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Simultaneous Evaluation of Postchallenge Glycemia and Lipidemia in Young Women

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Abstract: *Aim:* To determine the effects of simultaneously ingesting glucose and fat on postchallenge glycemia and lipidemia in 12 healthy young Japanese women. *Methods:* Three test trials were administered in a randomized crossover design: glucose (1 g/kg, G trial); fat cream (0.35 g/kg as fat; F trial); glucose + fat cream (GF trial). Blood samples were taken before and at 0.5, 1, 2, 4, and 6 h post-ingestion. *Results:* The GF trial's serum glucose peak value was lower than the G trial's, and its remnant lipoprotein-cholesterol (RLP-C) increase was less than the F trial's. The GF trial's apolipoprotein (Apo) B48 increase happened faster than the F trial's. The G and GF trials' insulinogenic index (I/G30) values were negatively correlated with the area under the curve (AUC) of glucose for 2 h. Fasting insulin level and HOMA-IR were positively (and QUICKI and I/G30 were negatively) correlated with the AUC of insulin for 2 h. The F and GF trials' fasting TG, RLP-C, ApoB48, and ApoC-III levels were positively correlated with the AUC of TG and RLP-C for 6 h. The fasting ApoB48 level predicted the AUC of ApoB48 for 6 h. *Conclusion:* The glucose peak was ameliorated by co-ingesting fat. I/G30 predicted an early postchallenge (0–2 h) glucose rise. The fasting insulin level, HOMA-IR, QUICKI, and I/G30 predicted an insulin rise. The RLP-C rise by fat ingestion was ameliorated by co-ingesting glucose. Fasting TG, RLP-C, ApoB48, and ApoC-III levels predicted postchallenge TG and RLP-C rises. The fasting ApoB48 level predicted postchallenge apoB48, i.e., a rise in intestinal lipoprotein.

Keywords: Remnant Lipoprotein-Cholesterol, Apolipoprotein B48, Apolipoprotein C-III, Insulin Sensitivity, Insulin Resistance

1. Introduction

People in developed countries spend most of their waking hours in the postprandial state, and postprandial hyperglycemia and lipidemia are significant risk factors for atherosclerosis. However, glucose and lipids in the blood are usually measured in the fasting state because most of the guidelines for glucose and lipid metabolism provide diagnostic criteria that are based on fasting values. When only fasting values are used, a significant proportion of at-risk subjects could be overlooked or evaluated incorrectly. Clinical evaluations of postprandial glucose metabolism are often based on the results of a 75-g oral glucose tolerance test, but there is no standardized oral fat tolerance test. In addition, although postprandial glycemia and lipidemia were usually studied separately, common foods contain both carbohydrate and fat. If only one nutrient is examined, the effects of another nutrient will be missed. For example, D-fructose and D-allulose both ameliorate the glycemic response compared to glucose, but they delay and even exacerbate postprandial lipidemia [1, 2]. O'Keefe et al. proposed that both postprandial hyperglycemia and postprandial hyperlipidemia be dealt with together as 'postprandial dysmetabolism' [3]. Postprandial dysmetabolism is a major but less recognized disturbance in the development of atherosclerosis and coronary heart disease (CHD).

Postprandial glucose values >7.8 mmol/L (140 mg/dL) are associated with an increase in all-cause mortality [4]. Diabetes mellitus is an independent risk factor for ischemic stroke in both men and women, whereas diabetes was an independent risk factor for CHD in the women but not in the men in a Japanese population [5]. Postprandial — but not

fasting — blood glucose is an independent risk factor for cardiovascular events in type 2 diabetes with a stronger predictive power in women than in men [6]. These findings suggest that glucose intolerance and diabetes may give rise to more serious outcomes in women than in men.

Approximately 40% of all cases of premature atherosclerosis develops in fasting normolipidemic individuals [7, 8], but most people are in the nonfasting state for most of the day, and this may be a better reflection of their true metabolic state. Although fat-loading studies demonstrated that the fasting triglyceride (TG) level is positively correlated with postprandial triglyceridemia [9], the nonfasting TG level has a stronger association with cardiovascular disease compared to fasting TG [10]. Small changes in fasting TG can lead to larger differences in diurnal triglyceridemia, probably due to the competition for removal mechanisms by lipoprotein lipase between endogenous and exogenous TG-rich lipoprotein (TRL) particles [11].

Nonfasting TG levels are associated with incident cardiovascular events independently of traditional cardiac risk factors, levels of other lipids, and markers of insulin resistance, particularly in women [11–14]. The elevation of the remnant-like particle-cholesterol level has been reported to be a better CHD risk marker than the serum TG level, particularly in women [15]. TG levels increase in older individuals and are thought to be a significant risk factor for CHD, particularly in women [16]. Regarding the mechanism of atherogenesis, both postprandial hyperglycemia and hyperlipidemia may initiate and accelerate the development of atherosclerosis by stimulating the inflammatory process and the production of active oxygen/free radicals [17–19].

In Japan, the ingestion of fast food that is rich in saturated fat and processed carbohydrates such as sucrose and high-fructose corn syrup is higher among university students than in high-school students [20]. Young women are generally rather resistant to glycemic and lipidemic loads and to diabetes mellitus, but once they develop such disease, the harm is more severe for women than for men.

We sought to determine the effects of the simultaneous ingestion of glucose and fat on postchallenge glycemia and lipidemia, and we attempted to identify the parameters that can be used to predict postprandial glycemia and lipidemia in healthy young Japanese women.

2. Methods

2.1. Subjects

Twelve healthy young Japanese women (age 21.1 ± 0.2 yr) with a normal ovarian cycle and apolipoprotein E phenotype 3/3 were enrolled. They were non-smokers, were not suffering from any apparent acute or chronic illness, and were not taking any medications or dietary supplements. This study was approved by the Institutional Review Board of the Sugiyama Jogakuen University School of Life Studies (No. 2017-28). The subjects provided written informed consent. The procedures were conducted in accord with the Helsinki Declaration of 1975 as revised in 1983.

2.2. Anthropometric and Body Composition Measurements

Each subject's body mass and height were measured according to standard methods. The waist circumference was assessed as the abdominal girth at the level of the umbilicus, and the hip circumference was measured at the level of the greater trochanters. The waist-to-hip (W/H) ratio was calculated. The body composition including the visceral fat area (VFA) was analyzed by an 8-polar bioelectrical impedance method (InBody720, BioSpace, Tokyo, Japan).

2.3. Glucose and Fat Loads

Each subject was studied on 3 trial occasions. At each test trial, the subjects ingested one of 3 beverages containing glucose (1 g/kg body mass) or OFTT cream (Jomo, Takasaki, Japan; 1 g/kg as cream, 0.35 g/kg as fat) or both the glucose and the cream, in a randomized crossover design. The OFTT cream was used as described [21]. The 3 beverages were prepared as follows. The glucose beverage (G trial) was prepared by mixing 1 g/kg of glucose (Marugo, Soka, Japan) with distilled water (5.1 mL/kg). The fat beverage (F trial) was prepared by mixing 1 g/kg of OFTT cream (0.35 g/kg as fat) with the same final amount of distilled water. The glucose + fat beverage (GF trial) was prepared by mixing glucose (1 g/kg) and OFTT cream (1 g/kg) with the same final amount of water. The total volume of each beverage was approx. 300 mL.

2.4. Experimental Design

One of the 3 beverages mentioned above was administered after a 12-h overnight fast. The subjects abstained from consuming alcohol on the day before each trial. Venous blood samples were taken before (0 h) and at 0.5, 1, 2, 4, and 6 h after the beverage ingestion. During the test, the subjects avoided exercise and eating, but had free access to water at 1 h after the ingestion. The blood samples were taken with the subject in the supine position. The trials were performed ≥4 wk apart. The interval of 4 wk between the trial days was to minimize the confounding effects of the subjects' menstrual status on their lipid metabolism. One of the subjects ingested the wrong formula of beverage in the G and GF trials and was excluded from the analysis. The number of subjects in the G and GF trials was thus 11.

2.5. Biochemical Analysis

Serum samples were immediately refrigerated (4°C) or frozen (−80°C) until analysis. The level of glucose was measured by a mutarotase-glucose oxidase method (Wako, Osaka, Japan). The level of insulin was measured by a chemiluminescent enzyme immunoassay (Fujirebio, Tokyo). The hemoglobin A1c (HbA1c) was measured by a latex agglutination method (Fujirebio) and was expressed as the National Glycohemoglobin Standardization Program (NGSP) value. The level of glycoalbumin (GA) was measured enzymatically (Sekisui Medical, Tokyo). 1,5-Anhydroglucitol

(1,5-AG) was determined by an enzymatic method (Determiner L 1,5-AG, Kyowa Medex, Tokyo). Insulin resistance was evaluated by the homeostasis model assessment for insulin resistance (HOMA-IR) [22]. The quantitative insulin sensitivity check index (QUICKI) [23] was calculated as follows;

$$QUICKI = 1/[\log(\text{fasting glucose, mg/dL}) + \log(\text{fasting insulin, mIU/L})]$$

The insulinogenic index (I/G30) was calculated as:

$$I/G30 = \Delta\text{insulin (30 min – 0 min, mIU/L)}/\Delta\text{glucose (30 min – 0 min, mg/dL)}$$

The level of free fatty acid (FFA) was measured enzymatically (Wako). The level of total cholesterol (TC) was measured enzymatically (Sysmex, Hyogo, Japan). The level of high-density lipoprotein-cholesterol (HDL-C) was measured by a direct method (Fujirebio). Low-density lipoprotein-cholesterol (LDL-C) was calculated by the Friedewald formula. Small, dense LDL-C (sdLDL-C) was measured enzymatically (Denka Seiken, Tokyo). TG was measured enzymatically (Sekisui Medical). The level of remnant lipoprotein-cholesterol (RLP-C) was measured with a homogeneous assay (MetaboRead RemL-C, Kyowa Medex, Tokyo). Lipoprotein(a), i.e., Lp(a) was measured by a latex agglutination method (Sekisui Medical). Apolipoproteins (Apo) A-I, A-II, B, C-II, C-III, and E were measured by the immunoturbidimetric method (Sekisui Medical). ApoB48 was measured by a chemiluminescent enzyme immunoassay (Fujirebio). The concentration of ApoB100 was calculated by subtracting the value of ApoB48 from the value of ApoB [24]. The ApoE phenotype was measured using the isometric electrophoresis method (Phenotyping ApoE IEF System, Joko, Tokyo).

2.6. The Quantification of the Subjects' Postchallenge Metabolism

Postchallenge changes in the concentrations of serum glucose, insulin, TG, RLP-C, ApoB100, and ApoB48 were calculated as the difference from the baseline mean value (as 0 at 0 h) and are shown as Δglucose, Δinsulin, ΔTG, ΔRLP-C, ΔApoB100, and ΔApoB48, respectively. The subjects' postchallenge metabolism was quantified by calculating the area under the curve (AUC) from 0 to 2, 4, or 6 h by the trapezoidal method.

2.7. Statistical Analyses

The statistical analyses were performed using SPSS ver. 25 software (IBM, Tokyo). A normal distribution of data was verified using the Shapiro-Wilk test for skewness and kurtosis of distribution. Non-normally distributed data were examined for a normal distribution when logarithmically transformed, and they were analyzed using parametric statistics. The data are presented as the mean ± SEM. The differences in the time-course compared with the fasting values were analyzed by performing a repeated measures analysis of variance (ANOVA), followed by the Dunnett test. The measured value differences at each time-point in the three trials were assessed by a repeated measures ANOVA followed by the Bonferroni test. For the correlation analysis, Pearson's correlation coefficient was calculated. For all data, $p<0.05$ was considered significant.

3. Results

The physical characteristics and fasting blood chemical data of the subjects are summarized in Table 1. There were no significant differences in any of the physical characteristics in the 3 trials (data not shown). All 3 beverages were well tolerated by the subjects, and none reported nausea, vomiting, or diarrhea during or after the trials.

Table 1. *Physical characteristics and fasting blood chemical data of the subjects.*

Age (years)	21.1	±	0.2	HOMA-IR	1.2	±	0.1
Height (cm)	160.5	±	1.4	HbA1c (%)	5.4	±	0.1
Mass (kg)	49.5	±	1.0	GA (mg/dL)	14.0	±	0.1
BMI (kg/m^2)	19.2	±	0.4	1,5-AG (mg/L)	21.3	±	1.7
Waist (cm)	67.8	±	0.9	TC (mg/dL)	161.2	±	7.8
Hip (cm)	89.3	±	1.0	HDL-C (mg/dL)	60.8	±	3.0
W/H	0.76	±	0.01	LDL-C (mg/dL)	88.0	±	6.3
VFA (cm^2)	22.4	±	3.0	sdLDL-C (mg/dL)	15.3	±	1.3
SBP (mmHg)	105.6	±	2.6	apoE phenotype		E3/E3	
DBP (mmHg)	66.6	±	2.4				
PR (beats/min)	84.0	±	3.4				

SBP: systolic blood pressure, DBP: diastolic blood pressure, PR: pulse rate.

3.1. Glucose, Insulin, and FFA

The fasting and postchallenge chemical data of serum glucose, insulin, and FFA in the 3 trials are presented in Table 2. The time courses of Δglucose and Δinsulin are shown in Figure 1. The serum concentrations of glucose in the G and GF trials were significantly increased at 0.5 h compared to the fasting levels. The concentrations of glucose in the GF trial were significantly lower compared to those in the G trial at 0.5 and 1 h. In the G and GF trials,

the glucose concentration peaked at 0.5–1 h and returned to baseline at 4 h. No significant rise in the glucose concentration was observed during the F trial (no glucose load).

The serum insulin concentrations in the G and GF trials were significantly higher compared to those in the F trial at 0.5, 1, 2, and 6 h. In the G and GF trials, the insulin concentration peaked at 0.5 h and returned to or was below baseline at 4 h. There were no significant differences in these data between the G and GF trials. In the F trial, the insulin level increased slightly but significantly at 0.5 h.

The serum concentrations of FFA decreased immediately in the G and GF trials but not in the F trial, reaching the nadir at 1–2 h. The FFA level then tended to increase toward the end of the trial. In the G and F trials, the FFA concentrations at 6 h were significantly increased compared to the fasting levels.

Table 2. *Fasting and postchallenge concentrations of serum glucose, insulin, and FFA.*

	Trial	0	0.5	1	2	4	6	(h)
Glucose (mg/dL)	G	77.5 ± 2.1	125.2 ± 4.4*	125.2 ± 6.7*	92.2 ± 4.9*	69.7 ± 1.9	75.7 ± 1.6	
	F	81.5 ± 1.3	77.5 ± 1.4*	77.8 ± 1.2*	81.1 ± 1.6	77.9 ± 1.3*	74.4 ± 2.1*	
	GF	78.3 ± 1.4	110.7 ± 5.0*	105.7 ± 6.6*	106.4 ± 9.5*	74.0 ± 1.7	75.8 ± 1.2	
Insulin (mIU/L)	G	6.1 ± 0.6	46.3 ± 5.0*	40.1 ± 5.0*	19.6 ± 2.7*	4.3 ± 0.5*	4.9 ± 0.4	
	F	6.0 ± 0.7	8.6 ± 0.9*	6.9 ± 0.7	5.2 ± 0.6	3.2 ± 0.5*	3.1 ± 0.5*	
	GF	7.1 ± 0.7	41.1 ± 7.2*	40.2 ± 7.0*	26.7 ± 1.6*	4.3 ± 0.64*	4.0 ± 0.4*	
FFA (mmol/L)	G	0.42 ± 0.05	0.36 ± 0.09	0.10 ± 0.01*	0.05 ± 0.01*	0.80 ± 0.09	0.92 ± 0.13*	
	F	0.45 ± 0.05	0.46 ± 0.04	0.41 ± 0.05	0.64 ± 0.08	0.70 ± 0.08	1.09 ± 0.14*	
	GF	0.58 ± 0.15	0.39 ± 0.07	0.16 ± 0.03*	0.16 ± 0.04*	0.56 ± 0.07	0.79 ± 0.08	

Values are mean ± SEM. *p<0.05 vs. the fasting value. #p<0.05 between the trials.

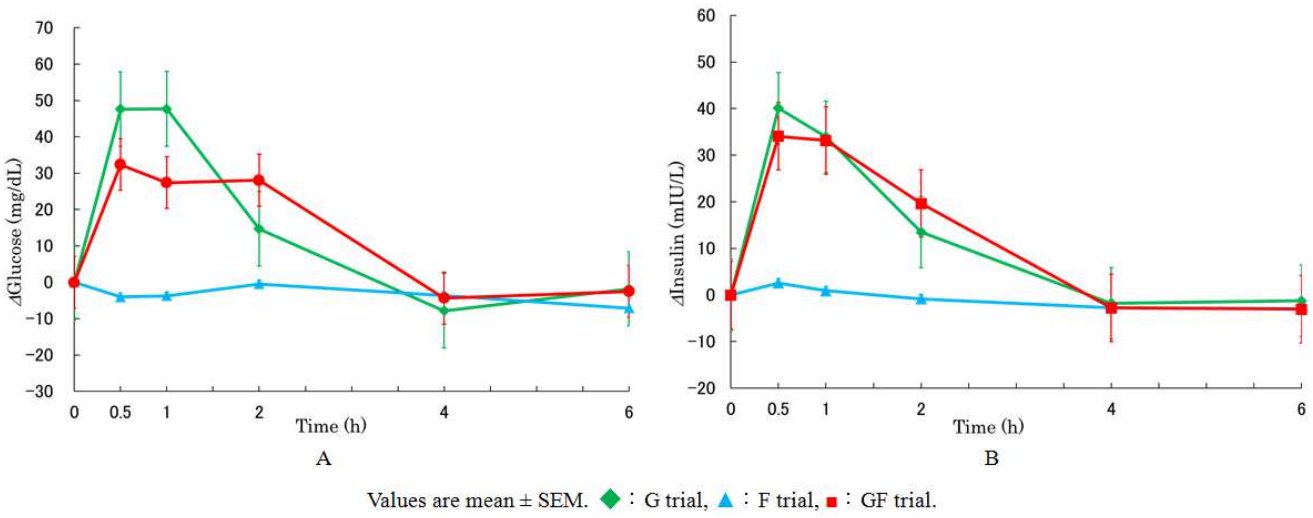

Figure 1. *Postchallenge Δglucose (A) and Δinsulin (B).*

3.2. TG, RLP-C, ApoB48, and ApoB100

The fasting and postchallenge chemical data of TG, RLP-C, ApoB48, and ApoB100 in the 3 trials are presented in Table 3. The time courses of *Δ*TG, *Δ*RLP-C, *Δ*ApoB48, and *Δ*ApoB100 are illustrated in Figure 2. The serum concentration of TG significantly increased at 2 and 4 h in the F and GF trials, and it peaked at 2 h in both trials. At 6 h, the TG level had returned to or was below baseline in the trials. The peak value of TG at 2 h was slightly lower in the GF trial compared to the F trial, but not significantly so. No increase in the TG concentration was observed in the G trial (no fat load), and the TG concentration was significantly decreased at 2 and 4 h compared to the baseline.

Table 3. *Fasting and postchallenge concentrations of serum TG, RLP-C, ApoB48, and ApoB100.*

	Trial	0	1	2	4	6	(h)
TG (mg/dL)	G	74.6 ± 10.1	62.0 ± 4.0	53.0 ± 4.4*	58.5 ± 4.6*	67.2 ± 6.2	
	F	66.6 ± 8.5	70.7 ± 8.7	88.2 ± 11.5*	77.0 ± 11.1*	53.3 ± 6.6*	
	GF	65.8 ± 11.5	72.6 ± 10.6	79.8 ± 12.6*	77.2 ± 8.9*	55.9 ± 6.4	
RLP-C (mg/dL)	G	2.7 ± 0.3	2.5 ± 0.3	2.3 ± 0.3	2.5 ± 0.3	2.9 ± 0.4	
	F	3.0 ± 0.5	3.1 ± 0.5	3.5 ± 0.6*	3.7 ± 0.7*	2.8 ± 0.4	
	GF	3.0 ± 0.6	3.1 ± 0.5	3.2 ± 0.5	3.4 ± 0.5	2.9 ± 0.4	
ApoB48 (mg/L)	G	2.4 ± 0.3	2.4 ± 0.3	2.5 ± 0.3	1.8 ± 0.3*	1.2 ± 0.1*	
	F	2.5 ± 0.4	3.0 ± 0.3	3.9 ± 0.5*	3.9 ± 0.5*	2.9 ± 0.2	
	GF	2.0 ± 0.4	3.6 ± 0.5*	3.7 ± 0.5*	3.5 ± 0.5	2.2 ± 0.3	

	Trial	0			1			2			4			6			(h)
ApoB100 (mg/dL)	G	64.6	±	4.3	61.5	±	4.1*	61.3	±	4.0*	63.4	±	4.0	63.3	±	4.0	
	F	63.2	±	3.9	62.3	±	4.2	61.7	±	4.2*	62.8	±	4.1	63.5	±	4.2	
	GF	64.4	±	4.7	62.6	±	4.4*	60.6	±	4.4*	63.3	±	4.8	64.5	±	4.8	
ApoC-II (mg/dL)	G	2.8	±	0.2	2.7	±	0.2*	2.8	±	0.2	2.8	±	0.2	2.7	±	0.2	
	F	2.6	±	0.2	2.6	±	0.2	2.5	±	0.2	2.3	±	0.2*	2.3	±	0.2*	
	GF	2.5	±	0.3	2.4	±	0.3	2.4	±	0.3	2.3	±	0.3*	2.1	±	0.3*	
ApoC-III (mg/dL)	G	7.8	±	0.4	7.6	±	0.40	7.6	±	0.4	7.4	±	0.4*	7.1	±	0.4*	
	F	7.4	±	0.5	7.1	±	0.50	6.7	±	0.5*	6.5	±	0.5*	6.3	±	0.4*	
	GF	7.2	±	0.7	7.0	±	0.66	6.6	±	0.6*	6.5	±	0.6*	5.9	±	0.5*	

Values are mean ± SEM. *p<0.05 vs. the fasting value. # p<0.05 between the trials.

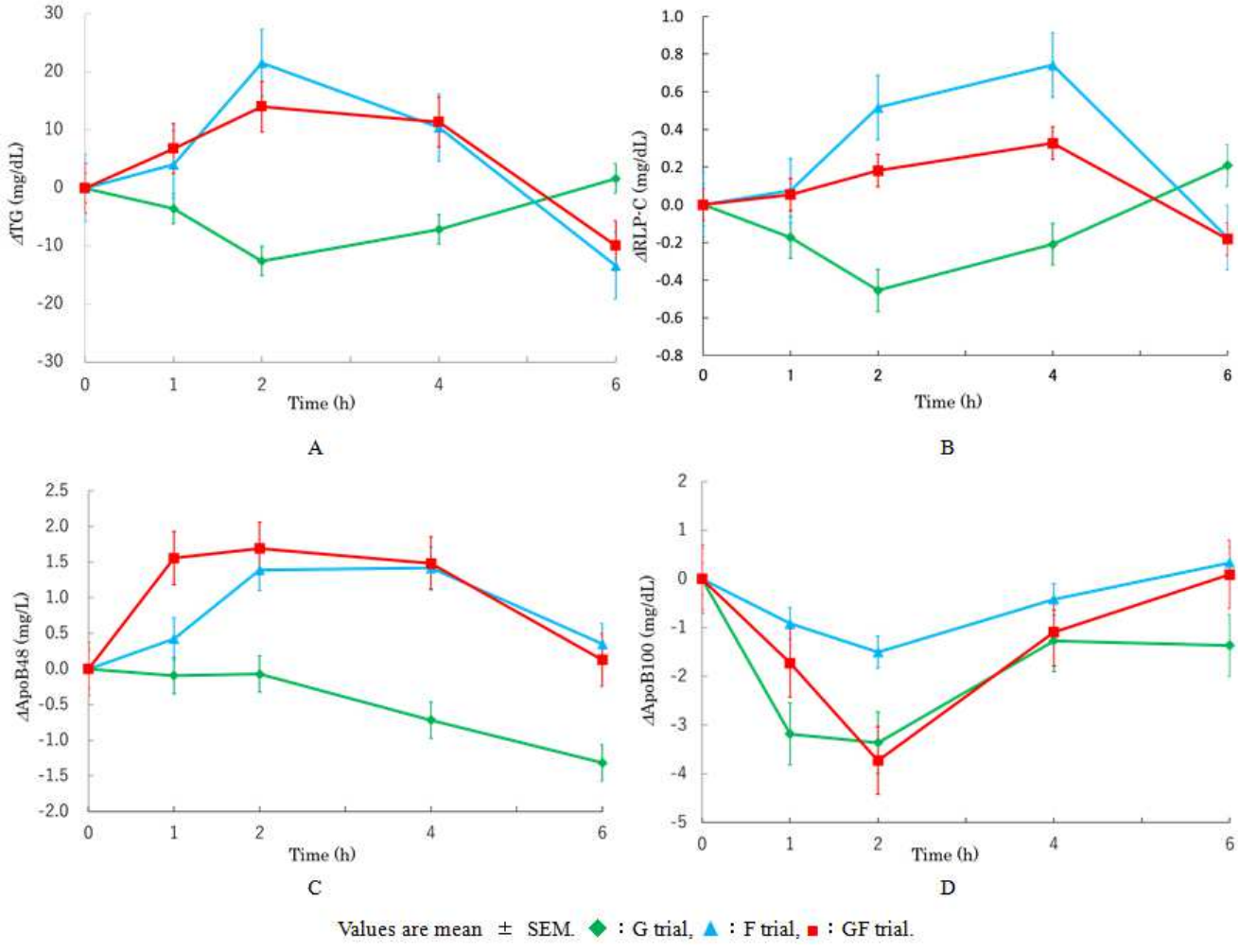

Figure 2. *Postchallenge ΔTG (A), ΔRLP-C (B), ΔApoB48 (C), and ΔApoB100 (D).*

The RLP-C concentration was significantly increased at 2–4 h, and returned to baseline at 6 h in the F trial. The increase in the RLP-C concentration in the GF trial was not significant. The RLP-C levels at 2 and 4 h tended to be lower in the GF trial compared to the F trial.

The concentration of ApoB48 was significantly increased at 2 h, peaked at 4 h, and returned to baseline at 6 h in the F trial. In the GF trial, it increased significantly at 1 h and peaked at 2 h. In the G trial, no increase in the ApoB48 concentration was observed, and it has fallen below the baseline value at 4 and 6 h. The ApoB100 concentration was decreased at 1 and 2 h in the G and GF trials, and it returned to baseline thereafter. In the F trial, a decrease in the ApoB100 concentration was observed at 2 h.

Table 4. *Correlation analysis between the indices of glucose metabolism and the AUC(0-2h) of insulin and glucose in the G and GF trials.*

	Trial	AUC(0–2h)	r
Fasting insulin	G	Insulin	0.648*
	GF	Insulin	0.713*
QUICKI	G	Insulin	-0.688*
	GF	Insulin	-0.688*
HOMA-IR	G	Insulin	0.686*
	GF	Insulin	0.691*
I/G30	G	Glucose	-0.756**
	GF	Glucose	-0.610*

*p<0.05, **p<0.01.

3.3. Correlation Analysis Between Indices of Glucose Metabolism and the AUC of Insulin and Glucose in the G and GF Trials

In the G and GF trials, the subjects' values of fasting insulin and HOMA-IR were positively correlated and the QUICKI values were negatively correlated with the AUC(0–2 h) of insulin. I/G30 was negatively correlated with the AUC(0–2h) of glucose (Table 4). HbA1c, GA, and 1,5-AG showed no significant correlation with the AUC(0–2h) of glucose or insulin (data not shown).

3.4. Correlation Analysis of the Fasting Values of Lipid Metabolism

In the F and GF trials, the fasting values of TG, RLP-C, ApoB48, and ApoC-III were positively correlated with each other (Table 5).

Table 5. *Correlation analysis between the fasting values and the AUC(0-6h) or the peaks of lipid metabolism in the F and GF trials.*

F trial		Fasting				AUC(0–6h))			Peak		
		TG	**RLP-C**	**ApoB48**	**ApoC-III**	**TG**	**RLP-C**	**ApoB48**	**TG**	**RLP-C**	**ApoB48**
Fasting	TG		0.906**	0.772**	0.680*	0.962**	0.850**	0.633*	0.957**	0.839**	0.650*
	RLP-C	0.906**		0.794**	0.661*	0.931**	0.983**	0.677*	0.910**	0.980**	0.654*
	ApoB48	0.772**	0.794**		0.802**	0.757**	0.783**	0.760**	0.737**	0.773**	0.710**
	ApoC-III	0.680*	0.661*	0.802**		0.670*	0.631*		0.615*	0.599*	

GF trial		Fasting				AUC(0–6h)			Peak		
		TG	**RLP-C**	**ApoB48**	**ApoC-III**	**TG**	**RLP-C**	**ApoB48**	**TG**	**RLP-C**	**ApoB48**
Fasting	TG		0.895**	0.729*	0.869**	0.957**	0.884**		0.942**	0.875**	
	RLP-C	0.895**		0.670*	0.802**	0.860**	0.935**		0.780**	0.928**	
	ApoB48	0.729*	0.670*		0.924**	0.762**	0.775**	0.793**	0.734*	0.778**	0.736**
	ApoC-III	0.869**	0.802**	0.924**		0.895**	0.882**	0.764**	0.890**	0.876**	0.726*

*p<0.05, **p<0.01.

3.5. Correlation Analysis of the Fasting Values and the AUC or Peaks of Lipid Metabolism in the F and GF Trials

In the F and GF trials, the fasting TG, RLP-C, ApoB48, and ApoC-III values were positively correlated with the AUC(0–6h) of TG and RLP-C (Table 5). Fasting ApoB48 was correlated with the AUC of ApoB48 in both the F and GF trials, but ApoC-III was correlated with the AUC of ApoB48 only in the GF trial. Fasting HDL-C and sdLDL-C showed no significant correlation with these parameters (data not shown).

4. Discussion

The rise of the serum glucose concentration of healthy young Japanese women was higher after the glucose load without fat compared to the glucose load with fat, suggesting that the co-presence of fat ameliorated the increase in the serum glucose concentration, although no such difference was observed in the insulin concentration. In the co-presence of glucose with fat, the subjects' serum TG peak was slightly lower and the rise of RLP-C tended to be lower compared to the fat load only. These results suggest that the absorption and metabolism of glucose and fat are mutually interrelated.

4.1. ApoC-III

In the fat load test (with or without glucose load), the fasting ApoC-III levels predicted postchallenge lipidemia and was correlated with the AUC of TG and RLP-C. A significant proportion of newly synthesized ApoC-III is secreted on very-low-density lipoprotein (VLDL) particles in the fasting state, and this proportion increases in the fed state [25]. Elevated plasma ApoC-III concentrations are associated with increased plasma concentrations of remnant-like particle-cholesterol and ApoB48, and the delayed catabolism of remnant-like particles is related to elevated ApoC-III [26]. When VLDL particles are cleared from the circulation, their ApoC-III components are not necessarily catabolized with them [25]. Accordingly, the fate of ApoC-III in the circulation is not the same as that of VLDL particles, and ApoC-III is an independent predictor of coronary events and the progression of CHD [27–32].

4.2. ApoB48

In the fat load tests (with or without glucose load), the subjects' fasting ApoB48 levels predicted the AUC of TG, RLP-C, and ApoB48 itself. The postprandial increase in the level of TG is accounted for by 80% ApoB48-containing exogenous TRL, with the remaining attributed to ApoB100-containing endogenous TRL [33]. The increase in TRL-TG is greater than the increase in ApoB48. ApoB48 secretion as chylomicron (CM) particles is stimulated by lipid absorption, but the increase in the capacity for the transport of ingested lipid occurs mainly by lipid enrichment and a marked enlargement of each CM particle [34, 35]. Therefore, the increase in ApoB48 (i.e., the CM particle number) was modest compared to the rise in TG. After a meal, the CM-TG concentration rises and peaks after 3–4 h, whereas the TG level in VLDL particles remains relatively constant or peaks later at 4–6 h [36–38].

The intestine secretes ApoB48-containing lipoproteins with sizes that vary widely [39], and, in the fasting state, the intestine still secretes ApoB48-containing lipoprotein as HDL-sized particles [40]. In the present G trial (without fat), the subjects' ApoB48 levels further decreased after 4 and 6 h, suggesting that the 'fasting' level of ApoB48 in the morning after the subjects'

overnight fast may not be the 'true' fasting level, consistent with our previous observations [41]. Interestingly, even in the present healthy young women without glucose intolerance, diabetes mellitus or fasting dyslipidemia, the fasting value of ApoB48 predicted postchallenge lipidemia.

TG-rich CM secreted by the intestine begins to appear in the blood approx. 1 h after an oral intake of fat, and the half-life of TG in CM is estimated to be approx. 5–8 min [42, 43]. It is calculated that approx. one-half of the TG in the CM particles leaves the CM particles in the process of remnant particle formation [44]. However, ApoB48 remains with the lipoprotein particles until receptor-mediated uptake occurs, indicating that the plasma ApoB48 level reflects the number of exogenous lipoprotein particles. The residence time of ApoB48 in the circulation was estimated to be 4.8 h [45].

One plausible causal risk factor for postprandial lipidemia may lie in the postprandial cholesterol metabolism, specifically the amount of cholesterol in remnant lipoproteins, with which plasma TG levels are strongly correlated [46]. It is likely that the late postchallenge increase of RLP-C observed in the present study took place largely in the endogenous TRL.

4.3. sdLDL

The atherogenicity of LDL depends mainly on its small size, increased density and increased particle number, but not by its cholesterol content per se [47]. These abnormalities in LDL have been known to be the result of defective TG metabolism. An increased number of LDL particles is the main source of atherogenicity associated with sdLDL. Although the presence of sdLDL is closely linked to the fasting plasma TG concentration, we observed no correlation between the sdLDL level and the postchallenge AUC of TG in the present study. This is probably because in young women, the TG level did not reach the threshold of approx. 1.5 mmol/L [48].

Young women usually have low plasma TG concentrations, reduced penetrance of sdLDL (pattern B), and low hepatic lipase activity [49]. Even if small amounts of sdLDL are formed, they would be efficiently removed from the circulation, and thus the accumulation of sdLDL is unlikely. However, prolonged or exaggerated postprandial lipidemia occurring after repeated meals could lead to more atherogenic changes in LDL particles. Interestingly, the LDL particle size was reported to be associated with male longevity, whereas the TG level, but not the LDL particle size, was associated with female longevity [50].

4.4. The HDL-C level and the VFA

An inverse relationship has been demonstrated between HDL-C levels and the magnitude of postprandial lipidemia [51], but in the present study, no correlation was observed between the fasting HDL-C level and postchallenge lipidemia. It was reported that increased fat mass, especially visceral fat, results in enhanced postprandial triglyceridemia [52–54]. However, we detected no correlation in the present study between postchallenge lipidemia and the VFA, probably because the subjects were all lean and their VFA values were low.

4.5. Insulin Sensitivity and Insulin Resistance

Insulin inhibits the assembly and secretion of VLDL1 particles by mechanisms involving an increase in ApoB100 degradation [55]. The lower postchallenge increase of RLP-C in the present study's GF trial compared to the F trial may have been caused by the inhibition of the secretion of VLDL (particularly VLDL1) by insulin. The concentration, the fractional clearance, and production of TRL-ApoB100 were not significantly affected by glucose [56]. However, in the present study, a slight but significant decrease in the ApoB100 concentration was observed after the glucose load (the G and GF trials). The postchallenge glycemic peak — but not the total area of glycemia — induced by the glucose load was ameliorated in the co-presence of fat (GF trial vs. G trial).

The fasting insulin value, the QUICKI value, and the HOMA-IR result predicted postchallenge insulinemia with or without fat co-loading, whereas I/G30 predicted postchallenge glycemia. These parameters may be valuable for predicting postprandial glycemia and insulinemia in individuals who do not have glucose intolerance or diabetes mellitus. However, the parameters that predicted postchallenge glucose metabolism were few in number compared to those predicting postchallenge lipidemia.

4.6. Study Limitations

This study has some limitations. Because of the small number of subjects, the results should be interpreted with caution. Postchallenge dysmetabolism (glycemia and lipidemia) can be used as a surrogate for postprandial dysmetabolism, but it is not equivalent to it. Therefore, postprandial dysmetabolism should be examined next after the consumption of a meal. Another study limitation is that only healthy young females were enrolled, and it remains unknown whether these results are applicable to other subjects.

5. Conclusion

The absorption and metabolism of glucose and fat are mutually interrelated. The rise of our subjects' serum glucose concentrations was higher after the glucose load only (without fat) compared to the glucose load with fat, with no difference in insulin concentration. In the co-presence of glucose with fat, the serum TG peak was slightly lower and the rise of RLP-C tended to be lower compared to the fat load only. The fasting TG, RLP-C, ApoB48, and ApoC-III levels predicted postchallenge TG and RLP-C increases. The fasting ApoB48 level also predicted postchallenge apoB48, i.e., an intestinal lipoprotein rise.

Conflicts of Interest

The authors declare that they have no competing interests.

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References

[1] Saito H, Kagaya M, Suzuki M, Yoshida A, Naito M: Simultaneous ingestion of fructose and fat exacerbates postprandial exogenous lipidemia in young healthy Japanese women. J Atheroscler Thromb, 2013; 20: 591-600.

[2] Kuzawa K, Sui L, Hossain A, Kamitori K, Tsukamoto I, Yoshida A, Tokuda M, Naito M: Effects of a rare sugar, D-allulose, coingested with fat on postprandial glycemia and lipidemia in young women. J Nutr Hum Health, 2019; 3: 1-6.

[3] O'Keefe JH, Bell DS: Postprandial hyperglycemia/hyperlipidemia (postprandial dysmetabolism) is a cardiovascular risk factor. Am J Cardiol, 2007; 100: 899-904.

[4] American Diabetes Association: Standards of medical care in diabetes–2016 abridged for primary care providers. Clin Diabetes, 2016; 34: 3-21.

[5] Doi Y, Ninomiya T, Hata J, Fukuhara M, Yonemoto K, Iwase M, Iida M, Kiyohara Y: Impact of glucose tolerance status on development of ischemic stroke and coronary heart disease in a general Japanese population: the Hisayama study. Stroke, 2010; 41: 203-209.

[6] Cavalot F, Petrelli A, Traversa M, Bonomo K, Fiora E, Conti M, Anfossi G, Costa G, Trovati M: Postprandial blood glucose is a stronger predictor of cardiovascular events than fasting blood glucose in type 2 diabetes mellitus, particularly in women: lessons from the San Luigi Gonzaga Diabetes Study. J Clin Endocrinol Metab, 2006; 91: 813-819.

[7] Genest JJ, McNamara JR, Salem DN, Schaefer EJ: Prevalence of risk factors in men with premature coronary artery disease. Am J Cardiol, 1991; 67: 1185-1189.

[8] Miller M, Seidler A, Moalemi A, Pearson TA: Normal triglyceride levels and coronary artery disease events: the Baltimore Coronary Observational Long-Term Study. J Am Coll Cardiol, 1998; 31: 1252-1257.

[9] Schaefer EJ, Lamon-Fava S, Cohn SD, Schaefer MM, Ordovas JM, Castelli WP, Wilson PW: Effects of age, gender, and menopausal status on plasma low density lipoprotein cholesterol and apolipoprotein B levels in the Framingham Offspring Study. J Lipid Res, 1994; 35: 779-792.

[10] Mora S, Rifai N, Buring JE, Ridker PM: Fasting compared with nonfasting lipids and apolipoproteins for predicting incident cardiovascular events. Circulation, 2008; 118: 993-1001.

[11] Brunzell JD, Hazzard WR, Porte D Jr, Bierman EL: Evidence for a common, saturable, triglyceride removal mechanism for chylomicrons and very low density lipoproteins in man. J Clin Invest, 1973; 52: 1578-1585.

[12] Nordestgaard BG, Benn M, Schnohr P, Tybjaerg-Hansen A: Nonfasting triglycerides and risk of myocardial infarction, ischemic heart disease, and death in men and women. JAMA, 2007; 298: 299-308.

[13] Freiberg JJ, Tybjaerg-Hansen A, Jensen JS, Nordestgaard BG: Nonfasting triglycerides and risk of ischemic stroke in the general population. JAMA, 2008; 300: 2142-2152.

[14] Langsted A, Freiberg JJ, Tybjaerg-Hansen A, Schnohr P, Jensen GB, Nordestgaard BG: Nonfasting cholesterol and triglycerides and association with risk of myocardial infarction and total mortality: the Copenhagen City Heart Study with 31 years of follow-up. J Intern Med, 2011; 270: 65-75.

[15] McNamara JR, Shah PK, Nakajima K, Cupples LA, Wilson PW, Ordovas JM, Schaefer EJ: Remnant-like particle (RLP) cholesterol is an independent cardiovascular disease risk factor in women: results from the Framingham Heart Study. Atherosclerosis, 2001; 154: 229-236.

[16] LaRosa JC: Triglycerides and coronary risk in women and the elderly. Arch Intern Med, 1997; 157: 961-968.

[17] Williams SB, Goldfine AB, Timimi FK, Ting HH, Roddy M-A, Simonson DC, Creager MA: Acute hyperglycemia attenuates endothelium-dependent vasodilation in humans in vivo. Circulation, 1998; 97: 1695-1701.

[18] Bae JH, Bassenge E, Kim KB, Kim YN, Kim KS, Lee HJ, Moon KC, Lee MS, Park KY, Schwemmer M: Postprandial hypertriglyceridemia impairs endothelial function by enhanced oxidant stress. Atherosclerosis, 2001; 155: 517-523.

[19] Ceriello A, Genovese S: Atherogenicity of postprandial hyperglycemia and lipotoxicity. Rev Endocr Metab Disord, 2016; 17: 111-116.

[20] Asano M, Fukakura N, Odachi J, Kawaraya C, Nanba A, Yasuda N, Yamamoto E: Use of fast foods among young people. Jpn J Nutr Diet, 2003; 61: 47-54 (In Japanese, Abstract in English).

[21] Nabeno-Kaeriyama Y, Fukuchi Y, Hayashi S, Kimura T, Tanaka A, Naito M: Delayed postprandial metabolism of triglyceride-rich lipoproteins in obese young men compared to lean young men. Clin Chim Acta, 2010; 411: 1694-1699.

[22] Matthews DR, Hosker JP, Rudenski AS, Naylor BA, Treacher DF, Turner RC: Homeostasis model assessment: insulin resistance and β-cell function from fasting plasma glucose and insulin concentrations in man. Diabetologia, 1985; 28: 412-419.

[23] Katz A, Nambi SS, Mather K, Baron AD, Follmann DA, Sullivan G, Quon MJ: Quantitative insulin sensitivity check index: a simple, accurate method for assessing insulin sensitivity in humans. J Clin Endocrinol Metab, 2000; 85: 2402-2410.

[24] Nakatani K, Sugimoto T, Masuda D, Okano R, Oya T, Monden Y, Yamashita T, Kawase R, Nakaoka H, Inagaki M, Yuasa-Kawase M, Tsubakio-Yamamoto K, Ohama T, Nishida M, Ishigami M, Komuro I, Yamashita S: Serum apolipoprotein B-48 levels are correlated with carotid intima-media thickness in subjects with normal serum triglyceride levels. Atherosclerosis, 2011; 218: 226-232.

[25] Batal R, Tremblay M, Barrett PHR, Jacques H, Fredenrich A, Mamer O, Davignon J, Cohn JS: Plasma kinetics of ApoC-III and ApoE in normolipidemic and hypertriglyceridemic subjects. J Lipid Res, 2000; 41: 706-718.

[26] Chan DC, Watts GF, Barrett PH, Mamo JCL, Redgrave TG: Markers of triglyceride-rich lipoprotein remnant metabolism in visceral obesity. Clin Chem, 2002; 48: 278-283.

[27] Chivot L, Mainard F, Bigot E, Bard JM, Auget JL, Madec Y, Fruchart JC: Logistic discriminant analysis of lipids and apolipoproteins in a population of coronary bypass patients and the significance of apolipoproteins C-III and E. Atherosclerosis, 1990; 82: 205-211.

[28] Hodis HN, Mack WJ, Azen SP, Alaupovic P, Pogoda JM, LaBree L, Hemphill LC, Kramsch DM, Blankenhorn DH: Triglyceride- and cholesterol-rich lipoproteins have a differential effect on mild/moderate and severe lesion progression as assessed by quantitative coronary angiography in a controlled trial of lovastatin. Circulation, 1994; 90: 42-49.

[29] Luc G, Fievet C, Arveiler D, Evans AE, Bard JM, Cambien F, Fruchart JC, Ducimetiere P: Apolipoproteins C-III and E in apoB- and non-apoB-containing lipoproteins in two populations at contrasting risk for myocardial infarction: the ECTIM study. Etude Cas Témoins sur 'Infarctus du Myocarde. J Lipid Res, 1996; 37: 508-517.

[30] Korena E, Corder C, Mueller G, Centurion H, Hallum G, Fesmire J, McConathy WD, Alaupovic P: Triglyceride enriched lipoprotein particles correlate with the severity of coronary artery disease. Atherosclerosis, 1996; 122: 105-115.

[31] Sacks FM, Alaupovic P, Moye LA, Cole TG, Sussex B, Stampfer MJ, Pfeffer MA, Braunwald E: VLDL, Apolipoproteins B, CIII, and E, and risk of recurrent coronary events in the Cholesterol and Recurrent Events (CARE) Trial. Circulation, 2000; 102: 1886-1892.

[32] Onat A, Hergenç G, Sansoy V, Fobker M, Ceyhan K, Toprak S, Assmann G: Apolipoprotein C-III, a strong discriminant of coronary risk in men and a determinant of the metabolic syndrome in both genders. Atherosclerosis, 2003; 168: 81-89.

[33] Cohn JS, Johnson EJ, Millar JS, Cohn SD, Milne RW, Marcel YL, Russell RM, Schaefer EJ: Contribution of apoB-48 and apoB-100 triglyceride-rich lipoproteins (TRL) to postprandial increases in the plasma concentration of TRL triglycerides and retinyl esters. J Lipid Res, 1993; 34: 2033-2040.

[34] Cohn JS, McNamara JR, Cohn SD, Ordovas JM, E J Schaefer EJ: Plasma apolipoprotein changes in the triglyceride-rich lipoprotein fraction of human subjects fed a fat-rich meal. J Lipid Res, 1988; 29: 925-936.

[35] Karpe F, Bell M, Björkegren J, Hamsten A: Quantification of postprandial triglyceride-rich lipoproteins in healthy men by retinyl ester labeling and simultaneous measurement of apolipoproteins B-48 and B-100. Arterioscler Thromb Vas Biol, 1995; 15: 199–207.

[36] Schneeman BO, Kotite L, Todd KM, Havel RJ: Relationships between the responses of triglyceride-rich lipoproteins in blood plasma containing apolipoproteins B-48 and B-100 to a fat-containing meal in normolipidemic humans. Proc Natl Acad Sci USA, 1993; 90: 2069-2073.

[37] Evans K, Kuusela PJ, Cruz ML, Wilhelmova I, Fielding BA, Frayn KN: Rapid chylomicron appearance following sequential meals: effects of second meal composition. Br J Nutr, 2007; 79: 425-429.

[38] Heath RB, Karpe F, Milne RW, Burdge GC, Wootton SA, Frayn KN: Dietary fatty acids make a rapid and substantial contribution to VLDL-triacylglycerol in the fed state. Am J Physiol Endocrinol Metab, 2007; 292: E732-739.

[39] Martins IJ, Sainsbury AJ, Mamo JCL, Redgrave TG: Lipid and apolipoprotein B48 transport in mesenteric lymph and the effect of hyperphagia on the clearance of chylomicron-like emulsions in insulin-deficient rats. Diabetologia, 1994; 37: 238-246.

[40] Guo Q, Avramoglu RK, Adeli K: Intestinal assembly and secretion of highly dense/lipid-poor apolipoprotein B48-containing lipoprotein particles in the fasting state: Evidence for induction by insulin resistance and exogenous fatty acids. Metabolism, 2005; 54: 689-697.

[41] Hashimoto S, Hayashi S, Yoshida A, Naito M: Acute effects of postprandial aerobic exercise on glucose and lipoprotein metabolism in healthy young women. J Atheroscler Thromb, 2013; 20: 204-213.

[42] Nestel PJ: Relationship between plasma triglycerides and removal of chylomicrons. J Clin Invest, 1964; 43: 943-949.

[43] Grundy SM, Mok HYI: Chylomicron clearance in normal and hyperlipidemic man. Metabolism, 1976; 1225-1239.

[44] Hultin M, Savonen R, Olivecrona T: Chylomicron metabolism in rats: lipolysis, recirculation of triglyceride-derived fatty acids in plasma FFA, and fate of core lipids as analyzed by compartmental modelling. J Lipid Res, 1996; 37: 1022-1036.

[45] Welty FK, Lichtenstein AH, Barrett PHR, Dolnikowski GG, Schaefer EJ: Human apolipoprotein (Apo) B-48 and ApoB-100 kinetics with stable isotopes. Arterioscler Thromb Vasc Biol, 1999; 19: 2966-2974.

[46] Musunuru K, Kathiresan S: Surprises from genetic analyses of lipid risk factors for atherosclerosis. Circ Res, 2016; 118: 579-585.

[47] Griffin BA: Lipoprotein atherogenicity: an overview of current mechanisms. Proc Nutr Soc, 1999; 58: 163-169.

[48] Sattar N, Greer IA, Louden J, Lindsay G, McConnell M, Shepherd J, Packard CJ: Lipoprotein subfraction changes in normal pregnancy: threshold effect of plasma triglyceride on appearance of small, dense low density lipoprotein. J Clin Endocrinol Metab, 1997; 82: 2483-2491.

[49] Millar JS, Packard CJ: Heterogeneity of apolipoprotein B-100-containing lipoproteins: what we have learnt from kinetic studies. Curr Opin Lipidol, 1998; 9: 197-202.

[50] Vaarhorst AM, Beekman M, Suchiman EHD, van Heemst D, Houwing-Duistermaat JJ, Westendorp RGJ, Slagboom PE, Heijmans BT, On behalf of the project group and the Leiden Longevity Study (LLS) Group: Lipid metabolism in long-lived families: the Leiden Longevity Study. Age, 2011; 33: 219-227.

[51] Patsch JR, Karlin JB, Scott LW, Smith LC, Gotto Jr AM: Inverse relationship between blood levels of high density lipoprotein subfraction 2 and magnitude of postprandial lipemia. Proc Natl Acad Sci USA, 1983; 80: 1449-1453.

[52] Després JP, Moorjani S, Lupien PJ, Tremblay A, Nadeau A, Bouchard C: Regional distribution of body fat, plasma lipoproteins, and cardiovascular disease. Arteriosclerosis, 1990; 10: 497-511.

[53] Ryu JE, Craven TE, MacArthur RD, Hinson WH, Bond MG, Hagaman AP, Crouse JR III: Relationship of intraabdominal fat as measured by magnetic resonance imaging to postprandial lipemia in middle-aged subjects. Am J Clin Nutr, 1994; 60: 586-591.

[54] Couillard C, Bergeron N, Prud'homme D, Bergeron J, Tremblay A, Bouchard C, Mauriège, Després JP: Postprandial triglyceride response in visceral obesity in men. Diabetes, 1998; 47: 953-960.

[55] Ginsberg HN, Fishe EA: The ever-expanding role of degradation in the regulation of apolipoprotein B metabolism. J Lipid Res, 2009; 50: S162-S166.

[56] Xiao C, Dash S, Morgantini C, Lewis GF: Novel role of enteral monosaccharides in intestinal lipoprotein production in healthy humans. Arterioscler Thromb Vasc Biol, 2013; 33: 1056-1062.