught). One group continued with the MFD in which 11%E was derived from lard (MFD-AF); in the other two groups the lard was substituted isocalorically with either SO (MFD-SO) or PO (MFD-PO).

The three groups were fed the respective experimental diets for 24 months and fasting blood samples were collected at 6-monthly intervals (6, 12, 18 and 24 months) for lipid analysis.

Composition of diets

The MFD consisted of skimmed milk powder, egg powder, precooked maize meal and maize kernels. The main sources

of fat were beef tallow as well as the experimental fat (either lard, SO or PO). No extra synthetic cholesterol was added to the diets. The diets were supplemented daily with minerals and vitamins to optimise the micronutrient intake of the vervets. Water was added to the ingredients, mixed to a stiff porridge and formed into food patties. Vervets were fed one patty in the morning and one in the afternoon to meet the energy and nutrient prescription. In addition, a piece of raw apple (~70 g) was supplied daily to each vervet. The suppliers of the eggs, beef tallow, lard, maize and fruit were constant.

The composition and preparation of the high-fat diet was as described in detail previously.³ Briefly, the HFD consisted of full cream milk powder, egg, chicken, beef, rice, dried beans, cabbage, carrot, potato, cake flour, sugar, salt and banana. The main sources of fat were lard, brick margarine, butter and fat from beef mince and chicken.

Refined, bleached, deodorised (RBD) PO was supplied by the Malaysian Palm Oil Board (Selangor, Malaysia). Refined and deodorized pure sunflower oil (SOMOL brand) was purchased from a local supplier (Cape Oil Products, Cape Town, South Africa). Lard consisted mainly of 23.5% palmitic, 11.9% stearic, 41.8% oleic and 16.4% linoleic acids (% by weight as determined by gas-liquid chromatography). The major fatty acids in beef tallow were 27.3% palmitic, 22.7% stearic, 43.3% oleic and 2.6% linoleic. PO contained 37.9% palmitic, 4.1% stearic, 44.7% oleic and 10.2% linoleic acids. The composition of sunflower oil was 6.0% palmitic, 3.4% stearic, 29.8% oleic and 58.9% linoleic acids.

The MRC Food Composition tables¹¹ were used to compile the diets and calculate the energy and nutrient content (Table 1). The total fat, cholesterol and fatty acid content of the diets were checked at regular intervals ($n = 7$) by laboratory analyses of a daily food portion of each diet, as described in detail previously.³

Dietary compliance was checked by weighing individual food portions before and the amount of wasted food after the vervets were fed. This was done on three consecutive days at regular intervals ($n = 5$).

Plasma lipids and lipoproteins

Isolation of plasma and plasma lipoproteins from fasting blood samples and all methods for measuring concentrations of total cholesterol (C), triacylglycerol (TAG), phospholipid (PL), apolipoprotein (apo) AI and LDL apo B were as described previously.³ The LDL molecular weight ($\text{g}/\mu\text{mol}$), a measure of particle size, was calculated² as described previously.³ Molecular weights (g/mol) used to calculate the number of molecules per LDL particle were: esterified cholesterol (CE), 651.1; unesterified (free) cholesterol (FC), 386.7; TAG, 885.4 for low and 879.4 for high P/S diets; and PL, 786.1. A molecular weight of 512 000 g/mol was used for apo B.

Ethics

The study was approved by the Ethics Committee for Research on Animals of the South African Medical Research Council. The MRC guidelines on ethics for medical research

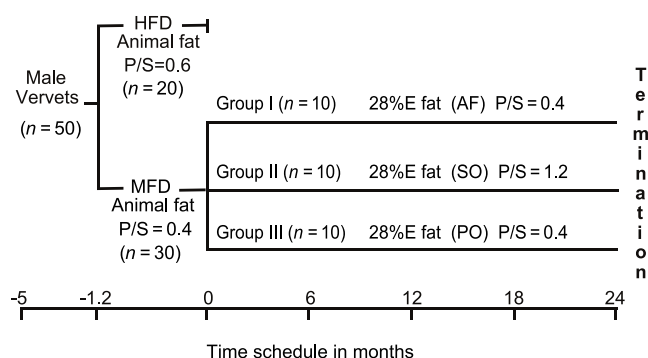


Figure 1. Diagram of the experimental design. AF, animal fat (lard); E, energy; HFD, high-fat diet; MFD, moderate-fat diet; PO, palm olein oil; P/S, polyunsaturated to saturated fatty acid ratio; SO, sunflower oil.

and the national code for animal use in research, education, diagnosis and testing of drugs and related substances in South Africa was followed.

Statistical analysis

Data were analysed with the SAS package for Unix (version 8; SAS Institute, Cary, NC, USA). A two-sample parametric *t*-test was used to compare the baseline measurements of the MFD ($n = 30$) and HFD ($n = 20$) groups. Data were first tested for homogeneity of variance and depending on whether this was equal or unequal, the Pooled *t*-test or Satterthwaite *t*-test, respectively, were used to indicate group differences (Tables 2,3). A repeated-measures analysis of variance was used to model the plasma and LDL lipid profile on the experimental factors group, time and group–time interaction, as well as the baseline measurement as covariate (Tables 4,5). Statistical significance was defined at $P < 0.05$ and results are presented as mean \pm SD.

Results

Matching

The three diets (MFD-AF, MFD-SO and MFD-PO) were applied to well-balanced adult males in terms of bodyweight, all the LDL component concentrations (Table 4; baseline values), as well as mean plasma and HDL total cholesterol concentrations (results not shown).

Diets and compliance in the moderate-fat groups

Regular laboratory analyses of daily food portions of the MFD showed that a constant composition in terms of total fat and cholesterol was maintained over the 24-month experimental period. The only marked difference between the moderate-fat experimental diets was the higher P/S ratio due to the sunflower oil (linoleic acid, C18:2) of the MFD-SO group (Table 1). Bodyweights over 24 months were not significantly different between groups (Table 4; no group–time interaction) indicating adequate energy intake. Dietary compliance during the 24 months was good and minimal food was wasted, as indicated by the percentage of food wasted (mean \pm SD): MFD-AF, 1.6 ± 2.0 ; MFD-SO, 4.5 ± 6.0 ; and MFD-PO, 2.7 ± 2.5 . The HFD differed from the MFD with regard to the quantities of total fat, cholesterol and *trans*-fatty acids.

Effect of high-fat and moderate-fat diets on plasma lipids after 6 weeks

Plasma lipid, lipoprotein and apolipoprotein concentrations. As expected, the HFD group had significantly higher mean plasma, LDL, HDL and very low-density lipoprotein (VLDL) + intermediate-density lipoprotein (IDL) total cholesterol, LDL component, and plasma apo AI concentrations, as well as a higher mean LDL-C : HDL-C ratio compared to the MFD group after 6 weeks (Table 2). Mean bodyweights

Table 1. Energy content and nutrient composition of the experimental diets per vervet per day

	High-fat diet		Moderate-fat diet	
		AF	SO	PO
Energy (kJ)‡	2289	2352	2348	2345
Protein (%E)	15.1	12.6	12.7	12.8
Total (g)	20.3	17.5	17.6	17.6
Animal (g)	14.5	11.3	11.3	11.3
Carbohydrate (%E)	50.7	59.2	59.2	59.3
Total (g)	62.5	73.3	73.2	73.2
Dietary fibre (g)	5.7	8.6	8.6	8.6
Fat (%E)	34.3	28.2	28.0	27.9
Total (g)†	21.2 \pm 1.4	17.9 \pm 0.8	17.8 \pm 1.2	17.7 \pm 1.0
% fat from oil§	–	39.1	39.3	39.5
Cholesterol (mg)†	224 \pm 58	60 \pm 11	57 \pm 8	60 \pm 5
Cholesterol (mg/1000 kJ)	98	26	24	26
Fatty acids (g)†				
Saturated	7.01 \pm 1.42	6.48 \pm 0.83	5.31 \pm 0.83	6.80 \pm 0.85
C14:0	0.62 \pm 0.31	0.26 \pm 0.09	0.22 \pm 0.09	0.25 \pm 0.07
C16:0	4.04 \pm 0.74	3.52 \pm 0.57	2.73 \pm 0.50	4.44 \pm 0.67
C18:0	2.14 \pm 0.48	2.63 \pm 0.39	2.25 \pm 0.41	2.01 \pm 0.38
Monounsaturated	6.13 \pm 1.10	6.36 \pm 1.16	6.02 \pm 0.87	6.26 \pm 0.92
C18:1	5.76 \pm 1.02	6.03 \pm 1.09	5.87 \pm 0.85	6.10 \pm 0.88
Polyunsaturated	4.03 \pm 0.90	3.00 \pm 0.47	6.11 \pm 0.91	2.60 \pm 0.61
C18:2	3.57 \pm 0.85	2.60 \pm 0.33	5.95 \pm 0.87	2.43 \pm 0.56
<i>Trans</i> -fatty acids (g)	1.51	0.01	0.01	0.01
P/S†	0.58 \pm 0.06	0.46 \pm 0.03	1.16 \pm 0.09	0.38 \pm 0.06

All values as obtained from the MRC Food Composition Tables,¹¹ except †those that were based on laboratory analyses representing the mean \pm SD of seven samples. ‡Energy was calculated by multiplying the weight (g) of fat, protein and carbohydrate + fibre by 37, 17 and 17 kJ, respectively. §All oils. AF, lard; E, energy; PO, palm olein oil; P/S, polyunsaturated to saturated fatty acid ratio; SO, sunflower oil.

and mean TAG concentrations in plasma and LDL were not significantly different between the HFD and MFD groups.

LDL particle characteristics. Results for LDL particle size and composition of the HFD and MFD groups are given in Table 3. The HFD group had significantly fewer TAG and significantly more unesterified cholesterol molecules per LDL particle compared to the MFD group. The mean number of phospholipid and esterified cholesterol molecules per LDL particle, as well as the mean LDL particle size (measured as LDL molecular weight) were not significantly different between the HFD and MFD groups.

Effect of a moderate-fat diet on LDL component concentrations and particle characteristics over 24 months
Low-density lipoprotein results of one individual in the MFD-PO group were excluded from the 18-month data set because of errors relating to analytical problems. For all the other time points the data set is complete; $n = 10$ per group.

LDL component concentrations. The effect of a moderate-fat diet containing either of the experimental fats (AF, SO, or PO) on bodyweight and LDL component concen-

trations over 24 months is given in Table 4. The most important result was that there was not a significant group–time interaction for bodyweight or any LDL component concentration, and therefore no significant time-specific group differences at 6, 12, 18 or 24 months.

The baseline measurement for bodyweight and for all the LDL components, except for TAG, had a significant effect on the values of the measurements at 6, 12, 18 and 24 months. The LDL-C, FC, CE, total phospholipid (TPL) and apo B concentrations, as well as bodyweight, showed a significant time effect; that is, the overall mean for the three groups ($n = 30$) changed over time. The LDL-TPL concentration showed a significant group effect, which means that the overall group means over time were at different concentration levels.

LDL particle characteristics. The effect of a MFD containing either of the experimental fats (AF, SO or PO) on LDL particle size and composition over 24 months is given in Table 5. There was not a significant group–time interaction for the mean number of phospholipid, TAG, unesterified and esterified cholesterol molecules per LDL particle, nor for the LDL particle size. The baseline measurement was

Table 2. Effect of high-fat and moderate-fat diets on plasma lipids, lipoproteins and apolipoproteins in vervets after 6 weeks

Variable	High-fat diet ($n = 20$)	Moderate-fat diet (AF) ($n = 30$)	<i>P</i> -value
Bodyweight (kg)	5.67 ± 0.71	5.39 ± 0.92	0.2583
Plasma-C (mmol/L)	8.94 ± 2.03	5.28 ± 0.95	<0.0001
VLDL + IDL-C (mmol/L)	0.63 ± 0.42	0.23 ± 0.13	0.0005
HDL-C (mmol/L)	1.66 ± 0.41	2.29 ± 0.52	<0.0001
Plasma-TAG (mmol/L)	0.40 ± 0.22	0.41 ± 0.11	0.8406
Plasma apo AI (g/L)	0.96 ± 0.27	1.27 ± 0.25	0.0001
LDL-C (mmol/L)	6.19 ± 1.84	2.58 ± 0.73	<0.0001
LDL-FC (mmol/L)	1.44 ± 0.39	0.54 ± 0.17	<0.0001
LDL-CE (mmol/L)	4.75 ± 1.53	2.04 ± 0.59	<0.0001
LDL-TPL (mmol/L)	36.99 ± 8.24	16.67 ± 4.13	<0.0001
LDL-TAG (mmol/L)	0.12 ± 0.05	0.10 ± 0.03	0.2403
LDL apo B (g/L)	0.62 ± 0.17	0.28 ± 0.07	<0.0001
LDL-C : HDL-C	4.13 ± 1.88	1.18 ± 0.49	<0.0001

All values are given as mean ± SD. AF, lard; apo, apolipoprotein; C, total cholesterol; CE, cholesteryl ester (esterified cholesterol); FC (free) unesterified cholesterol; HDL, high-density lipoprotein; IDL, intermediate-density lipoprotein; LDL, low-density lipoprotein; TAG, triacylglycerol; TPL, total phospholipid; VLDL, very low-density lipoprotein.

Table 3. Effect of high-fat and moderate-fat diets on low-density lipoprotein particle characteristics in vervets after 6 weeks

Variable	High-fat diet ($n = 20$)	Moderate-fat diet (AF) ($n = 30$)	<i>P</i> -value
Molecules/LDL particle			
Phospholipid	1231 ± 134	1221 ± 153	0.8038
Triacylglycerol	105 ± 61	190 ± 47	<0.0001
Unesterified cholesterol	1215 ± 272	996 ± 197	0.0018
Esterified cholesterol	3905 ± 537	3753 ± 421	0.2673
LDL MW (g/μmol)	4.59 ± 0.38	4.47 ± 0.39	0.3030

All values are given as mean ± SD. AF, lard; LDL MW, low-density lipoprotein molecular weight.

Table 4. Effect of a moderate-fat diet containing either of the experimental fats, lard, sunflower oil or palm olein oil on bodyweight and low-density lipoprotein component concentrations in vervets over 24 months

Variable	Groups (n = 10)	Time (months)						Significance of effects (P-values)			
		BL	6	12	18†	24	BL-value	Group	Time	Gr-Time	
Bodyweight (kg)	MFD-AF	5.22 ± 0.80	5.67 ± 0.38	5.48 ± 0.35	5.65 ± 0.30	5.60 ± 0.34	<0.0001	0.8615	0.0060	0.3137	
	MFD-SO	5.46 ± 1.08	5.74 ± 0.75	5.67 ± 0.72	5.65 ± 0.68	5.75 ± 0.64					
LDL-C (mmol/L)	MFD-PO	5.48 ± 0.94	5.80 ± 0.59	5.59 ± 0.54	5.67 ± 0.51	5.60 ± 0.51	<0.0001	0.2140	<0.0001	0.2399	
	MFD-AF	2.55 ± 0.62	3.05 ± 0.65	3.62 ± 1.21	2.83 ± 0.83	3.27 ± 1.16					
LDL-FC (mmol/L)	MFD-SO	2.59 ± 0.85	3.06 ± 1.91	3.10 ± 1.96	2.41 ± 1.14	2.38 ± 1.03	<0.0001	0.1707	<0.0001	0.0891	
	MFD-PO	2.60 ± 0.77	2.90 ± 0.80	3.02 ± 0.79	2.46 ± 0.56	2.78 ± 0.82					
LDL-CE (mmol/L)	MFD-AF	0.53 ± 0.15	0.72 ± 0.13	0.84 ± 0.24	0.67 ± 0.21	0.84 ± 0.31	<0.0001	0.2410	<0.0001	0.2936	
	MFD-SO	0.57 ± 0.19	0.73 ± 0.43	0.75 ± 0.44	0.57 ± 0.27	0.63 ± 0.22					
LDL-TPL (mmol/L)	MFD-PO	0.51 ± 0.17	0.70 ± 0.17	0.74 ± 0.19	0.51 ± 0.17	0.79 ± 0.26	<0.0001	0.0224	<0.0001	0.2605	
	MFD-AF	2.02 ± 0.51	2.33 ± 0.52	2.77 ± 0.97	2.17 ± 0.63	2.43 ± 0.86					
LDL-TAG (mmol/L)	MFD-SO	2.02 ± 0.67	2.33 ± 1.48	2.35 ± 1.52	1.84 ± 0.87	1.75 ± 0.82	0.3829	0.0523	0.1736	0.8783	
	MFD-PO	2.09 ± 0.65	2.19 ± 0.63	2.28 ± 0.61	1.95 ± 0.48	1.98 ± 0.62					
LDL apo B (g/L)	MFD-AF	15.87 ± 3.83	18.18 ± 3.37	22.30 ± 5.98	17.98 ± 5.65	16.78 ± 5.09	<0.0001	0.1127	0.0018	0.2895	
	MFD-SO	16.95 ± 4.53	17.34 ± 10.05	17.19 ± 8.05	15.66 ± 5.54	13.79 ± 5.21					
LDL apo B (g/L)	MFD-PO	17.18 ± 4.31	16.96 ± 4.30	17.22 ± 4.30	14.34 ± 2.11	14.51 ± 3.61	<0.0001	0.1127	0.0018	0.2895	
	MFD-AF	0.08 ± 0.02	0.10 ± 0.03	0.12 ± 0.03	0.13 ± 0.03	0.13 ± 0.03					
LDL apo B (g/L)	MFD-SO	0.11 ± 0.02	0.11 ± 0.03	0.12 ± 0.02	0.13 ± 0.03	0.13 ± 0.03	0.3829	0.0523	0.1736	0.8783	
	MFD-PO	0.11 ± 0.03	0.15 ± 0.07	0.14 ± 0.04	0.15 ± 0.06	0.14 ± 0.02					
LDL apo B (g/L)	MFD-AF	0.27 ± 0.06	0.32 ± 0.08	0.35 ± 0.10	0.31 ± 0.11	0.37 ± 0.10	<0.0001	0.1127	0.0018	0.2895	
	MFD-SO	0.28 ± 0.08	0.32 ± 0.20	0.32 ± 0.17	0.26 ± 0.11	0.28 ± 0.08					
LDL apo B (g/L)	MFD-PO	0.29 ± 0.07	0.31 ± 0.10	0.33 ± 0.07	0.26 ± 0.06	0.32 ± 0.08					

All values are given as mean ± SD. †The results of one individual in the MFD-PO group were excluded from the 18-month data set, n = 9 (see text). apo, apolipoprotein; AF, lard; BL, baseline; C, total cholesterol; CE, cholesteryl ester (esterified cholesterol); FC, (free) unesterified cholesterol; Gr-time, group-time interaction; LDL, low-density lipoprotein; MFD, moderate-fat diet; PO, palm olein oil; SO, sunflower oil; TAG, triacylglycerol; TPL, total phospholipid.

Table 5. Effect of a moderate-fat diet containing either of the experimental fats, lard, sunflower oil or palm olein oil on low-density lipoprotein particle characteristics in vervets over 24 months

Variable	Groups (<i>n</i> = 10)	Time (months)						Significance of effects (<i>P</i> -values)			
		BL	6	12	18 [†]	24	BL-value	Group	Time	Gr-Time	
Molecules/LDL particle Phospholipid	MFD-AF	1206 ± 120	1162 ± 104	1344 ± 385	1160 ± 76	907 ± 69	0.8929	0.4420	<0.0001	0.0908	
	MFD-SO	1224 ± 171	1103 ± 153	1120 ± 311	1237 ± 190	975 ± 126					
Triacylglycerol	MFD-PO	1231 ± 177	1142 ± 239	1074 ± 210	1156 ± 208	914 ± 131	0.0305	0.5029	0.0002	0.7399	
	MFD-AF	162 ± 43	172 ± 56	187 ± 49	232 ± 101	185 ± 61					
	MFD-SO	205 ± 42	201 ± 87	219 ± 79	298 ± 110	254 ± 103					
	MFD-PO	203 ± 46	259 ± 129	237 ± 97	323 ± 185	245 ± 107					
Unesterified cholesterol	MFD-AF	1017 ± 153	1168 ± 113	1256 ± 146	1095 ± 81	1145 ± 163	0.1887	0.9261	0.0075	0.1944	
	MFD-SO	1025 ± 124	1175 ± 185	1177 ± 197	1113 ± 184	1120 ± 109					
Esterified cholesterol	MFD-PO	947 ± 287	1188 ± 167	1164 ± 213	1025 ± 261	1246 ± 272					
	MFD-AF	3873 ± 327	3748 ± 260	4068 ± 462	3589 ± 376	3302 ± 346	0.0236	0.6575	<0.0001	0.1839	
	MFD-SO	3648 ± 435	3707 ± 521	3628 ± 661	3562 ± 383	3026 ± 580					
	MFD-PO	3736 ± 497	3669 ± 432	3577 ± 565	3889 ± 555	3121 ± 447					
LDL MW (g/μmol)	MFD-AF	4.52 ± 0.24	4.47 ± 0.28	4.87 ± 0.58	4.39 ± 0.29	3.98 ± 0.24	0.0813	0.7600	<0.0001	0.1037	
	MFD-SO	4.43 ± 0.44	4.42 ± 0.52	4.40 ± 0.64	4.50 ± 0.28	3.91 ± 0.33					
	MFD-PO	4.46 ± 0.49	4.49 ± 0.55	4.35 ± 0.63	4.64 ± 0.39	3.96 ± 0.39					

All values are given as mean ± SD. [†]The results of one individual in the MFD-PO group were excluded from 18-month data set, *n* = 9 (see text). AF, lard; BL, baseline; Gr-time, group-time interaction; LDL MW, low-density lipoprotein molecular weight; MFD, moderate-fat diet; PO, palm olein oil; SO, sunflower oil.

significant for the mean number of TAG and esterified cholesterol molecules per LDL particle. There was a significant time effect for the mean number of molecules of all the LDL components, as well as the LDL particle size.

Discussion

The main objective of this study was to compare the effects of the amount and different sources of dietary fat on the concentrations and composition of circulating plasma LDL in vervets consuming a MFD (28%E fat) containing either of the experimental fats (11%E), lard, SO or PO for 24 months, and to compare the baseline results of the MFD-AF ($n = 30$) with those from vervets ($n = 20$) fed a HFD (34%E fat).

Our results on LDL composition again confirmed our previous observations in vervets, namely that the increased concentrations of LDL-C seen as a result of consuming a HFD could be accounted for by an increase in the number of circulating LDL particles.^{2,3} This is clearly demonstrated by the number of esterified cholesterol molecules in the LDL particle, which was not statistically different between the high-fat and moderate-fat diets (Table 3). Further, the HFD/MFD LDL apo B ratio was approximately 2.3 while the HFD/MFD LDL-CE was also 2.3, indicating that the HFD increased the number of LDL particles more than two-fold because each particle contains one molecule of apo B.^{12,13} Enrichment of the LDL particles with cholesterol was not seen in the present study and confirmed our previous observations.^{2,3}

Our results are, however, in contrast to the findings of others reporting enrichment of LDL particles with cholesterol esters in non-human primates fed semisynthetic diets containing added amounts of cholesterol and fat from sources such as butter, lard, coconut oil, fish oil or corn oil.^{4–6,14,15} The diets used in our trials consisted entirely of natural cooked foods generally used by Westernised people. The main fat sources of the HFD were lard, brick margarine, butter and fat from beef mince and chicken, as described previously.³ No extra synthetic cholesterol was added to the diet, which contained considerably less cholesterol than that reported,^{6,14} namely 26–98 mg/1000 kJ as compared to 177–239 mg/1000 kJ, respectively.

The significantly lower number of TAG molecules in the LDL particles of vervets consuming the HFD compared to vervets consuming the MFD (AF) is in agreement with our results reported previously^{2,3} (Table 3). Associated with this significant loss of core TAG molecules was a significant increase in surface FC molecules. To what extent this will affect LDL metabolism and atherogenesis under these conditions is unclear. It was reported previously that the number of TAG molecules per LDL particle is negatively associated, while plasma LDL-FC and LDL-CE are positively associated, with measurements and scores of atherosclerosis in this non-human primate model.² Based on the composition of the HFD and MFD (AF), one could conclude that the major differences between these two diets are to be found in the total fat, cholesterol and *trans* fatty acid content. Whether these differences between the two diets could account for the

observed differences in LDL particle TAG and FC content cannot be excluded.

Results of a MFD containing either of the experimental fats (lard, SO or PO) showed no significant group–time interaction for bodyweight or any LDL component concentration, mean LDL molecular weight or LDL particle composition. It could therefore be concluded that the effects of the AF, SO and PO on LDL concentrations and LDL composition were not significantly different.

Based on these observations it is justified to assume that the relative risk for atherosclerosis when consuming a moderate-fat diet, namely MFD-AF, MFD-SO or MFD-PO, will be similar. Whether this is indeed the case needs to be established.

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