Effects of a high-fat meal on inflammatory and endothelial injury biomarkers in accordance with adiposity status: a cross-sectional study

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Abstract

Background It is known that consuming a high-fat meal (HFM) induces microvascular dysfunction (MD) in eutrophic women and aggravates it in those with obesity. Our purpose was to investigate if the MD observed after a single HFM intake is caused by endothelial damage or increased inflammatory state, both determined by blood biomarkers.

Methods Nineteen women with obesity (BMI 30-34.9 kg/m^2) and 18 eutrophic ones (BMI 20.0-24.9 kg/m^2) were enrolled into two groups: Obese (OBG) and Control (CG), respectively. Blood samples were collected at five-time points: before (fasting state) and 30, 60, 120, and 180 min after HFM intake to determine levels of adipokines (adiponectin, leptin), non-esterified fatty acid (NEFA), inflammatory [tumor necrosis factor-α (TNF-α), interleukin-6 (IL-6)] and endothelium damage [soluble E-selectin, soluble vascular cell adhesion molecule-1 (sVCAM-1), soluble intercellular adhesion molecule-1 (sICAM-1), plasminogen activator inhibitor-1 (PAI-1)] biomarkers.

Results Levels of soluble E-selectin, leptin, and PAI-1 were higher in OBG at all-time points (P < 0.05) compared to CG. In the fasting state, OBG had higher levels of NEFA compared to CG (P < 0.05). In intra-group analysis, no significant change in the levels of circulating inflammatory and endothelial injury biomarkers was observed after HFM intake, independently of the group.

Conclusion Our findings suggest that women with obesity have an increased pro-inflammatory state and more significant endothelial injury compared to eutrophic ones. However, the consumption of a HFM was not sufficient to change circulating levels of inflammatory and endothelial injury biomarkers in either group.

Registration number for clinical trials: NCT01692327.

Keywords Obesity, High fat meal, Inflammation, Endothelium, Postprandial period

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Background

Obesity has reached epidemic proportions globally [1]. According to World Health Organization (WHO), in 2016, more than 1.9 billion adults were overweight. Of these, over 650 million had obesity [2], and at least 2.8 million die each year due to obesity [1]. It is well known that unhealthy habits, such as smoking, alcohol abuse and excessive energy drinks consumption, diets with an excess of salt, sugar, high saturated fat, and also discretionary foods, and physical inactivity play a central role in obesity epidemic [3]. All these habits predispose to insulin resistance and dyslipidemia, which increase the risk for obesity-related comorbidities, such as cardiovascular diseases (CVD), type 2 diabetes mellitus (T2DM), nonalcoholic fatty liver disease (NAFLD) and dementia [2, 4, 5]. Nonetheless, CVD increases morbimortality rates being of particular concern [6, 7], in special due to ischemic heart disease and stroke ranking first and second global causes of death, respectively [9].

In addition, recent data have shown that a meaningful hallmark of obesity is chronic low-grade systemic inflammation, an important contributor to atherosclerosis onset [10]. In obesity, visceral adipose tissue downregulates the expression of anti-inflammatory adipocytokines, such as adiponectin, that protects the vascular endothelium and upregulates the expression of several proinflammatory mediators, including C-reactive protein (CRP) and interleukin-6 (IL-6) [11]. Consequently, the endothelium becomes activated and assumes a dysfunctional phenotype [12, 13], expressed by increased leukocyte adhesion, platelet aggregation, hypercoagulation, vascular constriction, and decreased fibrinolysis [14]. Dysfunctional endothelium becomes more permeable and allows low-density lipoprotein (LDL) particles to infiltrate and accumulate in the extracellular matrix, where they undergo oxidation and enzymatic modifications [15, 16]. Modified LDLs exacerbate the proinflammatory process eliciting the secretion of several cytokines, including TNF-α and IL-6, which in turn induce the expression of intercellular adhesion molecule-1 (ICAM-1) and vascular cell adhesion molecule-1 (VCAM-1) on endothelial surface and the release of monocyte chemoattractant protein (MCP-1) among other chemokines, consequently promoting the adherence and migration of monocytes and other immune cells. Once monocytes transmigrate and reach the subendothelial space, they differentiate into macrophages and express high levels of scavenger receptors, allowing them to internalize those modified LDLs [16]. Macrophages then become lipid-laden and convert into foam cells, which is a feature of early fatty streak lesions [17]. In parallel with the first stage of the disease, vascular smooth muscle cells (VSMCs) migrate into the intima, where they deposit extracellular matrix and promote the development of the fibrous cap [15].

As the atherosclerotic plaque progresses, the number of VSMCs decreases and foam cells become apoptotic, releasing metalloproteinases that degrade the fibrous cap, increasing the susceptibility of plaque rupture [18]. The innate and acquired immune responses contribute to the entire atherosclerotic process and are closely related to plaque vulnerability [15]. Thus, endothelial dysfunction (ED) occurs prior to the development of atherosclerotic plaques [14–16] and plays a pivotal role in the progression of CVD [17]. In 1979, Zilversmit proposed that atherosclerosis is a postprandial phenomenon [23] which attracted increased attention to postprandial lipid (PPL) metabolism. High-fat diet alters crucial protective functions of the endothelium, making its phenotype dysfunctional and predisposing it to the development of atherosclerosis [19, 20]. The most striking aspect of ED is the decreased nitric oxide (NO) bioavailability resulting in an impairment of endothelium-dependent vasodilation and loss of its anti-coagulant, anti-adhesive and anti-proliferative properties. The reduced NO bioavailability comes either from reduced production by the endothelium or its enhanced depletion by reactive oxygen species [21, 22].

Several studies have shown the postprandial deleterious effects on vascular function [23, 24]. In addition, our group have demonstrated that a single high-fat meal (HFM) elicited microvascular dysfunction (MD) in eutrophic women and worsened it in those with obesity [25]. However, the underlying mechanisms that induced or aggravated MD after lipid overload remain uncertain but may involve endothelial injury and increased inflammatory status. A study by Nappo et al. 2002 [30] conducted in healthy subjects and in patients with T2DM who ingested a high-fat meal has shown a significant increase in the circulating levels of proinflammatory cytokines, like tumor necrosis factor-α (TNF-α) and IL-6 and of soluble ICAM-1 and VCAM-1, both known as endothelial injury biomarkers. Additionally, Van Oostrom et al. 2003 [31] also studying the effects of high-fat meals demonstrated that they elicited a significant elevation in the number of circulating leukocytes with a concomitant reversible decline of endothelial function.

Based on these findings, this study aimed to investigate if the recorded MD observed after a single HFM intake is caused by endothelial damage or increased inflammatory state both tested by blood biomarkers.

Methods

Study Design

This is a cross-sectional study approved by the Research Ethical Committee at Hospital Universitário Pedro Ernesto (CAAE: 0190.0.228.000–10). All participants have read and signed the written informed consent form before being included. This study was registered on the
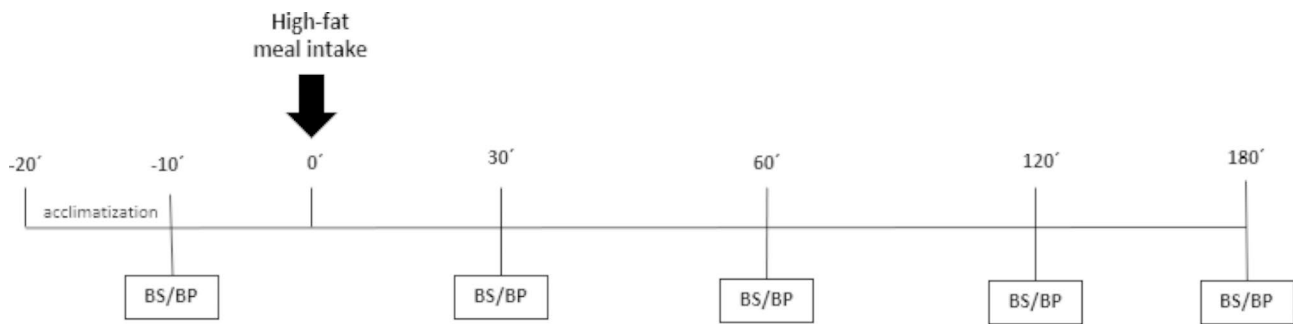
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Fig. 1 Experimental Design. (BS – blood sample collection; BP – blood pressure)

Clinical Trials website under "Study about the HFM and postprandial lipemia" and assigned the identifier NCT01692327.

Study Population
Patients were recruited in the Obesity Unit for outpatients' care at the State University of Rio de Janeiro, Brazil. Before inclusion, all participants attended an assessment appointment and underwent clinical, anthropometric, and body composition evaluations and blood collection for biochemical analysis and hemogram.

Eligibility criteria
The main inclusion criteria were women between 19 and 40 years with body mass index (BMI) between 30 and 34.9 kg/m² (Obesity group - OBG) or between 20 and 24.9 kg/m² (Control group - CG). The exclusion criteria were male sex, hypothyroidism, metabolic syndrome, any degree of glucose intolerance or diabetes mellitus; arterial hypertension; symptoms or history of lactose intolerance; weight gain or reduction by at least 5% of body weight in the past six months before recruitment; tobacco use and alcohol abuse; being involved in a regular practice of physical activity [32] and/or under simultaneous nutritional or pharmacological interventions.

Blood biochemical analysis
The biochemical analysis encompassed the circulating levels of insulin, total cholesterol (TC), triglycerides (TG), high-density lipoprotein cholesterol (HDL-c), and thyroid-stimulating hormone (TSH) after a 12-hour overnight fast. Fasting and 2-hour post-load (75 g, oral) plasma glucose were also evaluated. Glucose intolerance and metabolic syndrome were defined according to the American Diabetes Association (ADA) criteria[26] and the Joint Interim Statement[34], respectively. Plasma levels of glucose were measured by glucose oxidase colorimetric method. Blood was also collected into serum tubes for insulin and lipid profile analysis. Serum levels of TG, TC and HDL-c were assessed by glycerol phosphate oxidase/peroxidase, cholesterol oxidase/peroxidase and direct colorimetric methods, respectively. All

analyses were performed using commercially available kits appropriate for the Automatic Analyser A25 (BioSystems, Barcelona, Spain), following the protocols provided by the kit's manufacturer (BioSystems, Barcelona, Spain). LDL-c was calculated by the Friedewald equation [26]. Intra and inter-assay coefficient of variation of all analyses described above were below 15% and previously validated [27].

The test day
We included nineteen women in the OBG and eighteen eutrophics who met the eligibility criteria. On the test day, they were asked to arrive after a 12-hour fast at 7:00 a.m. and were placed to rest in an acclimatized room for 20-min. Systolic (SBP) and diastolic (DBP) blood pressures were subsequently assessed by auscultatory method, and then an intravenous catheter was placed and kept in place for blood sample collections during the test. Time points for blood pressure monitoring and blood samples harvesting were: at the fasting state (baseline) and 30, 60, 120, and 180-min after HFM intake (Fig. 1). The same meal was provided to both groups. Participants were instructed not to ingest fat-rich meals, drink caffeine or energy beverages, or practice any physical activity within 24 h before the test.

Test meal (high-fat meal)
Participants ingested a single high-fat breakfast consisting of whole milk (200ml), chocolate (10 g), margarine (20 g), croissant (1 unit), cheddar cheese (60 g), and salami (31 g). The meal was similar to the one used by Signori and coworkers[28], with minor changes to incorporate more palatable foods. The meal had 691.5 kcal and consisted of 24.8% carbohydrates, 59.5% lipids (including 21.9 g of saturated fat), and 15.7% proteins. The time limit for its consumption was 10 min.

Analysis of inflammatory and endothelial injury biomarkers
Blood samples were collected into EDTA tubes to determine levels of inflammatory [IL-6, tumor necrosis factor-α (TNF-α) and adiponectin] and endothelial injury biomarkers [sE-selectin, plasminogen activator

Table 1 Clinical, laboratory and body composition characteristics of the control group (CG) and group with obesity (OBG).

Variables	CG	OBG
Age (years)	27.89(5.38)	30.74(4.48)
Anthropometric measures		
Weight (kg)	57.82(6.24)	86.28(7.84)***
Height (cm)	1.63(0.07)	1.63(0.05)
BMI (kg/m^2)	21.81(1.79)	32.31(1.54)***
WC (cm)	79.47(5.09)	106.4(9.37)***
HC (cm)	100.3(4.47)	117.5(6.5)***
WHR	0.79(0.03)	0.91(0.85)***
Blood Pressure and heart rate		
SBP (mmHg)	109.1(7.53)	115.8(13.82)
DBP (mmHg)	69.09(6.12)	76.74(9.14)**
Heart rate (bpm)	73.19(9.56)	72.84(6.4)
Body Composition		
Fat mass (%)	27.88(3.11)	38.12(1.78)***
Muscle mass (%)	72.14(3.08)	61.81(1.75)***
Laboratorial analysis		
Glucose (mg/dl)	81.06(6.05)	88.79(13.06)*
Insulin (mU/l)	8.18(4.25)	12.74(4.76)**
TC (mg/dl)	169.40(32.18)	196.50(29.24)*
LDL-c (mg/dl)	86.70(24.92)	110.4(27.81)**
TG (mg/dl)	73.72(37.84)	109.50(56.42)*
HDL-c (mg/dl)	67.94(17.65)	62.37(16.57)

Data are expressed as mean (SD). BMI - body mass index; WC – waist circumference; HP -Hip circumference; WHR - Waist-to-hip ratio; SBP - systolic blood pressure; DBP - diastolic blood pressure; TC- total cholesterol; LDL-c - Low-density lipoprotein cholesterol; TG- triglycerides; HDL-c - High-density lipoprotein cholesterol; *Significantly different compared to CG group; *P<0.05; **P<0.01; ***P<0.001.

inhibitor-1 (PAI-1), soluble vascular cell adhesion molecule-1 (sVCAM-1) and soluble intercellular adhesion molecule-1 (sICAM-1)] at all-time points. Samples were centrifuged over 10 min at 4°C at 1000 g. Plasma was then transferred into cryotubes and stored at -80°C until analysis. Plasma levels of sE-selectin, PAI-1, sVCAM-1 and sICAM-1 were assessed by non-magnetic Human Cardiovascular Disease Panel 1 (EMD Millipore Corporation, MA, USA); sensitivity>0.079 ng/ml. IL-6 was quantified by Quantikine® HS Human IL-6 Immunoassay kit (R&D Systems, MN, USA); sensitivity>0.039 pg/ml. TNF-α was assayed by High Sensitivity Human Cytokine Magnetic Bead Panel (EMD Millipore Corporation MA, USA); sensitivity>0.07 pg/ml. Adiponectin was evaluated by Human Adipokine Panel 1 (EMD Millipore Corporation MA, USA); sensitivity>11 pg/ml. Plasma levels of leptin were determined by Human Metabolic Hormone Magnetic Bead Panel (EMD Millipore Corporation MA, USA); sensitivity>27 pg/ml. Blood was also collected in serum tubes for non-esterified fatty acids (NEFA) analysis. Serum tubes were centrifuged for 10 min at 18°C at 3000 rpm. Serum was then transferred into cryotubes and stored at -80°C until analysis using commercially

available reagents (Wako Chemicals, VA, USA). The between and within assay coefficient of variation of all analyses was less than 15%.

Statistical analysis

Statistical analysis was performed by GraphPad® Prism software, version 5. Normal Gaussian distribution was assessed using the Shapiro-Wilk normality test. For Gaussian and non-Gaussian distributions, data were expressed respectively by mean±SD or median [1st -3rd quartiles]. Intra-group comparisons were performed by ANOVA repeated measures or the Friedman test. Comparisons between groups were tested by unpaired t-test or Mann-Whitney U test. GPower 3.1.10 software was used for power analysis and sample size estimation. Significant differences were assumed to be present at P<0.05.

Results

Baseline data on body composition, clinical and laboratory evaluation of the participants

Table 1 depicts clinical, laboratory, and body mass characteristics of study participants during the assessment visit. As expected, OBG had significantly greater weight (P<0.001), BMI (P<0.001), waist and hip circumferences (P<0.001), waist-to-hip ratio (WHR, P<0.001), DBP (P<0.01), and percent of fat mass (P<0.001) compared to CG. Additionally, OBG had significantly higher levels of glucose (P<0.05), insulin (P<0.01), TC (P<0.05), LDL-c (P<0.01), and TG (P<0.05) than CG. On the other hand, OBG showed significantly lower muscle mass than CG (P<0.001). There were no significant differences between groups concerning age (P=0.083), height (P=0.73), SBP (P=0.073), heart rate (P=0.90) and HDL-c (P=0.33).

Differences in inflammatory and endothelial injury biomarkers between groups

Regarding adipocytokines (Fig. 2), we did not notice any differences in TNF-α levels between groups in the fasting state. However, at this state, levels of leptin, IL-6 and NEFA were higher in OBG than CG, while adiponectin was lower.

Concerning endothelial injury biomarkers, the levels of sICAM-1 and sVCAM-1 were similar between OBG and CG at the fasting state. However, levels of sE-selectin and PAI-1 were higher in OBG than in CG and remained unchanged during the postprandial state (P<0.05). However, no significant differences were observed in the levels of sICAM-1 and sVCAM-1 during this period (Fig. 3). Regarding adipocytokines, we noticed that leptin and IL-6 levels were significantly higher in OBG than CG during the postprandial period (P<0.05). In contrast, adiponectin levels were lower (P<0.05). Furthermore, no
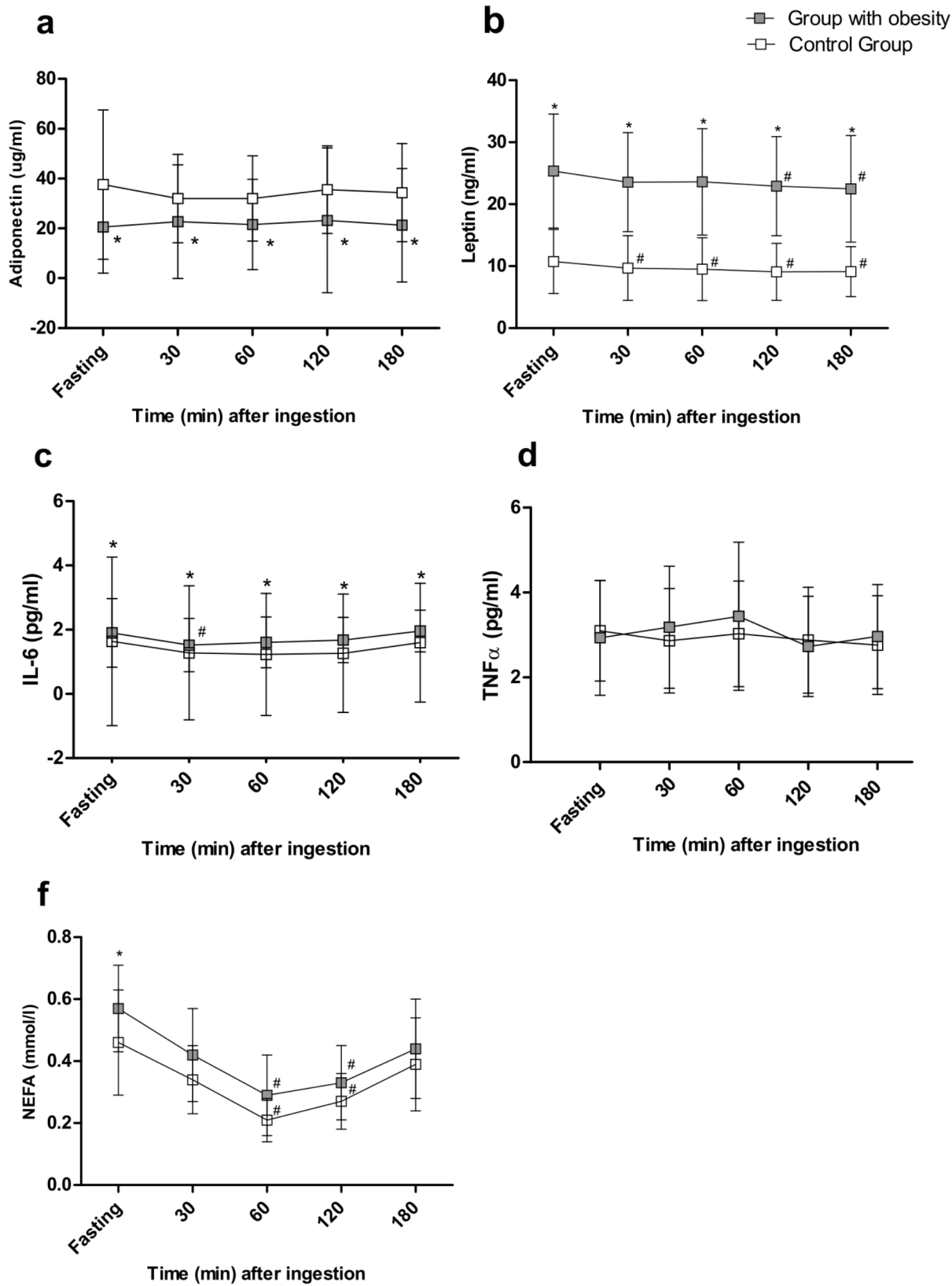
Fig. 2 - Adipokines responses elicited by high-fat meal in control (CG) and obese (OBG) groups. (* P<0.05 intergroup; # P<0.05 intragroup (compared to baseline))
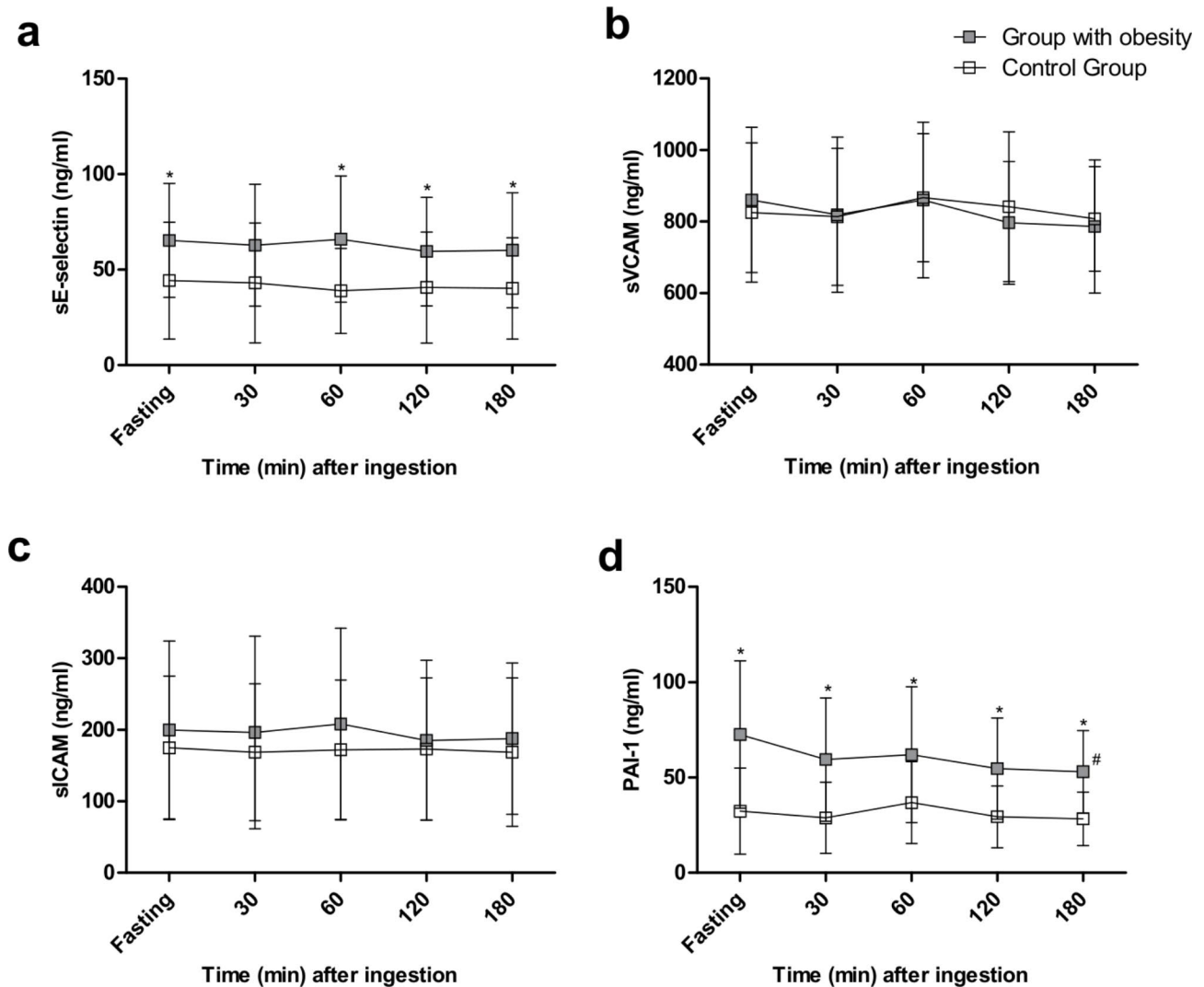
Fig. 3 - Endothelial biomarkers responses induced by high-fat meal in control (CG) and obese (OBG) groups. (* P < 0.05 intergroup; # P < 0.05 intragroup (compared to baseline))

significant differences in the levels of TNF-α and NEFA were observed between groups after HFM intake (Fig. 2).

Differences in inflammatory and endothelial damage biomarkers within groups

No significant differences within groups were noticed regarding endothelial injury biomarkers sE-selectin, s-ICAM-1, and s-VCAM-1 after an HFM intake. However, in OBG, it was observed a significant decrease in PAI-1 levels at 180-min compared to baseline (P < 0.05) (Fig. 3).

Furthermore, NEFA levels at 60 and 120-min after HFM were significantly lower in both groups in comparison to baseline (P < 0.05). The same was observed with respect to leptin levels, that was significantly reduced in both groups after a high-fat meal (OBG at 120-min and 180-min) and (CG at four-time points after HFM

ingestion) compared to baseline levels. In addition, we noticed that IL-6 concentration was significantly reduced in OBG (30-min) after meal intake. Nonetheless, no significant differences were observed in adiponectin and TNF-α levels (Fig. 2).

Discussion

The main finding of this study is that an intake of a single HFM was insufficient to elicit significant changes in inflammatory and endothelial injury biomarkers, independently of BMI. We also observed that women with obesity presented an increased proinflammatory state and endothelium damage before ingesting HFM and sustained it until 180-min after its intake, differently from eutrophic ones. In fact, in the present study, we enrolled participants with obesity who were age and gender-matched to CG. According to exclusion criteria, they

were all young, and they did not have T2DM, hypertension or metabolic syndrome, implying that obesity *per se* was responsible for the induction of the proinflammatory state and endothelial injury and not the ingestion of an HFM.

We have reported previously that obesity *per se* elicited MD in women in a fasting state [29]. More recently, we decided to investigate the microvascular function in young women with obesity during the postprandial period [19], considering atherogenesis a postprandial phenomenon. Our findings showed that the metabolic status reflected by changes in glucose, insulin, total cholesterol, HDL, LDL and triglycerides levels after the HFM aggravated MD, which was already present in women with obesity at fasting state and induced the deterioration of microvascular function in eutrophic ones [25]. Nonetheless, the underlying mechanisms responsible for MD after lipid overload remain unclear. We hypothesized that endothelial injury and inflammatory state were probably the main causes of microvascular damage evoked by HFM. In order to answer this question, we examined levels of inflammatory and endothelial injury biomarkers before and after a HFM intake. Adipocytokines, such as leptin and adiponectin, are an essential link between adipose tissue and vasculature, comprising the adipovascular axis, and alterations in their circulating levels will significantly influence vascular function [30]. In addition, we included some inflammatory cytokines, like TNF-α and IL-6, also some endothelial injury biomarkers (such as sE-selectin, sICAM-1, sVCAM-1, and PAI-1), and NEFA because their levels increase in response to subclinical inflammation, playing a significant role in the pathogenesis of the atherosclerotic process [31].

In the present study, we did not observe significant inflammatory and endothelial injury biomarkers changes within groups before and after HFM intake. In contrast, a meta-analysis performed by Tom et al. (2016) revealed that meal consumption reduces flow-mediated dilatation (FMD), evidencing the impairment of endothelial function [39]. The probable explanation for the observed endothelial injury was related to elevation of triglyceride-rich lipids (TRLs) levels after a high-fat consumption, which in turn increased oxidative stress. Oxidative stress is responsible for decreased NO bioavailability reflected by suppressed FMD response, the main feature of ED [40]. Furthermore, humans consume enormous amounts of food generally with high-fat content[34, 35], and consecutive intake of HFM appears to exacerbate lipemia effects[36]. All these factors may contribute to the appearance of atherosclerotic lesions, an independent risk factor for CVD [40].

Interleukin-6 and TNF-α are both critical mediators of inflammation, with increased levels in obesity. Diets may modulate the postprandial response of these two mediators, and we expected that HFM could have a more significant impact on postprandial inflammatory response, particularly in women with obesity. Instead, ingested diet did not change TNF-α levels, while IL-6 levels were reduced at 30-min after HFM. A study by Meyer et al. (2014) corroborates our findings, revealing that after a mixed meal, no significant changes were detected in T2DM and normoglycemic groups 2 and 4-hours after the intake [37]. On the other hand, other studies have reported increased levels of IL-6 in subjects with obesity after HFM ingestion. However, the meals investigated had higher quantities of saturated fat, and levels of circulating IL-6 were assessed for 6 h [19, 38].

Our study also showed that circulating levels of sICAM-1, sVCAM-1, and sE-selectin remained unchanged during the postprandial state in both groups. Corroborating our data, Rubin et al. (2008) did not find any changes in these biomarkers following a lipid-rich test meal. However, these authors pointed out an independent association of postprandial levels of triglycerides with sICAM-1, which could indicate a specific impact of postprandial lipid metabolism on endothelial response [46]. Peairs et al. (2011) claimed that the type of fat in meals could affect postprandial inflammation and endothelial activation differently. In their findings, sICAM-1 levels increased following acute saturated fat ingestion [40]. However, another study found that a meal rich in olive oil elicited a lower postprandial sICAM-1 level than a meal with a higher amount of saturated fat [48].

With regard to adiponectin and leptin, several studies have presented divergent findings [42, 43]. In our study, adiponectin levels in both groups were unaffected by the ingested meal, while leptin levels were reduced in the postprandial state in OBG (from 120-min to 180-min) and CG (from 30-min to 180-min). A recent study found that a fatty meal induces postprandial changes in adiponectin and leptin secretions in normal-weight subjects but not in individuals with obesity, implying that postprandial regulating role of adiponectin and leptin is reduced in obesity [44]. Furthermore, we may speculate that these disparities in postprandial adiponectin and leptin responses are influenced not only by participants' body weight but also by the meal's macronutrient composition[45].

Concerning NEFA and PAI-1, no significant changes in their levels following HFM was noticed. However, this is not a consensus since several studies have reported contradicting results. Nonetheless, these studies assessed different populations (e.g. with and without comorbidities), different diets and divergent observation periods following the meals intake [46–50].

Some limitations of our study warrant mention. Ideally, the observation period should be extended from 240 to 360-minutes [51]; however, sequential blood sampling

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for more than 4 h is very difficult to be performed due to the discomfort inflicted on the participants. Additionally, the literature is controversial with respect to the HFM stimulus. In the present study, it was probably insufficient to cause significant endothelial damage and inflammatory response. We could probably have found different results by adding more saturated fat and testing them for at least three hours or even after three meals over a day. We should have included a regular meal (control meal) to clarify the impact of HFM on inflammatory and endothelial injury biomarkers. We assessed postprandial lipemia only in women. However, Orem et al. (2018) determined postprandial TG ranges in healthy subjects by considering gender differences and noticed that postprandial TG cut-off values for female and male subjects should be determined separately; in other words, postprandial lipemia may display considerable gender differences, and all of them should be studied[52]. Our tests did not consider the women's menstrual cycle phase. The literature regarding the influence of the menstrual cycle on inflammatory and endothelial biomarkers is controversial. It depends on what biomarker is being tested, adiposity status, the age of the participant, and also on the sample size of the study. Since many disparities occur regarding this issue [60–65], we opted not to fix an exact menstrual cycle phase for the tests, even though this must be viewed as one of our limitations.

Conclusion

Our findings suggest that women with obesity have an increased proinflammatory state and more significant endothelial damage than eutrophic ones. However, the consumption of a single HFM was insufficient to change the levels of circulating inflammatory and endothelial injury biomarkers. Other factors, like blood viscosity, may be involved in HFM induced microvascular dysfunction. Therefore, more studies are needed to elucidate the underlying mechanisms that induce or aggravate microvascular dysfunction after a single HFM.

Abbreviations

ADA	American Diabetes Association
BMI	Body mass index
BP	Blood pressure
BS	Blood sample
CG	Control group
DBP	Diastolic blood pressure
ED	Endothelial dysfunction
HDL	High-density lipoprotein
HFM	High-fat meal
NEFA	Non-esterified free fatty acids
OBG	Obese group
PPL	Postprandial lipemia
SBP	Systolic blood pressure
sICAM	soluble intercellular adhesion molecule-1
sVCAM-1	soluble Vascular cell adhesion molecule −1
T2DM	Diabetes Mellitus type 2
TRLs	Triglycerides-rich lipoproteins
TSH	Thyroid-stimulating hormone
WHO	World Health Organization

Author's contributions
MGCS, PAM, EB, LGK-A: study design, conceived the experiments and writing of the manuscript. MGCS and LGK-A: data analysis and interpretation.MGCS and PAM: literature search and writing of the manuscript. MGCS, PAM, DGP, JFNN: data collection.

Data Availability
Data will be presented upon forwarding the request to the corresponding author (luiz.aguiar@hupe.uerj.br).

Declarations

Ethics approval and consent to participate
We obtained written informed consent from all participants. Based on the ethical guidelines of the 1975 Declaration of Helsinki, the study protocol was approved by the Ethics Research Council of the Pedro Ernesto University Hospital - State University of Rio de Janeiro.

Consent for publication
Not applicable.

Competing interests
The authors have no conflict of interest.

References
1. Obesity. https://www.who.int/news-room/facts-in-pictures/detail/6-facts-on-obesity.
2. Obesity. **and overweight**https://www.who.int/news-room/fact-sheets/detail/obesity-and-overweight.
3. Chatterjee A, Gerdes MW, Martinez SG. **Identification of Risk Factors Associated with Obesity and Overweight-A Machine Learning Overview**. Sensors (Basel) 2020, 20.
4. Afshin A, Forouzanfar MH, Reitsma MB, Sur P, Estep K, Lee A, Marczak L, Mokdad AH, Moradi-Lakeh M, Naghavi M, et al. Health Effects of Overweight and Obesity in 195 Countries over 25 Years. N Engl J Med. 2017;377:13–27.
5. Blüher M. Obesity: global epidemiology and pathogenesis. Nat Rev Endocrinol. 2019;15:288–98.
6. Ormazabal V, Nair S, Elfeky O, Aguayo C, Salomon C, Zuñiga FA. Association between insulin resistance and the development of cardiovascular disease. Cardiovasc Diabetol. 2018;17:122.
7. Dwivedi AK, Dubey P, Cistola DP, Reddy SY. Association Between Obesity and Cardiovascular Outcomes: Updated Evidence from Meta-analysis Studies. Curr Cardiol Rep. 2020;22:25.
8. Roth GA, Mensah GA, Johnson CO, Addolorato G, Ammirati E, Baddour LM, Barengo NC, Beaton AZ, Benjamin EJ, Benziger CP, et al. Global Burden of Cardiovascular Diseases and Risk Factors, 1990–2019: Update From the GBD 2019 Study. J Am Coll Cardiol. 2020;76:2982–3021.
9. **The top 10 causes of death**https://www.who.int/news-room/fact-sheets/detail/the-top-10-causes-of-death.
10. Libby P. The changing landscape of atherosclerosis. Nature. 2021;592:524–33.
11. Wang CC, Goalstone ML, Draznin B. Molecular mechanisms of insulin resistance that impact cardiovascular biology. Diabetes. 2004;53:2735–40.

12. Takaoka M, Nagata D, Kihara S, Shimomura I, Kimura Y, Tabata Y, Saito Y, Nagai R, Sata M. Periadventitial adipose tissue plays a critical role in vascular remodeling. Circ Res. 2009;105:906–11.

13. Ouchi N, Parker JL, Lugus JJ, Walsh K. Adipokines in inflammation and metabolic disease. Nat Rev Immunol. 2011;11:85–97.

14. Dzobo KE, Hanford KML, Kroon J. Vascular Metabolism as Driver of Atherosclerosis: Linking Endothelial Metabolism to Inflammation. Immunometabolism. 2021;3:e210020.

15. Badimon L, Vilahur G. Thrombosis formation on atherosclerotic lesions and plaque rupture. J Intern Med. 2014;276:618–32.

16. Wang D, Wang Z, Zhang L, Wang Y. Roles of Cells from the Arterial Vessel Wall in Atherosclerosis. Mediators Inflamm. 2017;2017:8135934.

17. Gimbrone MA Jr, García-Cardeña G. Endothelial Cell Dysfunction and the Pathobiology of Atherosclerosis. Circ Res. 2016;118:620–36.

18. Koga J, Aikawa M. Crosstalk between macrophages and smooth muscle cells in atherosclerotic vascular diseases. Vascul Pharmacol. 2012;57:24–8.

19. Davignon J, Ganz P. Role of endothelial dysfunction in atherosclerosis. Circulation. 2004;109:Iii27–32.

20. Ross R. The pathogenesis of atherosclerosis: a perspective for the 1990s. Nature. 1993;362:801–9.

21. Mudau M, Genis A, Lochner A, Strijdom H. Endothelial dysfunction: the early predictor of atherosclerosis. Cardiovasc J Afr. 2012;23:222–31.

22. Silveira A, Carlo A, Adam M, McLeod O, Lundman P, Boquist S, Woodhams BJ, Hamsten A. VIIaAT complexes, procoagulant phospholipids, and thrombin generation during postprandial lipemia. Int J Lab Hematol. 2018;40:251–7.

23. Zilversmit DB. Atherogenesis: a postprandial phenomenon. Circulation. 1979;60:473–85.

24. . Zhao Y, Liu L, Yang S, Liu G, Pan L, Gu C, Wang Y, Li D, Zhao R, Wu M. Mechanisms of Atherosclerosis Induced by Postprandial Lipemia. Front Cardiovasc Med. 2021;8:636947.

25. Incalza MA, D'Oria R, Natalicchio A, Perrini S, Laviola L, Giorgino F. Oxidative stress and reactive oxygen species in endothelial dysfunction associated with cardiovascular and metabolic diseases. Vascul Pharmacol. 2018;100:1–19.

26. Shaito A, Aramouni K, Assaf R, Parenti A, Orekhov A, Yazbi AE, Pintus G, Eid AH. Oxidative Stress-Induced Endothelial Dysfunction in Cardiovascular Diseases. Front Biosci (Landmark Ed). 2022;27:105.

27. Shafieesabet A, Scherbakov N, Ebner N, Sandek A, Lokau S, von Haehling S, Anker SD, Lainscak M, Laufs U, Doehner W. Acute effects of oral triglyceride load on dynamic changes in peripheral endothelial function in heart failure patients with reduced ejection fraction and healthy controls. Nutr Metab Cardiovasc Dis. 2020;30:1961–6.

28. Whisner CM, Angadi SS, Weltman NY, Weltman A, Rodriguez J, Patrie JT, Gaesser GA. **Effects of Low-Fat and High-Fat Meals, with and without Dietary Fiber, on Postprandial Endothelial Function, Triglyceridemia, and Glycemia in Adolescents**. Nutrients 2019, 11.

29. Maranhão PA, de Souza M, Panazzolo DG, Nogueira Neto JF, Bouskela E, Kraemer-Aguiar LG. Metabolic Changes Induced by High-Fat Meal Evoke Different Microvascular Responses in Accordance with Adiposity Status. Biomed Res Int. 2018;2018:5046508.

30. Nappo F, Esposito K, Cioffi M, Giugliano G, Molinari AM, Paolisso G, Marfella R, Giugliano D. Postprandial endothelial activation in healthy subjects and in type 2 diabetic patients: role of fat and carbohydrate meals. J Am Coll Cardiol. 2002;39:1145–50.

31. van Oostrom AJ, Sijmonsma TP, Verseyden C, Jansen EH, de Koning EJ, Rabelink TJ, Castro Cabezas M. Postprandial recruitment of neutrophils may contribute to endothelial dysfunction. J Lipid Res. 2003;44:576–83.

32. WHO guidelines. on physical activity and sedentary behaviour [https://www.who.int/publications/i/item/9789240015128].

33. Alberti KG, Eckel RH, Grundy SM, Zimmet PZ, Cleeman JI, Donato KA, Fruchart JC, James WP, Loria CM, Smith SC Jr. Harmonizing the metabolic syndrome: a joint interim statement of the International Diabetes Federation Task Force on Epidemiology and Prevention; National Heart, Lung, and Blood Institute; American Heart Association; World Heart Federation; International Atherosclerosis Society; and International Association for the Study of Obesity. Circulation. 2009;120:1640–5.

34. Friedewald WT, Levy RI, Fredrickson DS. Estimation of the concentration of low-density lipoprotein cholesterol in plasma, without use of the preparative ultracentrifuge. Clin Chem. 1972;18:499–502.

35. Signori LU, Vargas da Silva AM, Della Méa Plentz R, Geloneze B, Moreno H Jr, Belló-Klein A, Irigoyen MC. D'Agord Schaan B: **Reduced venous endothelial responsiveness after oral lipid overload in healthy volunteers**. Metabolism. 2008;57:103–9.

36. Maranhão PA, de Souza M, Kraemer-Aguiar LG, Bouskela E. Dynamic nailfold videocapillaroscopy may be used for early detection of microvascular dysfunction in obesity. Microvasc Res. 2016;106:31–5.

37. Adya R, Tan BK, Randeva HS. Differential effects of leptin and adiponectin in endothelial angiogenesis. J Diabetes Res. 2015;2015:648239.

38. Bošanská L, Michalský D, Lacinová Z, Dostálová I, Bártlová M, Haluzíková D, Matoulek M, Kasalický M, Haluzík M. The influence of obesity and different fat depots on adipose tissue gene expression and protein levels of cell adhesion molecules. Physiol Res. 2010;59:79–88.

39. Thom NJ, Early AR, Hunt BE, Harris RA, Herring MP. Eating and arterial endothelial function: a meta-analysis of the acute effects of meal consumption on flow-mediated dilation. Obes Rev. 2016;17:1080–90.

40. Moncada S, Higgs A. The L-arginine-nitric oxide pathway. N Engl J Med. 1993;329:2002–12.

41. Cohen JC, Noakes TD, Benade AJ. Serum triglyceride responses to fatty meals: effects of meal fat content. Am J Clin Nutr. 1988;47:825–7.

42. Dubois C, Beaumier G, Juhel C, Armand M, Portugal H, Pauli AM, Borel P, Latgé C, Lairon D. Effects of graded amounts (0–50 g) of dietary fat on postprandial lipemia and lipoproteins in normolipidemic adults. Am J Clin Nutr. 1998;67:31–8.

43. Jackson KG, Robertson MD, Fielding BA, Frayn KN, Williams CM. Measurement of apolipoprotein B-48 in the Svedberg flotation rate (S(f)) > 400, S(f) 60–400 and S(f) 20–60 lipoprotein fractions reveals novel findings with respect to the effects of dietary fatty acids on triacylglycerol-rich lipoproteins in postmenopausal women. Clin Sci (Lond). 2002;103:227–37.

44. Meher D, Dutta D, Ghosh S, Mukhopadhyay P, Chowdhury S, Mukhopadhyay S. Effect of a mixed meal on plasma lipids, insulin resistance and systemic inflammation in non-obese Indian adults with normal glucose tolerance and treatment naïve type-2 diabetes. Diabetes Res Clin Pract. 2014;104:97–102.

45. Barrett EJ, Wang H, Upchurch CT, Liu Z. Insulin regulates its own delivery to skeletal muscle by feed-forward actions on the vasculature. Am J Physiol Endocrinol Metab. 2011;301:E252–63.

46. Rubin D, Claas S, Pfeuffer M, Nothnagel M, Foelsch UR, Schrezenmeir J. s-ICAM-1 and s-VCAM-1 in healthy men are strongly associated with traits of the metabolic syndrome, becoming evident in the postprandial response to a lipid-rich meal. Lipids Health Dis. 2008;7:32.

47. Peairs AD, Rankin JW, Lee YW. Effects of acute ingestion of different fats on oxidative stress and inflammation in overweight and obese adults. Nutr J. 2011;10:122.

48. Pacheco YM, López S, Bermúdez B, Abia R, Villar J, Muriana FJ. A meal rich in oleic acid beneficially modulates postprandial sICAM-1 and sVCAM-1 in normotensive and hypertensive hypertriglyceridemic subjects. J Nutr Biochem. 2008;19:200–5.

49. Kennedy A, Spiers JP, Crowley V, Williams E, Lithander FE. Postprandial adiponectin and gelatinase response to a high-fat versus an isoenergetic low-fat meal in lean, healthy men. Nutrition. 2015;31:863–70.

50. Ferreira TDS, Antunes VP, Leal PM, Sanjuliani AF, Klein M. The influence of dietary and supplemental calcium on postprandial effects of a high-fat meal on lipaemia, glycaemia, C-reactive protein and adiponectin in obese women. Br J Nutr. 2017;118:607–15.

51. Larsen MA, Isaksen VT, Paulssen EJ, Goll R, Florholmen JR. Postprandial leptin and adiponectin in response to sugar and fat in obese and normal weight individuals. Endocrine. 2019;66:517–25.

52. Adamska-Patruno E, Ostrowska L, Goscik J, Fiedorczuk J, Moroz M, Kretowski A, Gorska M: **The Differences in Postprandial Serum Concentrations of Peptides That Regulate Satiety/Hunger and Metabolism after Various Meal Intake, in Men with Normal vs. Excessive BMI**. Nutrients 2019, **11**.

53. Mager DR, Mazurak V, Rodriguez-Dimitrescu C, Vine D, Jetha M, Ball G, Yap J. A meal high in saturated fat evokes postprandial dyslipemia, hyperinsulinemia, and altered lipoprotein expression in obese children with and without nonalcoholic fatty liver disease. JPEN J Parenter Enteral Nutr. 2013;37:517–28.

54. Parry SA, Smith JR, Corbett TR, Woods RM, Hulston CJ. Short-term, high-fat overfeeding impairs glycaemic control but does not alter gut hormone responses to a mixed meal tolerance test in healthy, normal-weight individuals. Br J Nutr. 2017;117:48–55.

55. Jackson KG, Wolstencroft EJ, Bateman PA, Yaqoob P, Williams CM. Acute effects of meal fatty acids on postprandial NEFA, glucose and apo E response: implications for insulin sensitivity and lipoprotein regulation? Br J Nutr. 2005;93:693–700.

56. Liu L, Zhao SP, Wen T, Zhou HN, Hu M, Li JX. Postprandial hypertriglyceridemia associated with inflammatory response and procoagulant state after a high-fat meal in hypertensive patients. Coron Artery Dis. 2008;19:145–51.

57. Jensen L, Sloth B, Krog-Mikkelsen I, Flint A, Raben A, Tholstrup T, Brünner N, Astrup A. A low-glycemic-index diet reduces plasma plasminogen activator inhibitor-1 activity, but not tissue inhibitor of proteinases-1 or plasminogen activator inhibitor-1 protein, in overweight women. Am J Clin Nutr. 2008;87:97–105.

58. Emerson SR, Kurti SP, Harms CA, Haub MD, Melgarejo T, Logan C, Rosenkranz SK. Magnitude and Timing of the Postprandial Inflammatory Response to a High-Fat Meal in Healthy Adults: A Systematic Review. Adv Nutr. 2017;8:213–25.

59. Orem A, Yaman SO, Altinkaynak B, Kural BV, Yucesan FB, Altinkaynak Y, Orem C. Relationship between postprandial lipemia and atherogenic factors in healthy subjects by considering gender differences. Clin Chim Acta. 2018;480:34–40.

60. Wyskida K, Franik G, Wikarek T, Owczarek A, Delroba A, Chudek J, Sikora J, Olszanecka-Glinianowicz M. The levels of adipokines in relation to hormonal changes during the menstrual cycle in young, normal-weight women. Endocr Connect. 2017;6:892–900.

61. Rafique N, Salem AM, Latif R, MH AL. Serum leptin level across different phases of menstrual cycle in normal weight and overweight/obese females. Gynecol Endocrinol. 2018;34:601–4.

62. Bonello N, Norman RJ. Soluble adhesion molecules in serum throughout the menstrual cycle. Hum Reprod. 2002;17:2272–8.

63. Koh SCL, Prasad RNV, Fong YF. Hemostatic Status and Fibrinolytic Response Potential at Different Phases of the Menstrual Cycle. Clin Appl Thromb Hemost. 2005;11:295–301.

64. Giardina EG, Chen HJ, Sciacca RR, Rabbani LE. Dynamic variability of hemostatic and fibrinolytic factors in young women. J Clin Endocrinol Metab. 2004;89:6179–84.

65. Magkos F, Fabbrini E, Patterson BW, Mittendorfer B, Klein S. Physiological interindividual variability in endogenous estradiol concentration does not influence adipose tissue and hepatic lipid kinetics in women. Eur J Endocrinol. 2022;187:391–8.

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