Dietary Palmitic and Oleic Acids Exert Similar Effects on Serum Cholesterol and Lipoprotein Profiles in Normocholesterolemic Men and Women

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To compare the effects of dietary palmitic acid (16:0) vs oleic acid (18:1) on serum lipids, lipoproteins, and plasma eicosanoids, 33 normocholesterolemic subjects (20 males, 13 females; ages 22–41 years) were challenged with a coconut oil-rich diet for 4 weeks. Subsequently they were assigned to either a palm olein-rich or olive oil-rich diet followed by a dietary crossover during two consecutive 6-week periods. Each test oil served as the sole cooking oil and contributed 23% of dietary energy or two-thirds of the total daily fat intake. Dietary myristic acid (14:0) and lauric acid (12:0) from coconut oil significantly raised all the serum lipid and lipoprotein parameters measured. Subsequent one-to-one exchange of 7% energy between 16:0 (palm olein diet) and 18:1 (olive oil diet) resulted in identical serum total cholesterol (192, 193 mg/dl), low-density lipoprotein cholesterol (LDL-C) (130, 131 mg/dl), high-density lipoprotein cholesterol (HDL-C) (41, 42 mg/dl), and triglyceride (TG) (108, 106 mg/dl) concentrations. Effects attributed to gender included higher HDL in females and higher TG in males associated with the tendency for higher LDL and LDL/HDL ratios in men. However, both sexes were equally responsive to changes in dietary fat saturation. The results indicate that in healthy, normocholesterolemic humans, dietary 16:0 can be exchanged for 18:1 within the range of these fatty acids normally present in typical diets without affecting the serum lipoprotein cholesterol concentration or distribution. In addition, replacement of 12:0+14:0 by 16:0+18:1, but especially 16:0 or some component of palm olein, appeared to have a beneficial impact on an important index of thrombogenesis, i.e., the thromboxane/prostacyclin ratio in plasma.

Abbreviations: BMI = body mass index, FA = fatty acid(s), HDL-C = high-density lipoprotein cholesterol, LDL-C = low-density lipoprotein cholesterol, LDL_r = LDL receptor, $PGF_{1\alpha}$ = prostaglandin $F_{1\alpha}$, PORIM = Palm Oil Research Institute of Malaysia, RBD = refined, bleached and deodorized, SFA = saturated fatty acid, TC = total cholesterol, TG = triglyceride(s), TXB_2 = thromboxane B_2, VLDL = very-low-density lipoprotein, 12:0 = lauric acid, 14:0 = myristic acid, 16:0 = palmitic acid, 18:1 = oleic acid, 18:2 = linoleic acid

INTRODUCTION

Interest in the ability of the various dietary fatty acids (FA) to modulate the plasma cholesterol concentration has been rekindled by the realization that specific lipoproteins are differentially affected by the relative concentration of these FA. In addition, the original observation that saturated fatty acids (SFA) may not contribute equally to the cholesterolemic effect [1] has been reaffirmed by the suggestion that myristic acid (14:0) appeared to be primarily responsible for elevating the cholesterol concentration in normocholesterolemic monkeys when diets were not complicated by simultaneous cholesterol feeding [2]. Subsequent studies demonstrated that palmitic (16:0) and oleic acids (18:1) were equivalent in terms of their effect on plasma total cholesterol (TC) and similar to linoleic acid

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(18:2) regarding low-density lipoprotein (LDL) concentration and metabolism, again in normocholesterolemic monkeys [3]. Similar results can be inferred from other human studies where normocholesterolemic subjects were evaluated [4,5].

On the other hand, in humans with cholesterol concentrations > 225 mg/dl, dietary 16:0 was cholesterolemic relative to either dietary 18:1 or 18:2, both of which appeared to have similar ability in lowering TC and LDL cholesterol (LDL-C) concentrations [6]. This not only contrasts with the data in normocholesterolemic monkeys just mentioned, but conflicts with the neutrality of 18:1 vs the cholesterol-lowering effect of 18:2 demonstrated by Keys et al [7] and Hegsted et al [1]. However, the latter investigators usually manipulated the percent of dietary energy from the specific FA in question within the range normally encountered in human diets, as opposed to the atypical distribution of FA (e.g., percent energy > 10% from 18:2 or > 20% from 18:1) in recent studies [6]. The theoretical significance of the relative FA distribution has been emphasized previously [8,9].

In a previous human study [10] we initiated examination of certain FA plasma cholesterol relationships and found that a diet rich in lauric acid (12:0)+14:0 (coconut oil) was markedly cholesterolemic relative to one rich in 16:0+18:1 (palm olein) which produced substantially higher cholesterol concentrations than a diet containing high 18:2 (corn oil). To more closely examine the 16:0 vs 18:1 impact on lipoproteins, we designed a similar study in humans where the major source of dietary fat was exchanged between coconut oil (hypercholesterolemic control) and palm olein or olive oil. The latter two oils were selected to compare an exchange between 16:0 and 18:1, while 18:2 and "other" FA were held relatively constant. In addition, both 16:0 and 18:1 were fed within the range of energy normally contributed by these two FA in typical human diets.

METHODS

Subjects

A total of 39 male and female adult volunteers were screened by the combined use of a questionnaire, a physical examination by a physician, and biochemical liver-function and renal-function tests. Finally, 33 volunteers (20 males, 13 females; ages 22–41 years) were selected based on the following inclusion criteria: 1) normolipidemic: serum TC <220 mg/dl, fasting TG <190 mg/dl; 2) normotensive: systolic pressure <140 mm Hg, diastolic pressure <90 mm Hg; 3) non-obese: body mass index (BMI) <28 kg/m^2; 4) free from hepatic, renal or bleeding disorders; and 5) willingness to adhere to the dietary guidelines provided in the study. Written informed consent was obtained from every subject. The study protocol was approved by the National Committee on Ethics for Medical Research.

Experimental Design

The subjects were free-living but had to consume the daily cooked meal (lunch) prepared for them, keep personal dietary records, use only the test oil provided for cooking purposes at home, and adhere to specific dietary guidelines provided in the study. Two weeks before the start of the trial, the habitual dietary intake of every subject was estimated on 2 separate days by two research dietitians using the 24-hour recall technique [11]. During the study the mean daily energy intake by nutrient class was generated from diet records and proximate analysis of the two daily meals consumed at the Palm Oil Research Institute of Malaysia (PORIM) and at home.

Energy intake and nutrient composition were determined using a computerized database of 267 common Malaysian foodstuffs, the composition of which were obtained from food composition tables (Nutrient Composition of Malaysian Foods, ASEAN Food Habits Project, Malaysia, 1988) and analysis of selected items by chemists at the Malaysian Institute for Medical Research. Average servings of the six PORIM menus and household meals were also analyzed for a complete FA profile [12] (Table 1).

During the first intervention period subjects underwent 4 weeks of standardization when their meals were prepared with refined, bleached and deodorized (RBD) coconut oil as cooking oil both at the PORIM canteen and at home. At the end of this period, subjects were stratified according to sex, race, serum TC response, TG, and menstrual cycles, and then randomly allocated to one of two dietary fats,

Table 1. Dietary Fatty Acid Profiles

Fatty acid	Coconut	Palm	Olive
	Percent distribution		
<12:0	5.0 (1.7)	0.1 (—)	0.1 (—)
12:0	36.8 (12.5)	0.9 (0.3)	0.2 (0.1)
14:0	16.0 (5.4)	1.5 (0.5)	0.5 (0.2)
16:0	14.5 (4.9)	39.5 (13.4)	18.6 (6.3)
16:1	0.7 (0.2)	0.7 (0.2)	1.6 (0.5)
18:0	4.0 (1.4)	5.2 (1.8)	5.5 (1.9)
18:1	10.8 (3.7)	40.5 (13.8)	62.8 (21.3)
18:2	2.7 (0.9)	10.4 (3.5)	7.7 (2.6)
18:3	0.1 (—)	0.5 (0.2)	0.8 (0.3)
Other	9.4 (3.2)	0.7 (0.2)	2.2 (0.7)

Portions of the daily PORIM and household menus were collected as consumed, homogenized, Soxhlet-extracted and analyzed by GLC. Values represent the mean of several such analyses.
( ) represents % of total dietary energy.

i.e., olive oil or RBD palm olein. Diets were prepared as previously described [10]. Briefly they represented a 6-day menu rotation with all the fat used for cooking and baking supplied from a single source. It is apparent from the FA profile of the diets (Table 1) that approximately one-half of the dietary kilocalories from 16:0 during the palm olein period were replaced by 18:1 during the olive oil period, (i.e., 7% total dietary energy exchanged), with 18:2 held relatively constant between 2.6–3.5% of kilocalories. Total dietary fat represented approximately 34% dietary calories. Group A (10 males, 7 females) initially received virgin olive oil, while group B (10 males, 6 females) ate meals cooked with RBD palm olein (the main cooking oil in Malaysia). After 6 weeks a crossover of the test oils was followed by a second dietary period of 6 weeks.

Two cooks employed for the study used 500 ml of the designated cooking oil (RBD palm olein, virgin olive oil or RBD coconut oil) to prepare lunch for each group of 16–17 people. Skimmed milk was used in place of coconut milk for the preparation of curries. During weekdays (Monday–Saturday), the subjects consumed the major meal (lunch) at the PORIM canteen. Food served was based on six rotating menus which were developed around subjects' preferences. The subjects were given specific servings for certain food items, such as chicken, fish and egg (e.g., one large piece each), but ate servings of rice, beef, squid and leafy vegetables according to their tastes and needs. Their body weights were monitored at weekly intervals to ensure no appreciable change during the study.

For each dietary period the subjects were provided with the test oil to serve as the sole home cooking oil. With each succeeding period, excess cooking oil used in the previous period was retrieved from households to preclude it continued use. All subjects kept daily dietary records including a cooking oil log book at home. Adherence to the guidelines by all subjects was monitored by two research dietitians who interviewed each subject at least twice at PORIM and once in the subject's home per dietary period. At this time dietary records and household use of the test oil in food preparation were monitored.

At entry and at the end of each dietary period, 8–10 ml fasting venous blood was collected from every subject for the analysis of serum lipids and plasma thomboxane (TXB_2) and prostacyclin ($PGF_{1\alpha}$).

Biochemical Measurements

Fasting serum TC, high density lipoprotein cholesterol (HDL-C) following phosphotungstic acid and magnesium chloride precipitation, and TG were analyzed using enzymatic diagnostic kits purchased from Human Diagnostica (Germany). LDL-C was estimated by the formula of Friedewald [13].

TXB_2 and $PGF_{1\alpha}$ were measured in collagen-activated plasma prepared by incubating 5 μg collagen (Hormon-Chemie, Munich), in a 10 μl glucose solution with 1.0 ml of citrated whole blood in a 37°C water bath for exactly 5 minutes. The plasma was then separated by centrifugation and stored temporarily at −70°C until analysis. Plasma TXB_2 and 6-keto $PGF_{1\alpha}$ were determined by ^{3}H-radioimmunoassay kits obtained from Amersham (United Kingdom). For the TXB_2 assay, plasma samples were first diluted 100-fold with phosphate buffer from the kit.

Statistical Analysis

Data from the three test periods were analyzed by repeated measures two-way analysis of variance for gender and fat effects and Fischer's protected least significant difference test between periods when a diet effect was encountered [14].

RESULTS

Age and Body Mass

The 20 males (30±4.5 years) averaged almost 3 years older than the 13 females (26.6±3.9 years) and had a slightly greater BMI (22.3±2.8 vs 21.6±3.4), but the latter difference was not significant. BMI indicate a low to modest amount of body fat was present in this Malaysian population. Body weight did not change appreciably throughout the three dietary periods (data not shown).

Dietary Compliance

The record of attendance for lunch served at the PORIM canteen for all subjects averaged 78%. Since this figure did not take into consideration subjects who took leave during the study and adhered to the dietary guidelines at home (based on dietary records), the mean for dietary compliance was estimated to be close to 90%.

In addition, observations made for the household use of the test oils and records of daily entries into the household cooking oil log book supported this estimate of compliance. The daily amounts of cooking oil used per adult (disappearance data) within the home did not differ between test oils with means of 45, 48, and 45 g. These data served as a measure of compliance but were considered too crude an estimate of actual consumption because frying pan loss and plate waste were not accounted for.

Food Consumption

Mean daily energy intake by nutrient class was generated from diet records (Table 2) and proximate analysis of the two daily meals consumed at PORIM and at home (Table 3). The proximate composition generated from these two daily meals accounted for 78% of all energy consumed daily with an additional 22% accounted for

Table 2. Daily Energy Intake and Diet Composition According to Diet Records for All Subjects During the Three Dietary Fat Periods

Diet period	Energy (kcal)	Fat (g)	Protein (g)	CHO (g)
Coconut:				
Males (n = 20)	2196 ± 319[a]	80 ± 16	88 ± 16	281 ± 84
Females (n = 13)	1963 ± 279	74 ± 13	78 ± 17	246 ± 70
Palm:				
Males	2204 ± 373	82 ± 18	89 ± 19	274 ± 96
Females	1998 ± 392	77 ± 14	84 ± 19	243 ± 75
Olive:				
Males	2100 ± 403	78 ± 20	85 ± 20	267 ± 82
Females	1956 ± 380	74 ± 18	81 ± 16	241 ± 88

[a] Mean ± SD.

from small meals or snacks included in dietary records, i.e., the difference between Tables 2 and 3. In the case of fat, approximately 75 g/person of cooking oil "disappeared" in the preparation of the two meals. From our analysis 69 g fat (of an average daily total 77 g) were consumed during these two meals. Of the 69 g, approximately 46 g was thought to represent the test oil, an amount similar to our previous estimate of cooking oil (44 g) used in urban Malaysian diets [15]. Our generous oil allowance provided on a per person basis for cooking was reflected in the slightly oily appearance/taste of the prepared meals which were, nevertheless, well-accepted by the subjects. Accordingly our estimate of 34% of total dietary energy as fat is presumably greater than the fat intake in the typical Malaysian diet.

Serum Cholesterol and Triglycerides

Entry level plasma cholesterol concentrations were <200 mg/dl for both males and females (Table 4), and gender had no effect on this parameter. TC was significantly elevated (18%) during coconut oil consumption compared to either the palm olein or olive oil period. The increase was slightly greater in males than females. Consumption of either palm olein (16:0-rich) or olive oil (18:1-rich) diets resulted in identical TC values and were similar to the entry level concentration. The pattern for LDL-C was identical to that for TC, with males again tending toward higher concentrations than females (Table 4). The gender difference was not significant, but coconut oil caused a significant elevation (20%) compared to both the other dietary periods, which were similar and comparable to the ad libitum (entry) LDL-C concentration.

HDL-C revealed both diet and gender effects with women having significantly higher concentrations than men for all dietary periods. Coconut oil caused a rise in HDL-C (approximately 10%) which was significant when compared to either palm olein or olive oil in men, but only compared to palm olein in women. Entry level values for HDL-C were similar to those achieved with palm olein and olive oil (Table 4).

These changes in lipoprotein cholesterol were reflected in the LDL/HDL ratio which also revealed gender and diet effects. This ratio in males was considerably higher than that in females during all periods. A 10% rise occurred during the coconut oil period compared to the entry level ratio, which, in turn, was slightly higher than the ratio after palm olein and olive oil intake, especially in males. The increase in this ratio during coconut oil consumption was significant when compared to either the palm olein or olive oil periods (Table 4).

Fasting plasma TG concentrations were normal and appreciably higher in males than in females. A rise oc-

Table 3. Proximate Composition of Daily Meals Prepared at the PORIM Canteen and at Home

Type of diet	Dietary component per meal (mean ± SD, n = 6)							
	PORIM canteen				Household			
	Energy (kcal)	Fat (g)	Prot. (g)	CHO (g)	Energy (kcal)	Fat (g)	Prot. (g)	CHO (g)
Coconut	862 ± 85	38.2 ± 4.6	39.0 ± 6.0	94.6 ± 6.5	739 ± 48	31.3 ± 3.7	33.4 ± 5.6	81.3 ± 9.1
Palm	856 ± 67	39.8 ± 3.1	38.8 ± 7.9	90.0 ± 10.1-	743 ± 77	29.1 ± 4.9	35.9 ± 3.3	84.3 ± 12.8
Olive	878 ± 3.5	40.6 ± 74	38.0 ± 11.4	90.3 ± 13.7	783 ± 36	29.2 ± 5.0	35.1 ± 5.5	90.8 ± 4.1

curred with coconut oil intake, especially for males. The concentration declined during palm olein and olive oil consumption, the decrease being significant for both sexes during olive oil intake and for men consuming palm olein (Table 4).

Eicosanoid Metabolites

The estimates of eicosanoid metabolism measured ex vivo in plasma harvested from collagen-activated whole blood revealed effects attributable both to gender and dietary fat intake (Table 5). TXB_2 concentrations were highest during coconut oil intake, which was significant for both sexes when compared to palm olein and in males fed olive oil-rich diets. No gender differences were observed.

The metabolite of prostacyclin, $PGF_{1\alpha}$, was depressed by coconut oil in both males and females by comparison to palm olein. This depression by coconut oil was extreme in females, the concentration being reduced to less than half that observed with either palm olein or olive oil consumption or males consuming coconut oil.

These differences were reflected in the $TXB_2/PGF_{1\alpha}$ ratio such that males consuming the palm olein-rich diet had a significantly lower ratio than that obtained during olive oil intake, and dramatically lower than the elevated ratio encountered during coconut oil consumption. In females the ratio was equally low during both palm olein and olive oil periods, which were approximately one-third that attributed to the striking elevation in this ratio during the coconut oil period, which was, in fact, significantly higher than that in males eating coconut oil.

Table 5. Thromboxane B_2 and Prostacylin ($PGF_{1\alpha}$) Concentrations in Collagen-Activated Plasma Following Three Dietary Fat Periods

	Coconut	Palm	Olive
TXB_2 (ng/ml)			
Males (n = 20) NA	$153 \pm 70^{a,b}$	113 ± 31^{a}	124 ± 41^{b}
Females (n = 13) NA	133 ± 44^{a}	100 ± 39^{a}	107 ± 30^{b}
$PGF_{1\alpha}$ (pg/dl)			
Males NA	$135 \pm 104^{a,x}$	180 ± 53^{a}	145 ± 46
Females NA	$66 \pm 20^{a,b,x}$	156 ± 60^{a}	145 ± 49^{b}
$TXB_2/PGF_{1\alpha}$ ratio			
Males NA	$1446 \pm 558^{a,b,x}$	$641 \pm 89^{a,c}$	$898 \pm 334^{b,c}$
Females NA	$2050 \pm 576^{a,b,x}$	670 ± 175^{a}	780 ± 252^{b}

NA: not available.

[a,b,c] Means sharing common superscript indicate a significant fat effect ($p < 0.05$).

[x] Indicates significant gender effect ($p < 0.05$).

DISCUSSION

These data reveal several interesting and important facts concerning the impact of dietary FA on plasma lipids and eicosanoid metabolism in humans. First, they are original in their comparison of a one-to-one exchange between 16:0 and 18:1 using natural oils in normolipemic subjects (<200 mg/dl cholesterol, <110 mg/dl TG). This comparison strongly suggests that these two dietary FA exerted equivalent effects on both the plasma cholesterol concentration and distribution of cholesterol among lipoproteins in normocholesterolemic subjects, at least when the sub-

Table 4. Plasma Lipid Profiles of All Subjects at Entry and at the End of Each Dietary Period

Criterion	Ad libitum entry	Dietary fat		
		Coconut	Palm	Olive
Plasma cholesterol (mg/dl)				
Males (n = 20)	194 ± 26	$234 \pm 36^{a,b}$	195 ± 26^{a}	197 ± 32^{b}
Females (n = 13)	191 ± 30	$227 \pm 47^{a,b}$	188 ± 27^{a}	188 ± 30^{b}
Plasma triglyceride (mg/dl)				
Males	107 ± 65	$143 \pm 80^{a,b,x}$	125 ± 81^{a}	$126 \pm 59^{b,x}$
Females	72 ± 37	$90 \pm 30^{a,x}$	83 ± 41	$76 \pm 37^{a,x}$
LDL cholesterol (mg/dl)				
Males	136 ± 23	$165 \pm 38^{a,b}$	133 ± 25^{a}	134 ± 31^{b}
Females	130 ± 29	$158 \pm 45^{a,b}$	125 ± 27^{a}	127 ± 30^{b}
HDL cholesterol (mg/dl)				
Males	36 ± 8^{x}	$40 \pm 9^{a,b,x}$	$37 \pm 8^{a,x}$	$37 \pm 7^{b,x}$
Females	46 ± 7^{x}	$51 \pm 8^{a,x}$	$46 \pm 7^{a,x}$	46 ± 8^{x}
LDL/HDL ratio				
Males	3.91 ± 1.08^{x}	$4.32 \pm 1.43^{a,b,x}$	$3.74 \pm 1.09^{a,x}$	$3.71 \pm 1.23^{b,x}$
Females	2.89 ± 0.82^{x}	$3.25 \pm 1.25^{a,b,x}$	$2.80 \pm 0.94^{a,x}$	$2.82 \pm 0.99^{b,x}$

Values represent mean ± SD.

[a,b] Means within rows having common superscript differ significantly ($p < 0.05$).

[x] Significant gender effect ($p < 0.05$).

stitution of 18:1 by 16:0 represented an increase in 16:0 from 6 to 13% of total energy and 18:2 represented about 3% total energy. In that sense our results are similar to those of Becker et al [4] where 16:0 and 18:0 were exchanged for 18:1 (>16% energy exchanged) in subjects with even lower entry concentrations of cholesterol.

Recent human studies [6,16], where dietary 16:0 was found to be cholesterolemic relative to 18:1, differ from the current experiment in at least three respects. First, those diets were fed as liquid formulas, which characteristically generate aberrant results [17]. Such data appear atypical for unknown reasons and have been discounted in major literature reviews [18]. Second, previous subjects were older, more ponderous, and relatively hypercholesterolemic (average >225 mg/dl) and presumably had relatively depressed LDL receptor (LDL_r) activity at the time of study by comparison to our subjects [18]. Third, the percent energy exchanged between 16:0 and 18:1 in the former studies was extreme (>15% en) by comparison to either our exchange (7% en) or what might be achieved through other practical diets (<10% en). This resulted in unusually low 16:0 (2% en) and exceptionally high substitution of 18:1 (32% en) in the previous studies compared to that provided by most practical diets (5–12% en for 16:0 or 8–18% for 18:1). Furthermore, in one of the studies [6] the 18:1-rich diet also provided 50% more energy as 18:2 compared to the 16:0-rich diet, which may have enhanced LDL_r activity in the former situation. However, the last two caveats also pertain to the Becker study [4].

Thus, all things considered, anthropomorphic and associated metabolic differences in subjects under study might best explain the different cholesterolemic response to 16:0 observed in these studies. On one hand, the adiposity of previous subjects has been associated with increased cholesterolemia attributed to down-regulated LDL_r [19,20]. On the other hand, similar to the Becker study [4] and the present human investigation, normocholesterolemic monkeys fed the same three oils (16:0, 18:1, 18:2) in the absence of dietary cholesterol revealed minimal differences in their LDL-C [3], whereas monkeys fed these fats with their LDL_r partially down-regulated by added dietary cholesterol elevated their plasma cholesterol when fed 16:0 in a manner similar to overweight, hypercholesterolemic humans [21]. This apparent discrepancy involving LDL_r activity may reflect metabolism of specific FA because it has been previously found that dietary 16:0+18:1 increased very-low-density lipoprotein (VLDL) apoB transport relative to 12:0+14:0 in vivo [22], in keeping with predictions generated on the basis of FA chain length and degree of unsaturation affecting TG secretion by perfused livers [23]. Thus, any circumstance, e.g., anthropomorphic or dietary, that decreases LDL_r activity would hinder not only LDL removal but clearance of the expanded pool of VLDL remnants generated by 16:0+18:1 as well, leading to expansion of the circulating cholesterol pool [24]. Under such circumstances 16:0 might generate more VLDL remnants than 18:1, since the latter undergoes β-oxidation at a much greater rate than 16:0 [25]. When LDL_r are fully up-regulated (normal healthy host), the greater VLDL remnant load would clear normally and not result in expanded VLDL-LDL cholesterol pool.

Our results differ from predictions generated by the classical Keys-Hegsted regression equations. For example, according to the Keys equation the cholesterol value during palm olein consumption should have been 18–20 mg/dl higher than olive oil. However, if 16:0 is considered neutral, the difference disappears in accord with our observed results. Whereas both Keys and Hegsted considered 18:1 neutral, 16:0 was thought to be minimally cholestrolemic by Hegsted [1] and equal to 12:0 and 14:0 by Keys [7]. It is possible that their interpretations were complicated by use of moderately hypercholesterolemic men (average cholesterol, 225 mg/dl) whose LDL receptors presumably would be partially down-regulated [20]. We think at least two factors contribute to the recurrent discrepancy that numerous investigators have noted between their results and the predictions generated by these classical regressions. One is that the initial cholesterolemia (i.e., LDL_r setpoint) of the study population seems to impact the response, particularly that to 16:0 [3]. The second is the failure of the Keys-Hegsted regressions to recognize the nonlinear plasma cholesterol response associated with dietary 18:2 intake [26]. Keys inferred these points when he noted that his regression applies to individuals with an initial cholesterol of 225 mg/dl and consuming diets with normal P/S ratios (i.e., 0.2–1.0) [8].

Because the main difference in FA between the coconut oil-rich and the palm olein-rich diets was the substitution of 12:0+14:0 for 16:0+18:1, respectively, the inference is that 12:0+14:0 is the primary source of the cholesterolemic activity in normocholesterolemic humans [27], just as it proved to be in monkeys [2]. Also because butter routinely increases cholesterol in monkeys [28] and humans [1,7,29], yet contains much less 12:0 than 14:0, the latter FA would appear to be the common denominator and major contributor to the SFA-induced cholesterolemia in normocholesterolemic primates. Previous studies in humans that suggested 12:0, 14:0 and 16:0 were equally cholesterolemic may have reflected entry level cholesterol values that were somewhat elevated [1,7,29], or the fact that modified or "structured" TG were substituted for natural fats and oils in the comparisons [30,31]. As discussed above and elsewhere [3,22], 16:0 may be cholesterolemic depending on the degree of reduced LDL_r activity in individuals with already elevated plasma cholesterol. To the second point, modified TGs may be metabolized differently from natural TGs and exert a different impact on cholesterol metabolism [32].

The present results pertained even though 200 mg cholesterol/day was consumed by our subjects. Higher cholesterol intake or substantial cholesterol absorption might be expected to elicit different results because excess cholesterol flux eventually decreases LDL_r activity [33], which would decrease VLDL remnant return of the increased VLDL flux associated with the 16:0+18:1 [24]. In fact, cholesterol feeding is often used as a means to demonstrate the SFA effect in animals, and unless dietary cholesterol is added, most so-called SFA fail to raise the plasma cholesterol concentration appreciably [5,21,28,34]. The usual exceptions are butter fat and coconut oil [1,7,28,29], the first of which contributes some dietary cholesterol.

In our study it is noteworthy that coconut oil raised the entry level cholesterol concentration, underscoring the relative potency of 14:0 (+12:0) to raise plasma cholesterol and dispelling the notion that plasma cholesterol routinely decreases during dietary intervention as subjects become more conscious of the dietary fat-cholesterol relationship and unwittingly contribute to the placebo effect and regression toward the mean. It is interesting that during the palm olein period, the subjects in our study scarcely changed from their prestudy, ad libitum plasma cholesterol values, in part because palm olein represents the major source of fat in the typical Malaysian diet [15].

The entry level cholesterol values (192±28) in these subjects averaged 13% higher than the mean value (171± 29) recorded in a previous study from this lab [10] using a slightly different population. Differences that might contribute to this observation are the facts that the present population was on average 6–8 years older and BMI averaged 16% greater than the previous population. Plasma cholesterol is known to increase with age and adiposity [19], and the greater adiposity of the subjects in this study might be expected to exert a positive influence on cholesterol production to reflect negatively on LDL_r [20].

Gender Effects on Lipoproteins

Ours represents one of the few studies to directly compare the plasma lipid response of both males and females during changes in dietary fat saturation. Two gender effects were noteworthy: higher HDL in females and higher TG in males associated with the tendency for higher LDL and higher LDL/HDL ratios in men. However, the basic response to dietary fat was the same, i.e., coconut oil caused cholesterol and TG to increase in a similar fashion in both sexes, except for TG which expanded slightly more in males. In both sexes, palm oil and olive oil elicited identical responses. This suggests that the mechanism of SFA-induced cholesterolemia is the same for both sexes and not complicated by normal levels of sex hormones.

Both diet and gender effects on lipoproteins might reflect differences in LDL_r activity. For example, during coconut oil ingestion the primary increase occurred in LDL-C (about 25%) with a lesser rise in HDL-C (about 10%), consistent with previously reported reductions in LDL fractional catabolic rate and LDL_r activity associated with coconut oil intake [34,35]. In addition, the slightly greater BMI in men is consistent with lower LDL_r activity [16] and the higher plasma LDL, TG, and LDL/HDL ratio observed in males. The lower LDL in females is in keeping with the fact that estrogen increases LDL_r [31].

Eicosanoid Profile

The striking difference in plasma eicosanoids between sexes was not totally unanticipated. The plasma TXB_2 produced by platelets ex vivo was consistent with that predicted from the gender-dietary fat effects on the lipoprotein response, i.e., high LDL (males) favors TXB_2 production by platelets, whereas high HDL (females) tends to inhibit TXB_2 [37]. Thus, coconut oil in males resulted in the highest LDL/HDL ratio and the highest TXB_2 concentration compared to the other two fats. And females with lower LDL/HDL ratios than males tended to have lower TXB_2 for any given fat. Gender differences in the plasma levels of prostacyclin are more puzzling, however, and interpretation is difficult because assessment of whole body biosynthesis (primarily by vascular endothelium and smooth muscle cells) and the implications thereof are still unclear [38]. Nonetheless, the depressed level of prostacyclin in females fed coconut oil was impressive and worthy of further investigation.

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