ORIGINAL

Relationship between serum concentrations of saturated fatty acids and unsaturated fatty acids and the homeostasis model insulin resistance index in Japanese patients with type 2 diabetes mellitus

Masataka Kusunoki[1], Kazuhiko Tsutsumi[2], Meiho Nakayama[1], Tsuyoshi Kurokawa[1], Takao Nakamura[3], Hitoshi Ogawa[1], Yoshitaka Fukuzawa[1], Munehiko Morishita[1], Tatsuro Koide[4], and Tetsuro Miyata[5]

[1]*Aichi Medical University, Medical Clinic, Aichi, Japan ;* [2]*Otsuka Pharmaceutical Factory, Inc., Tokushima, Japan ;* [3]*Faculty of Medicine, Yamagata University, Yamagata, Japan ;* [4]*Aichi Gakuin University, Aichi, Japan ; and* [5]*University of Tokyo School of Medicine, Tokyo, Japan*

Abstract :
Background : Consumption of polyunsaturated fatty acids (PUFA) improves the lipid metabolism of diabetics, leading to prevents of arteriosclerosis. Exact relationship between saturated fatty acids (SFA) or PUFA and the insulin resistance of diabetics are unknown.
Subjects and Methods : We investigated the relationship between the serum concentrations of saturated and unsaturated fatty acids and the homeostasis model insulin resistance index (HOMA-R) in Japanese patients with type 2 diabetes mellitus.
Results : The SFA, i.e., lauric acid, myristic acid, palmitic acid, and stearic acid ; the monounsaturated fatty acids (MUFA), i.e., palmitoleic acid, oleic acid, and erucic acid ; and the PUFA, i.e., eicosadienoic acid, dihomo-γ-linolenic acid, docosatetraenoic acid, and docosapentaenoic acid were positively correlated with HOMA-R. However, no correlations were found between HOMA-R and SFA, i.e., arachidic acid, behenic acid, and lignoceric acid ; the MUFA, i.e., eicosenoic acid and nervonic acid ; and the PUFA, i.e., linoleic acid, γ-linolenic acid, linolenic acid, 5-8-11 eicosatrienoic acid, arachidonic acid, eicosapentaenoic acid, and docosahexaenoic acid.
Conclusions : Some PUFA as well as SFA were positively correlated with HOMA-R. These results indicate that the intake of diet fatty acid must be well balanced in diabetic patients and it is not always true to refrain from taking SFA and increase the unsaturated fatty acids in their diets. J. Med. Invest. 54 : 243-247, August, 2007

Keywords **:** *HOMA-R, PUFA, SFA, Japanese patients, type 2 diabetes mellitus*

INTRODUCTION

There have been many reports that a high-fat diet causes insulin resistance (1-3) and consumption of saturated fatty acids (SFA) induces hyperlipidemia and obesity, causing progression of arterioscle-

Received for publication December 1, 2006 ; accepted March 20, 2007.

Address correspondence and reprint requests to Masataka Kusunoki, MD, Medical Clinic, Aichi Medical University, 2-12-1, Higashisakura, Higashi-ku, Nagoya, Aichi 261-0005, Japan and Fax : +81-052-243-1114.

rosis (4, 5). There have been many reports that consumption of polyunsaturated fatty acids (PUFA) leads to improvement of hyperlipidemia and prevention of arteriosclerosis (6, 7). However, there have been a few reports of an association between blood saturated fatty acid concentrations or unsaturated fatty acid concentrations and insulin resistance. In present study, we measured the serum fatty acid concentrations in patients with type 2 diabetes mellitus and investigated the relations between the homeostasis model insulin resistance index, HOMA-R (8), and the serum fatty acid concentrations. Early morning fasting blood specimens were collected from 93 outpatients (70 men and 23 women) attending the outpatient clinic of Aichi Medical University Hospital, and their serum fatty acid concentrations were measured. Their serum insulin concentration was measured at the same time, and the correlations between serum fatty acid concentrations and HOMA-R were calculated.

SUBJECTS AND METHODS

Study Population

The study was conducted on 93 type 2 diabetes mellitus patients (70 men and 23 women, mean age 54 (SD 11) years old, body mass index 23.3 (SD 4.8)) attending the outpatient clinic for the treatment of diabetes at the Aichi Medical University Hospital. They were treated with hypoglycemic agents (pioglitazone, sulfonylurea, or α-glucosidase inhibitor) and dietetic therapy or both for 2-3 years.

Morning blood samples were collected after a 12-hour fast. The serum was separated from the blood within 1 hour, and serum samples were used to measure the concentrations of glucose, insulin, and fatty acids. All procedures were performed at 4°C or on ice water.

Informed consent was obtained from all the subjects, and the study protocol was approved by the ethics committee.

Serum glucose and insulin analysis

Serum glucose was determined by conventional enzymatic methods at SRL Inc. (Tokyo, Japan). Serum insulin was determined by a conventional enzyme immunoassay with the Glazyme insulin-EIA test (Wako Pure Chemical Industries) at SRL Inc.

Serum fatty acids

Approximately 0.2 ml of serum samples and 2 ml of chloroform-methanol (2 : 1) solution were placed in Pyrex centrifuge tubes, homogenized with a Polytron (PCU-2-110, KINEMATICA GmbH, Switzerland), and then centrifuged at 3000 rpm for 10 min. An aliquot of the chloroform-methanol extract was transferred to another Pyrex tube and dried under a stream of nitrogen gas. The dried specimens were dissolved in 100 μl 0.4M potassium methoxide-methanol/14% boron trifluoride-methanol solution, and the fatty acid concentrations in the solution were measured with a gas chromatograph (Shimazu GC 17A, Kyoto, Japan) at SRL Inc.

Statistical analysis

The correlation analysis was performed by Spearman's test. P values less than 0.05 were considered significant.

RESULTS

The fasting serum glucose levels in the diabetic patients were 135±43 mg/dl, insulin levels were 8.61±8.05 μU/ml, and HOMA-R were 2.91±2.75.

The fasting serum FFA levels are shown in Table 1.

Regarding the fasting serum glucose, insulin, HOMA-R, and FFA levels, no sex difference was noted.

The correlations between serum SFA concentrations and HOMA-R are shown in Table 2. The SFA that were positively correlated with HOMA-R were lauric acid (C12 : 0), myristic acid (C14 : 0), palmitic acid (C16 : 0), and stearic acid (C18 : 0). No correlations were found with regard to arachidic acid (C 20 : 0), behenic acid (C22 : 0), and lignoceric acid (C24 : 0).

The correlations between the serum MUFA concentrations and HOMA-R are shown in Table 3. The MUFA that were positively correlated with HOMA-R were palmitoleic acid (C16 : 1), oleic acid (C18 : 1), and erucic acid (C22 : 1). No correlations were found with regard to eicosanoic acid (C20 : 1) and nervonic acid (C24 : 1).

The correlations between the PUFA serum concentration and HOMA-R are shown in Table 4. The PUFA that were positively correlated with HOMA-R were eicosadienoic acid (C20 : 2), dihomo-γ-linolenic acid (C20 : 3), docosatetraenoic acid (C22 : 4), and docosapentaenoic acid (C22 : 5). No correlations were found with regard to linoleic acid (C18 : 2), γ-linolenic acid (C18 : 3), linolenic acid (C18 : 3), 5-8-11 eicosatrienoic acid (C20 : 3), arachidonic acid

(C20 : 4), eicosapentaenoic acid (C20 : 5), and docosahexaenoic acid (C22 : 6).

Table 1. Fasting serum FFA levels in patients with type 2 diabetic meditus

FFA	μg/ml
Lauric acid (C12 : 0)	1.9±1.4
Myristic acid (C14 : 0)	32.1±18.3
Palmitic acid (C16 : 0)	837.7±245.2
Stearic acid (C18 : 0)	247.5±60.8
Arachidic acid (C20 : 0)	8.3±1.7
Behenic acid (C22 : 0)	21.2±4.6
Lignoceric acid (C24 : 0)	18.8±3.9
Palmitoleic acid (C16 : 1)	79.9±35.4
Oleic acid (C18 : 1)	734.0±247.6
Eicosenoic acid (C20 : 1)	6.3±3.0
Erucic acid (C22 : 1)	1.8±0.8
Nervonic acid (C24 : 1)	39.2±7.9
Linoleic acid (C18 : 2, ω6)	955.9±218.3
γ-linolenic acid (C18 : 3, ω6)	11.3±7.1
Linolenic acid (C18 : 3, ω3)	28.5±14.3
Eicosadienoic acid (C20 : 2, ω6)	6.8±2.0
5-8-11 eicosatrienoic acid (C20 : 3, ω9)	1.9±0.9
Dihomo-γ-kinolenic acid (C20 : 3, ω6)	37.4±13.8
Arachidonic acid (C20 : 4, ω6)	191.3±48.2
Eicosapentaenoic acid (C20 : 5, ω3)	92.5±46.5
Docosatetraenoic acid (C22 : 4, ω6)	4.3±1.5
Docosapentaenoic acid (C22 : 5, ω3)	27.4±9.8
Docosahexaenoic acid (C22 : 6, ω3)	175.3±57.9

Data expressed as means ± S.D. (n = 93).

Table 2. The relationship between the serum levels of saturated fatty acids and the HOMA-R

	Correlation coefficient (r)	P value
Lauric acid (C12 : 0)	0.2169	0.0497
Myristic acid (C14 : 0)	0.3903	0.0001
Palmitic acid (C16 : 0)	0.3523	0.0006
Stearic acid (C18 : 0)	0.2543	0.0144
Arachidic acid (C20 : 0)	0.0438	0.6787
Behenic acid (C22 : 0)	-0.0327	0.7567
Lignoceric acid (C24 : 0)	-0.0589	0.5767

Table 3. The relationship between the serum levels of monounsaturated fatty acids and the HOMA-R

	Correlation coefficient (r)	P value
Palmitoleic acid (C16 : 1)	0.4053	0.0001
Oleic acid (C18 : 1)	0.2890	0.0052
Eicosenoic acid (C20 : 1)	0.1053	0.3177
Erucic acid (C22 : 1)	0.2482	0.0171
Nervonic acid (C24 : 1)	-0.0894	0.3966

Table 4. The relationship between the serum levels of polyunsaturated fatty acids and the HOMA-R

	Correlation coefficient (r)	P value
Linoleic acid (C18 : 2, ω6)	0.1115	0.2901
γ-linolenic acid (C18 : 3, ω6)	0.1321	0.2094
Linolenic acid (C18 : 3, ω3)	0.1769	0.0916
Eicosadienoic acid (C20 : 2, ω6)	0.2570	0.0134
5-8-11 eicosatrienoic acid (C20 : 3, ω9)	0.1428	0.1744
Dihomo-γ-kinolenic acid (C20 : 3, ω6)	0.3568	0.0005
Arachidonic acid (C20 : 4, ω6)	0.1137	0.2804
Eicosapentaenoic acid (C20 : 5, ω3)	0.1740	0.0971
Docosatetraenoic acid (C22 : 4, ω6)	0.2241	0.0318
Docosapentaenoic acid (C22 : 5, ω3)	0.2834	0.0064
Docosahexaenoic acid (C22 : 6, ω3)	0.1877	0.0732

DISCUSSION

Consumption of SFA induces hyperlipidemia, causing progression of arteriosclerosis (4, 5). However, there have been many reports that consumption of PUFA leads to improvement of hyperlipidemia and prevention of arteriosclerosis (6, 7), In human (6, 7) and animal studies, a hyperlipidemia-ameliorating action and cardiovascular disease preventing action have also been found to result from intake of omega-3 fatty acids (9, 10). On the other hand, there have been few reports on the relation between fatty acid intake and glucose metabolism or insulin resistance in diabetics, and there have also been few reports describing an association between blood SFA or unsaturated fatty acid concentrations and insulin resistance.

In an animal study, Storlien LH, *et al.* fed rats saturated fatty acids and reported that when they fed the rats food containing omega-3 fatty acids after inducing insulin resistance, the insulin resistance improved (11). In the present study, we measured the serum fatty acid concentrations of diabetic patients and investigated the relation between an index of insulin resistance, HOMA-R, and serum fatty acid concentrations.

In the present study, we predicted that there would be a positive correlation between serum SFA concentrations and HOMA-R and there would be a negative correlation between the serum concentrations of PUFA and HOMA-R. More specifically, we predicted that the serum concentrations of PUFA in patients with mild insulin resistance would be higher than those of patients with severe insulin resistance and that their serum concentrations of SFA would be low.

The results showed positive correlations between HOMA-R and the concentrations of the SFA, i.e., lauric acid (C12), myristic acid (C14), palmitic acid (C16), and stearic acid (C18), but no correlations with arachidic acid (C20), behenic acid (C22), or lignoceric acid (C24). These results suggest that the concentrations of SFA containing a short carbon atom chain (C12-C18) are more positively correlated with HOMA-R than the concentrations of SFA with a long carbon atom chain (C20-C24), and that the severity of insulin resistance is related to a short carbon chain.

With regard to the MUFA, the concentrations of palmitoleic acid (C16 : 1), oleic acid (C18 : 1), and erucic acid (C22 : 1) were positively correlated with HOMA-R, but no correlations were observed with eicosanoic acid (C20 : 1) or nervonic acid (C24 : 1). No associations were found between the carbon chain length of MUFA and HOMA-R.

Kusunoki, *et al.* administered omega-3 fatty acids to OLETF rats, an animal model of diabetes, and reported that they improved their insulin resistance (12). There are reports on omega-3 fatty acids as representative PUFA, stating that intake of PUFA improves hyperlipidemia in diabetics and prevents the progression of arteriosclerosis (13-15). Therefore, negative correlations were predicted between the concentrations of PUFA and HOMA-R. However, none of the PUFA were found to be negatively correlated with HOMA-R in this study. In contrast, positive correlations were found with eicosadienoic acid (C20 : 2, ω6), dihomo-γ-linolenic acid (C20 : 3, ω6), docosapentaenoic acid (C20 : 5, ω3), and docosatetraenoic acid (C22 : 4, ω6), but no correlations were found between HOMA-R and following seven fatty acids, linoleic acid (C18 : 2, ω6), γ-linolenic acid (C 18 : 3, ω6), linolenic acid (C18 : 3, ω3), 5-8-11 eicosatrienoic acid (C20 : 3, ω9), arachidonic acid (C 20 : 4, ω6), eicosapenataenoic acid (C20 : 5, ω3), or docosahexaenoic acid (C22 : 6, ω3). No correlations were found between the carbon chain length of PUFA and HOMA-R, and no associations were found between HOMA-R and the ω3 fatty acids or ω6 fatty acids.

CONCLUSION

The results of this study showed that some PUFA as well as SFA were positively correlated with HOMA-R, in other words, indicating that intake of those fatty acids aggravates insulin resistance. Among the SFA, positive correlations were found with regard to the concentrations of SFA that have short carbon chains as compared to the concentrations of SFA that have long carbon chains, and there appeared to be an association between the severity of insulin resistance and SFA with short carbon chains. No trends were observed in the correlations between the concentrations of MUFA or PUFA and HOMA-R. These results indicate that the intake of diet fatty acid must be well balanced in diabetic patients and it is not always true that they should refrain from taking SFA and increase the unsaturated fatty acids in their diets.

REFERENCES

1. Storlien LH, James DE, Burleigh KM, Chisholm DJ, Kraegen EW : Fat feeding causes widespread *in vivo* insulin resistance decreased energy expenditure and obesity in rats. Am J Physiol 251 : E576-E583, 1986
2. Okamoto Mi, Okamoto Mo, Kono S, Inoue G, Hayashi T, Kosaki A, Maeda I, Kubota M, Kuzuya H, Imura H : Effects of a high-fat diet on insulin receptor kinase and the glucose transporter in rats. J Nutr BioChem 3 : 241-250, 1992
3. Kusunoki M, Hara T, Tsutsumi K, Nakmura T, Miyata T, Sakakibara F, Sakamoto S, Ogawa H, Nayaya Y, Storlien LH : The lipoprotein lipase activator, NO-1886, suppresses fat accumulation and insulin resistance in rats fed a high-fat diet. Diabetologia 43 : 875-880, 2000
4. YIN W, Tsutsumi K, Yuan Z, Yang B : Effects of the lipoprotein lipase activator NO-1886 as a suppressor agent of atherosclerosis in aorta of mild diabetic rabbits. Arzneim-Forsch./Drug Res 52 : 610-614, 2002
5. Yin W, Yuan Z, Tsutsumi K, Xie Y, Zhang Q, Wang Z, Fu G, Long G, Yang Y : A lipoprotein lipase-promoting agent, NO-1886, improves glucose and lipid metabolism in high fat, high sucrose-fed New Zealand white rabbits. Experimental Diab Res 4 : 427-434, 2003
6. Hu FB, Bromner L, Willett WC, Stampfer MJ, Rexrode KM, Albert CM, Hunter D, Manson JE : Fish and omega-3 fatty acid intake and risk of coronary heart disease in women. JAMA 287 : 1815-1821, 2002
7. Siscovick DS, Raghunathan TE, King I, Weinmann S, Wicklund KG, Albright J, Bovjerg V, Arbogast P, Smith H, Kushi LH, Cobb LA,

Copass MK, Pasty BM, Lemaitre R, Retzlaff B, Childs M, Knoop RH : Dietary intake and cell membrane levels of long-chain n-3 polyunsaturated fatty acids and the risk of primary cardiac arrest. JAMA 274 : 1363-1367, 1995
8. Nagasaki S, Iwamoto Y, Ishikawa S, Kuzuya T, Saito T : Efficacy of troglitazone measured by insulin resistance index. Lancet 350 : 184, 1997
9. Weiner BH, Ockene IS, Levine PH, Cuenoud HF, Fisher M, Johnson BF, Daoud AS, Jarmolych J, Hosmer D, Johnson MH, Natale A, Vaudreuil C, Hoogasian JJ. : Inhibition of atherosclerosis by cod-liver oil in a hyperlipidemic swine model. N Engl J Med 315 : 841-846, 1986
10. Reiner G, Skamene E, DeSanctis J, Radzioch D : Dietary n-3 polyunsaturated fatty acids prevent the development of atherosclerotic lesions in mice : modulation of macrophage secretory activities. Arterioscler Thromb 13 : 1515-1524, 1993
11. Storlien LH, Kraegen EW, Chisholm DJ, Ford GL, Bruce DG, Pascoe WS : Fish oil prevents insulin resistance induced by high-fat feeding in rats. Science 237 : 885-888, 1987
12. Kusunoki M, Tsutsumi K, Hara T, Ogawa H, Nakamura T, Miyata T, Sakakibara F, Fukuzawa Y, Suga T, Kato K, Hirooka Y, Nakaya Y : Ethyl icosapentate (omega-3 fatty acid) causes accumulation of lipids in skeletal muscle but suppresses insulin resistance in OLETF rats. Metabolism 52 : 30-34, 2003
13. Hu FB, Cho E, Rexrode KM, Albert CM, Manson JE : Fish and long-chain omega-3 fatty acid intake and risk of coronary heart disease and total mortality in diabetic women. Circulation 107 : 1852-1857, 2003
14. Dewailly E, Blanchet C, Gingras S, Lemieux S, Bruce JH. : Fish consumption and blood lipids in three ethic groups of Quebec (Canada). Lipids 38 : 359-365, 2003
15. Erkkila AT, Lichtenstein AH, Mozaffarian D, Herrington DM : Fish intake is associated with a reduced progression of coronary artery atherosclerosis in postmenopausal women with coronary artery disease. Am J Clin Nutr 80 : 626-632, 2004