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ORIGINAL ARTICLE

Postprandial Endothelial Function: The Effect of Various Types of Fat and of Red Wine Intake on Flow-Mediated Dilation in Healthy Volunteers

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KEY WORDS: *fat, red wine, endothelial function, flow-mediated vasodialtion*

List of Abbreviations
ED: Endothelial dysfunction
NO: Nitric oxide
FMD: Flow-mediated dilation
VTI: Velocity time integral
Apo A1: Apolipoprotein A1
Apo B100: Apolipoprotein B-100
Lp(a): lipoprotein (a)

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ABSTRACT

BACKGROUND: Current evidence suggests that postprandial lipid composition may have either atherogenic or anti-atherogenic properties. This study investigated the acute postprandial effect of three different types of fat (saturated, mono-unsaturated, poly-unsaturated) and of red wine upon endothelial function of healthy subjects.

METHODS: We fed 21 healthy young subjects four meals containing 1000 kcal and 50 g fat each. Three of the meals contained different fat sources: yellow cheese (saturated fat-meal A), olive oil (monounsaturated fat-meal B) and margarine (polyunsaturated fat-meal C). The fourth meal contained the same amount of saturated fat as the first one together with 250 cc of red wine (meal D). We measured serum lipoproteins, with the use of standard laboratory techniques. Brachial artery flow-mediated vasodilation (FMD), an index of endothelial function, was also assessed with B-Mode vascular ultrasound. All measurements were performed before, 2 and 4 hours after each meal

RESULTS: All four meals significantly raised plasma triglycerides. Meal A was associated with a decrease in FMD 2 and 4 hours after its intake (16.5±5% vs. 13±4% vs. 12.3±6%; p=**0.030**) while with meal D FMD profoundly decreased 2 hours postprandially with a rapid return to baseline levels at 4 hours (14.3±9.5 vs. 8.6±5.5 vs. 12.6±7.5; p=0.214). This type of response was associated with a concomitant increase of basal brachial artery diameter at 2 hours postpandially (3.7±0.6 mm vs 4.2±0.7 mm vs 3.9±0.7mm at 0, 2 and 4 hours respectively, p=0.001). Meals B and C did not significantly affect FMD.

CONCLUSION: Saturated fat acutely decreases FMD while mono-unsaturated and poly-unsaturated fat do not have any harmful effect on endothelium. The concomitant intake of red wine with saturated fat is associated with a delayed counteracting effect on lipid-induced postprandial endothelial dysfunction.

INTRODUCTION

Endothelial dysfunction (ED) is thought to play a key role in the development of atherosclerosis and predates clinically obvious vascular pathology by many years.[1,2] A

key and quantifiable feature of ED is the inability of arteries and arterioles to dilate fully in response to an appropriate stimulus that stimulates release of vasodilators from the endothelium like nitric oxide (NO). Endothelial dysfunction is commonly associated with decreased NO bioavailability, which is due to impaired NO production by the endothelium and/or increased inactivation of NO by reactive oxygen species.[3] Flow-mediated dilation (FMD) is the most widely used non-invasive test for assessing endothelial function. This technique measures endothelial function by inducing reactive hyperemia via temporary arterial occlusion and measuring the resultant relative increase in blood vessel diameter via ultrasound.[4] As people with endothelial dysfunction have low NO bioavailability, their blood vessels have a decreased capacity to dilate in response to certain stimuli, compared to those with normal endothelial function.

Dyslipidemia, an important risk factor for atherosclerosis, is also closely associated with the development of ED.[5] Fasting lipid profile has been routinely used to define dyslipidemia in both research and clinical grounds. However, dyslipidemia in the postprandial state, the so called postprandial lipemia, has been recently recognized as an important atherogenic factor.[6,7] Indeed, ingestion of a single fatty meal has been shown to acutely impair endothelium dependent vasodilation in healthy volunteers[8] and this has been proposed as a possible mechanism of postprandial lipemia-induced atherosclerosis.[9] However, not all types of fat are considered harmful and some of them may even have a beneficial effect on the vasculature.[10]

To investigate whether postprandial ED can be induced by different types of fat we assessed brachial artery FMD after ingestion of 3 meals with different fat composition by healthy volunteers. A fourth meal containing saturated fat accompanied by red wine was also given to assess how red wine modifies endothelial response to postprandial lipemia.

MATERIALS AND METHODS

STUDY SUBJECTS

Twenty-one healthy volunteers, 18 men and 3 women, aged 33±11.2 years without any known metabolic or cardiovascular disorders were included in the study. Subjects taking systematically agents known to affect endothelial function (nitrates, angiotensin converting enzyme inhibitors, angiotensin blockers, calcium channel blockers and antioxidant vitamins) or habitual smokers were excluded from the study. Written informed consent was obtained from each participant. The study protocol was in accordance with the Declaration of Helsinki (2000) of the World Medical Association and was approved by the Ethics Committee of the Evagelismos Hospital.

STUDY PROTOCOL

Since several factors including temperature, food ingestion, drugs and autonomic tone may affect FMD, all study participants were kept in a comfortable waiting room for 30 minutes and the examinations were subsequently performed in a relatively silent room with normal temperature. They were also instructed to avoid strenuous activity, food and caffeine ingestion for at least 4-6 hours before the examination. All volunteers received the following meals, each containing approximately 1000 Kcal:

1. Meal A containing 50 g of saturated fat (yellow cheese)
2. Meal B containing 50 g of mono-unsaturated fat (olive oil)
3. Meal C containing 50 g of trans-poly-unsaturated fat (margarine)
4. Meal D containing 50 g of saturated fat (yellow cheese) and 300 ml of red wine (12% vol)

The exact composition of each meal is shown in Table 1.

All measurements were performed before the meal intake and were repeated 2 and 4 hours after the meal.

ULTRASONOGRAPHIC EVALUATION OF ENDOTHELIAL FUNCTION

Endothelial function was assessed by ultrasonographic evaluation of the dilatation of the brachial artery after limb ischemia induced by cuff inflation (FMD). The test was repeated after the administration of sublingual nitrates as positive control (non FMD). The sonograph Hewlett-Packard Sonos 2500 equipped with a high frequency vascular transducer (7.5 MHz) was used. It was able to provide B-mode vascular imaging, perform colour and spectral Doppler analysis and allow simultaneous electrocardiographic monitoring in order to ensure that all measurements were obtained in the same phase of the cardiac cycle (end-diastole). The individual was placed in supine position and the brachial artery was located in the antero-medial surface of the right arm within 4-5 cm from the elbow joint. After an adequate image was achieved, the transducer exact position was marked in order to be found again quickly for the subsequent measurements. Baseline blood flow in the brachial artery was assessed by the velocity time integral (VTI) using pulse wave Doppler imaging in the mid portion of the vessel. Subsequently, the blood flow was interrupted using a cuff inflated for 5 minutes in pressure exceeding the systolic arterial pressure by at least 50 mmHg and positioned above the point the transducer was placed. The ischemia that occurs causes the vessels distal to the obstruction to dilate. The subsequent release of the cuff triggers a brief period of high flow to match with the increased capacity of the dilated vessels. Doppler and B-mode images were recorded within 15 seconds after the cuff deflation and VTI was measured again. The above mentioned reactive hyperemia lasts for several minutes and at least 15 minutes later a second B-mode along with Doppler images and VTI

TABLE 1. Nutrition components of the four test meals. Sat: Saturated, Mono-unsat: Mono-unsaturated, Poly-unsat: Poly-unsaturated

ingredients	Quantity (g)	C/H_2O (g)	Protein (g)	Fat				Chol (mg)	Energy (Kcal)
				g	Sat(%)	Mono-unsat(%)	Poly-unsat(%)		
Meal A								1	
EDAM cheese	214	trace	52.22	49.01	62.2	28.9	3.69	54.08	650.56
Boiled vegetable marrow	300	23.7	3	0.9	0	0	0.9	0	99
Boiled potatoes	100	18.3	1.6	0.1	0	0	trace	0	76
Bread	62	30.81	4.84	1.05	30.03	18.8	48.7	0	144.46
Apple	200	23.8	0.6	trace	0	0	0	0	92
Total		96.61	62.26			51.06		154.08	1062.02
Meal B									
Olive oil	50	0	0	43.395	14.7	73	11.5	0	449.5
Yoghurt 2%	400	24.8	20	1	0	0	0	0	180
White of egg	4	0	18	0	0	0	0	0	72
Boiled vegetable marrow	300	23.7	3	0.9	0	0	0.9	0	99
Bread	50	24.85	3.9	0.85	30.3	18.8	48.7	0	116.5
Apple	200	23.8	0.6	trace	0	0 0		0	92
Total		97.15	45.5			52.7		0	1009
Meal C	50								
Margarine	400	0.05	0.05	40.5	33.1	12.6	48.9	0	365
Yoghurt 2%	4	24.8	20	1	0	0	0	0	180
White of egg	300	0	18	0	0	0	0	0	72
Boiled vegetable marrow	50	23.7	3	0.9	0	0	0.9	0	99
Bread	200	24.85	3.9	0.85	30.3	18.8	48.7	0	116.5
Apple		23.8	0.6	trace	0	0	0	0	92
Total		97.2	45.55			43.25		0	924.5
Meal D									
EDAM cheese	214	trace	52.22	49.01	62.2	28.9	3.69	154.08	650.56
Boiled vegetable marrow	300	23.7	3	0.9	0	0	0.9	0	99
Boiled potatoes	100	18.3	1.6	0.1	0	0	trace	0	76
Bread	62	30.81	4.84	1.05	30.3	18.8	48.7	0	144.46
Apple	200	23.8	0.6	trace	0	0	0	0	92
Red wine	300	0.9	0.6	0	0	0	0	0	204
Total		97.51	62.86			51.06		154.08	1266.02

measurement were performed. At that time, 5 mg of sublingual nitroglycerin were administered to provoke vasodilation and evaluate the function of the vascular smooth muscle cells (non - FMD). The nitroglycerin effect reaches its maximum in 3-4 minutes and for the whole period the vessel and its flow were monitored. This provocation test was omitted if the person had a systolic blood pressure lower than 90 mmHg. The whole study was videotaped for subsequent off- line analysis. One of the authors performed measurements blindly. A frame of the B-mode image in end-diastole was chosen and the brachial artery diameter was measured manually with the use of digital calipers. Three measurements at three points approximately 1 cm apart were done and the mean was used for subsequent calculations. Both FMD and non-FMD were expressed as percentage change in vessel diameter from baseline.

BIOCHEMICAL ANALYSIS

Venous blood samples were drawn immediately after each ultrasonographic examination. Full lipid profile analysis (total cholesterol, triglycerides, HDL-C, LDL-C, Apolipoprotein A1 (Apo A1), apolipoprotein B-100 (Apo B100), lipoprotein (a) (lp(a)) was performed for all participants in the laboratory

facilities of the department of Biochemistry of Evagelismos hospital. Total cholesterol, triglycerides and HDL-C were determined by colorimetric enzymatic methods (reactants from ROCHE), while LDL-C was calculated using the Friedewald formula (for triglycerides levels <400 mg/dl). Apolipoprotein A, Apolipoprotein B and Lp(a) were measured by immunoenzymatic assay (antisera from DADE BEHRING GmbH).

STATISTICAL ANALYSIS

Sample size was based on preliminary data, which allowed us to estimate the standard deviation and the difference between the means. For a desired 5% level of significance and an 80% power to detect an actual difference of 2 units in FMD, a sample size of at least 18 patients was considered satisfactory. For the statistical process of the study data, the SPSS for Windows Release 11.0 (SPSS Inc, Chicago, Illinois) was used. All variables were quantitative and were controlled in terms of the normality of the distribution of their values using the Liliefors test and Shapiro-Wilks' test. The comparison of the means for variables following normal distribution was performed with paired t-test, when the variables were two, or the one-way analysis of variance (ANOVA) test for more than two variables. Variables following other than normal distributions were compared using Wilcoxon test (two variables) and Friedman test (more than two variables). The level of statistical significance was defined at 5% ($p<0.05$).

RESULTS

As measurements associated with the different meals were obtained at different times, it was important to assure that baseline values of lipid and functional vessel parameters did not differ significantly among the four individual assessment sessions. Indeed no significant differences were noted (Table 2). While no significant change was noted regarding total cholesterol levels after any of the meals, triglycerides were significantly higher postprandially after each meal with the lowest increase presented after meal C. The concentration of HDL-C was greatly reduced 4 hours after meal D, while a hardly statistically significant decrease was established in relation with meal A. The LDL-C levels were significantly reduced with meals A and D. With regards to apolipoproteins, a progressive reduction of ApoA1 was recorded in association with meal D. ApoB100 concentration was significantly reduced 2 hours after meal C and recovered to its former levels 4 hours post-prandially. Finally, Lp(a) showed a significant decrease 2 hours after meal D remaining unchanged in the 4-hour postprandial measurement (Table 3). The changes in FMD are presented in Table 4. Endothelium-mediated vasodilation was found significantly reduced after the rich in saturated fat meal (meal A). The reduction of FMD with regard to the baseline was statistically significant both at the 2 hour and 4-hour measurements ($Z=2.104$, $P=0.030$ and $Z=2.641$, $P=0.008$ respectively). Consumption of meals rich in mono-unsaturated and trans- poly-unsaturated fat did not affect notably FMD. In meal D, which was accompanied by red wine, FMD was significantly reduced 2 hours postprandially and was increased again 2 hours later (4 hour postprandial measurement). The decrease noted in the first 2 hours was significant in comparison to the baseline as well as the 4-hour assessment ($Z=2.740$, $P=0.006$ and $Z=2.057$, $P=0.04$ respectively). Non-FMD remained essentially unchanged regardless of the type of meal the subject consumed (Table 5). In Table 6, the significant increase in the diameter of brachial artery 2 hours after the wine containing meal 4 is presented. This phenomenon may account for the reduced endothelial response recorded in this case.

TABLE 2. Comparison of baseline values in the four different sessions

	meal A	meal B	meal C	meal D	p
Cholesterol (mg/dl)	178.4±31.7	186.2±30.5	182.2±33.6	179.2±34.2	0.554
Triglycerides (mg/dl)	84.6±44.3	83.8±43.2	98.1±55.7	95.8±50.2	0.198
HDL (mg/dl)	43±8.5	41.7±9.4	43.2±8	43.1±9.5	0.907
LDL (mg/dl)	117.3±24.9	119.8±30.5	114.2±37.9	120.8±38.4	0.345
Apo A 1(mg/dl)	134.5±17.5	138.5±22.6	136.9±15.2	136.5±19.8	0.477
Apo B 100 (mg/dl)	91.3±25	93.8±23.1	98.4±25	91.9±32.4	0.801
Lp(α) (mg/dl)	11.2±11.4	12.2±14.3	8.8±10.6	9.4±9.8	0.701
Baseline brachial artery diameter (mm)	3.77±0.53	3.90±0.57	3.84±0.46	3.74±0.62	0.339
Pre-prandial FMD (%)	16.5±5.4	13.2±3.9	16.2±8.3	14.3±9.5	0.253
Pre-prandial non-FMD (%)	15.1±7.9	13.5±5.1	14.3±5	14±3.4	0.211

TABLE 3. Pre- and post-prandial lipid levels in correlation with the four different types of meals.

	Meal A			Meal B		
mg/dl	before	2h	4h	before	2h	4h
CHOL	178.4±31.7	176±30.9	181.9±31.7	186.2±30.5	187.8±30	189.5±28.6
TG	84.6±44.3	130.3±72.5	**145.7±76.1***	83.8±43.2	121.6±58.7	**122.7±60.1***
HDL-C	43±8.5	41.5±7.3	40±9.3	41.7±9.4	42.9±8	41.3±9
LDL-C	117.3±24.9	107±22.4	**111±24.9***	119.8±30.5	113.3±28	117.2±28.9
Apo A1	134.5±17.5	133.7±16.6	130.1±8.6	138.5±22.6	133.8±20.6	134±16.1
Apo B100	91.3±25	88.7±27.6	91.3±26.5	93.8±23.1	92±22.6	93.7±21.1
Lp(a)	11.2±11.4	12.6±12.3	12.8±14.2	12.2±14.3	11.9±13.9	12.1±13.7
	Meal C			**Meal D**		
mg/dl	before	2h	4h	before	2h	4h
CHOL	182.2±33.6	181.5±32.1	182.5±30.4	179.2±34.2	174.8±31.6	176.5±30.3
TG	98.1±55.7	132.5±97	**130.1±80.8***	95.8±50.2	136.2±59.4	**177.9±75***
HDL-C	43.2±8	43.2±8	43.2±8	43.1±9.5	42.2±8.5	**39.9±8.5***
LDL-C	114.2±37.9	111.8±21.8	113.3±24.7	120.8±38.4	110.3±38.2	**102.6±32.8***
Apo A1	136.9±15.2	135.9±20.5	136.8±18.5	136.5±19.8	133.2±20.9	**130.2±20.1***
Apo B100	98.4±25	95.3±22.8	**100.1±25.6***	91.9±32.4	93.7±30	94.7±31.1
Lp(a)	8.8±10.6	9±10.7	9.3±11.4	9.4±9.8	8.6±9.9	**8.4±9.1***

The asterisk (*) indicates statistical significance. CHOL: Total cholesterol, TG: Triglycerides, HDL-C: HDL cholesterol, LDL-C: LDL cholesterol, Apo A1: Apolipoprotein A1, Apo B100: Apolipoprotein B 100, LP(a): lipoprotein (a)

TABLE 4. Endothelium mediated vasodilation before and after meals

	meal A	meal B	meal C	meal D
before	16.5±5,4 %	13.2±3.9 %	16.2±8.3 %	14.3±9.5 %
2h	13±4.6 %	14.8±7 %	15.5±6.9 %	8.6±5.5 %
4h	12.3±6 %	12.1±3.4 %	15.8±4.7 %	12.6±7.5 %
P	**0.030**	0.145	0.623	0.214

TABLE 5. Non-endothelium-mediated vasodilation before and after meals

	meal A	meal B	meal C	meal D
before	15.1±7.9 %	13.5±5.1 %	14.3±5 %	14±3.4 %
4 hour	14.2±5 %	16.5±6.2 %	13.3±4.5 %	12.3±6 %
p	0.352 %	0,683 %	0,373 %	0,278 %

TABLE 6. Baseline brachial artery diameter before and after meals

	meal A	meal B	meal C	meal D
Baseline brachial artery diameter (mm)	3.7±0.5	3.9 ±0.5	3.8±0.4	3.7±0.6
Brachial artery diameter in 2 hours (mm)	3.9±0.4	3.9±0.6	3.9±0.6	4.2±0.7
Brachial artery diameter in 4 hours (mm)	3.9±0.5	3.9±0.6	3.9±0.6	3.9±0.7
p	0.258	0.596	0.142	**0.000**

DISCUSSION

The main finding of the present study was that the ingestion of a saturated fat-rich meal acutely impaired endothelial vasodilatory function, while after the intake of either mono- or poly-unsaturated fat FDM remained unchanged. In addition, the administration of red wine with the saturated fat decreased FMD 2 hours after the meal. However, FMD completely recovered 4 hours postprandially. This last finding could be attributed to an acute vasodilatory effect caused by the red wine leading to a significant increase in the basal diameter of the brachial artery. It has been well established that FMD is inversely related to the baseline arterial diameter. In other words, bigger arteries dilate less compared to smaller ones.[11] Thus, the acute drop in FMD at 2 hours should not be perceived as evidence of ED. On the other hand, the complete

recovery of endothelial function at 4 hours could be attributed to a counteracting effect of red wine to the saturated fat-induced ED.

Conflicting results have been reported from others who have, similarly to us, investigated the acute effect of saturated fat intake on endothelial function. Vogel et al[12] found a significant decrease of FMD in 10 healthy volunteers 2 and 4 hours after the administration of a fatty meal. Marchesi et al[13] reported similar results in a healthy but younger population who developed ED 2 and 4 hours postprandially. In this study, FMD returned back to baseline levels 6 hours postprandially. On the other hand, Maggi et al[14] showed that in middle-aged volunteers FMD remains low even 6 hours after the fat intake, while Tsai et al[15] reported that even in younger healthy people FMD remains low at 6 hours postprandially. Others have reported a neutral effect of postprandial lipemia on the endothelium. Raitakari et al[6] did not find any significant FMD drop after the administration of a fatty meal in 12 healthy volunteers and they attributed their results to an increase of the mean brachial artery diameter postprandially, a finding confirmed by others[17] but not from our results. In addition, Djousse et al[18] who fed 12 healthy persons with a fat- rich meal reported no FMD changes at 2, 4 and 6 hours postprandially. Finally, Muntwyler et[19] al did not find any relationship between postprandial lipemia and endothelium dependent dilation as assessed by impedance plethysmography in 12 twelve healthy men. The reason for the above-mentioned discrepancies is not entirely clear. However, the different types of fat loading may had affected endothelial response since in some of the studies a pure fat emulsion was used while in others a regular meal was administered. Indeed, other components of an ordinary meal beyond fat may affect endothelial function. It has been shown, for example, that the addition of 50 g of protein counteracts the harmful effect of postprandial lipemia possibly by increasing the supply of L-arginin, a well known substrate for the production of endothelial NO.[20] On the other hand, oxidative modification of poly-unsaturated fat through deep frying may render the cooked fat even more toxic for the endothelium compared to the regular fat used in some of the studies mentioned above.[21]

The acute effect of mainly mono- or poly-unsaturated fatty acids containing meal on endothelial function has been less extensively studied. We found that this type of meals did not adversely affect postprandial endothelial function while in a similar study Vogel et al[22] who compared the acute effect of 3 types of fatty meal on endothelial function (olive oil, canola oil and fish oil) reported that the highly containing mono-unsaturated fat olive oil adversely affected endothelium while the rich in poly-unsaturated fat canola and fish oils did not affect FMD. A similar harmful effect on the endothelium was noticed by Ong et al[23] who administered a meal rich in oleic and linoleic acids (the main components of olive oil). However, Ros et al[24] who gave mon-unsaturated fat in 21 hypercholesterolemic adults for 4 weeks did not report any endothelial dysfunction in their population. In the same study, the substitution of mono-unsaturated with poly-unsaturated fat (in the form of nuts) was associated with a similar neutral effect on endothelial function. As suggested by Ruano et al,[25] other components of the olive oil such as its phenolic compounds which possess antioxidant, anti-inflammatory and antithrombotic properties, may account for its vascular protective effects. Thus, the vegetable fat may improve endothelial function only if contains sufficient quantities of these compounds. Beyond all the above discrepancies, the main body of evidence as derived from several large epidemiologic studies, clearly supports a vasoprotective role for mono- and poly-unsaturated fat in contrast to the atherogenic effect of saturated fat.[26,27] How much of this effect could be directly related to endothelial function remains to become fully elucidated.

The combined effect of red wine and fat has not been extensively studied. In the most relevant study Djousee et al[18] in contrast to us, did not find any effect of fat either alone or combined with red wine on the endothelial function of healthy volunteers. Other studies which examined the acute effect of wine alone, have reported conflicting results. Whelan et al[28] found that both red and white wine acutely increased FMD 6 hours after their intake while Karantzi et al[29] demonstrated, similarly to us, that red wine deteriorated endothelial function 60 minutes after its intake with a complete recovery 30 minutes later. In a subsequent study, the same research team reported that only the combination of red wine with a rich in antioxidants type of olive oil was able to improve FMD 2 hours postprandially.[30] It is unclear why the red wine had a variable effect on endothelial function in the studies mentioned above. Nevertheless, the baseline vasodilation which similarly to us was observed in most of these studies might have acted as a confounding factor obscuring the expected improvement of endothelial vasodilatory response and the blunting of fat-induced ED. Indeed, a very early (within 30 minutes) improvement of FMD has been demonstrated with the intake of alcohol-free red wine which notably does not induce any baseline arterial vasodilation.[31,32] The pathophysiologic mechanisms underlying the wine-induced vasorelaxation are not fully understood. The high content of red wine in polyphenolic compounds may partially explain its beneficial effect on endothelium since polyphenoles as potent antioxidants increase nitric oxide production and inhibit postprandial absorption of lipid peroxides.[33,34]

Postprandial hypertriglyceridemia is probably the most important mechanism associated with the blunted endothelial vasodilatory response following a fatty meal. Indeed, triglyceride and chylomicron levels have been related to the development and progression of atherosclerosis.[35,36] Moreover, in most[8,12] but not all of the studies[20,37] an inverse relationship between postprandial changes in FMD and plasma triglycerides has been reported. As expected, we found a significant

postprandial increase in plasma triglycerides. Postprandial hypertryceridemia was significantly higher after the ingestion of saturated fat (77±6.7% 4 hours after meal A and 106.7±24.1% 4 hours after meal D) compared to the one developed after unsaturated fat intake (65.1±20.1% 4 hours after meal B and 33.5±6.7% after meal C). This finding may offer an explanation about the saturated fat - induced postprandial ED. Nevertheless, since a general consensus about the relationship between triglyceride levels and FMD is lacking, not only the magnitude but also the pattern postprandial lipemia may need to be taken into account. It has been shown that various types of fat induce different patterns of postprandial lipemia with vegetable oils been associated with a less atherogenic one characterized by low levels of oxidized LDL cholesterol[38] and chylomicron remnants.[39]

Several *limitations* of our study need to be reported. This study was designed to address short-term effects of different fatty meals and of red wine on endothelial function. Long term effects of chronic ingestion of various types of fat or of red wine cannot be inferred from the present data. Since the population of our study was young with a "healthy" endothelium, an acute adverse effect of a fat load might have been difficult to demonstrate. Postprandial ED could have been easier to assess in a middle-aged population with several cardiovascular risk factors. Thus, extrapolation of our data to other populations would not be appropriate. We used common mixed meals instead of experimental diets. Other than the fat components of the meal could have modified the lipid effect of endothelial function. Nevertheless, our data represent more closely real life conditions. In this crossover trial each participant was examined 4 times 15-30 days apart. Although all subjects were instructed not to change their dietary habits in between examinations minor changes may had happened and the goal of a steady state during the whole study may had not been achieved for every one of them. Finally, since we did not measure any oxidative or inflammatory markers, we can not speculate on potential differences in anti-oxidant or anti-inflammatory properties of the various types of fat and of red wine as the underlying pathophysiologic mechanism of their diverse effect on endothelial function.

CONCLUSION

In conclusion, the consumption of a diet with a high saturated fat content acutely impairs FMD while diets containing mono- and poly-unsaturated fat have a neutral acute effect on endothelial function. The concomitant intake of red wine with saturated fat is associated with a delayed counteracting effect on lipid-induced postprandial ED. Low levels of hypertriglyceridemia after the consumption of mono- and poly-unsaturated fat diets may account for the beneficial effect of these types of fat on endothelial function in the postprandial state. Since endothelial dysfunction is an important step in the development of atherosclerosis, a diet containing mainly vegetable oil as fat source along with the intake of moderate quantities of red wine with meals may contribute to the prevention of future cardiovascular events.

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