Journal of Atherosclerosis and Thrombosis Vol. 11, No. 6

Reviews

Postprandial Hyperlipidemia and Atherosclerosis

Akira Tanaka

Department of Health and Nutrition, College of Human and Environmental Studies, Kanto-gakuin University, Kanagawa, Japan.

The development of the remnant like particle (RLP) method for conveniently measuring serum remnant lipoprotein levels in 1993 promoted much research on atherogenic significance and metabolism of remnant lipoproteins. This research brought about many results as the following. A novel apolipoprotein B48 receptor incorporating remnant lipoproteins into macrophages in arterial wall was discovered and the structure of the gene of the receptor was clarified. The expression of apolipoprotein B100 was recognized in the human small intestine, suggesting that dietary very low density lipoproteins (VLDL) might be synthesized in the human small intestine and converted into VLDL remnants and low density lipoproteins (LDL). It is recognized that the atherosclerotic risk of postprandial hyperlipidemia is derived from an increase of remnant lipoproteins and that measurement of serum RLP levels in postprandial state is more sensitive and necessary for evaluating an atherosclerotic risk because serum RLP levels remain high all day in patients with diabetes mellitus or coronary heart disease. The relation between postprandial hyperlipidemia and insulin resistance was clarified. *J Atheroscler Thromb, 2004; 11: 322–329.*

Key words: Postprandial hyperlipidemia, Remnant lipoprotein, RLP, Atherosclerosis

Introduction

Since the report of Zilversmit (1) in 1979, postprandial hyperlipidemia has been noticed as a risk factor for atherosclerosis. We have focused on an increase of triglyceride (TG) rich lipoproteins, especially remnant lipoproteins, as a cause of atherosclerotic risk of postprandial hyperlipidemia and researched on the atherogenic significance and metabolism of remnant lipoproteins.

Remnant Lipoproteins and Remnant Like Particles (RLP)

There are two types of remnant lipoproteins, one is chylomicron remnant which is derived from chylomicron synthesized in small intestine, and the other is very low density lipoprotein (VLDL) remnant which is derived from VLDL synthesized in liver. Because of rapid metabolism of remnant lipoproteins, fasting serum of healthy subjects does not include remnant lipoproteins (Fig. 1).

An increase of remnant lipoproteins is a risk factor for atherosclerosis (2). The remnant lipoprotein is as well as the oxidized low density lipoprotein (LDL), easily taken into the macrophage in the arterial wall, promoting foam cell formation of macrophages and forming the atherosclerotic lesion (Fig. 2). However, the remnant lipoprotein was not clinically familiar, since there was no method for conveniently measuring the remnant lipoprotein. In 1993, we developed the remnant like particle (RLP) method for measuring serum remnant lipoproteins in clinical samples (3–5). As a result, clinical research revealed that RLP was easily taken into macrophages in arterial wall as well as oxidized LDL (6), promoted platelet aggregation (7), increased PAI-1 activity (8), impaired endothelial function (9), promoted proliferation of vascular smooth muscle cells (10, 11), and promoted adhesion of monocytes to endothelial cells (12), showing atherogenic

Address for correspondence: Akira Tanaka, Department of Health and Nutrition, College of Human and Environmental Studies, Kanto-gakuin University, 1–50–1, Mutsuura-higasi, Kanazawa-ku, Yokohama 236–8501, Japan.
E-mail: tnkakr@kanto-gakuin.ac.jp
Received May 7, 2004.
Accepted for publication July 15, 2004.

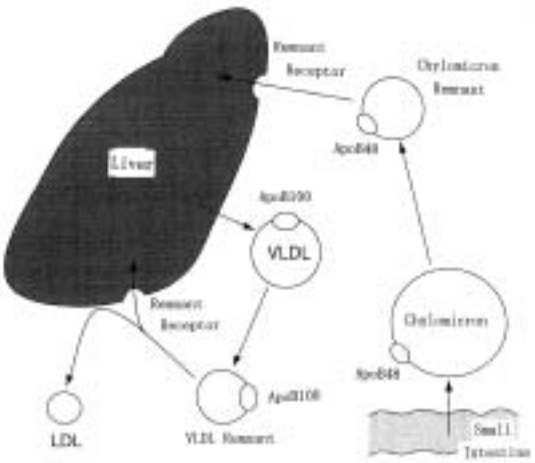

Fig. 1. Metabolism of remnant lipoproteins.

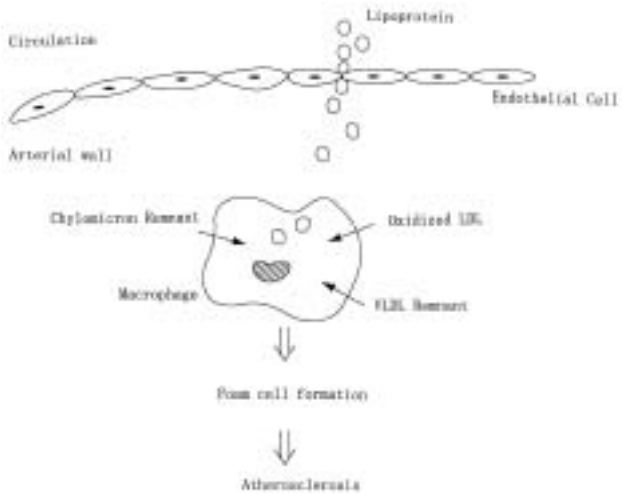

Fig. 2. Incorporation of remnant lipoproteins and oxidized LDL into macrophages in arterial wall.

mechanism of RLP. Recently we clarified that the HMG CoA reductase inhibitor (statin) inhibited adhesion of monocytes to endothelial cells (12).

There are also many clinical reports showing that an increase of RLP is a risk factor for atherosclerosis (13–14). The Framingham Heart Study reported that an increase of RLP cholesterol (RLP-C) is a significant risk factor for coronary artery disease in women (15). And Kugiyama *et al.* (16) reported that the incidence of cardiovascular events in the high RLP-C group (RLP-C ≥ 5.1 mg/dl) was higher than that in the low RLP-C group (RLP-C ≤ 3.3 mg/dl) in a 3-year following clinical study. As the result, Food and Drug Administration (FDA) of the USA recognized that an increase of RLP was a risk factor for coronary heart disease in 2000.

Significance of Postprandial Hyperlipidemia

Study in healthy subjects

Figure 3 shows the results of the fat load test in 19 healthy Subjects (17). Levels of total cholesterol (TC) and high density lipoprotein cholesterol (HDL-C) do not

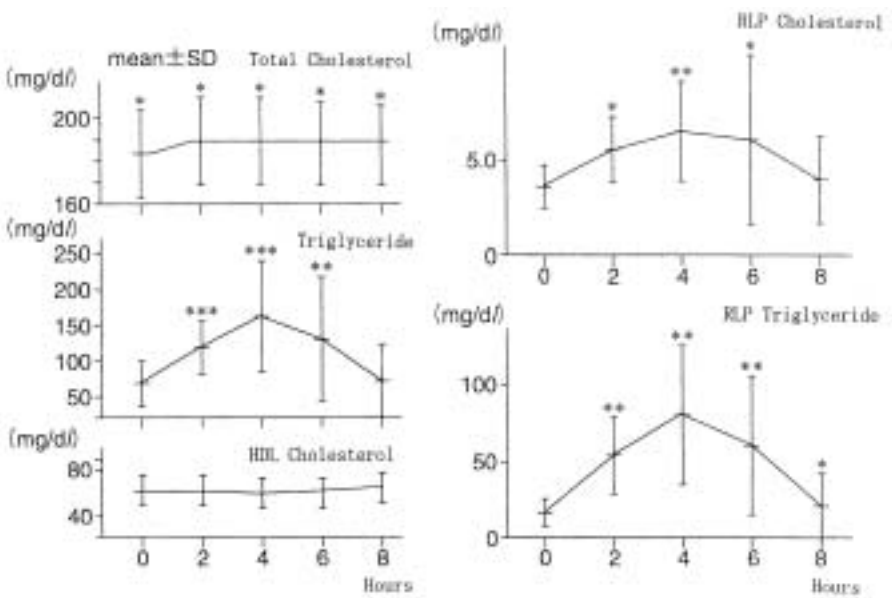

Fig. 3. Changes of lipid levels before and after OFTT cream load.
$*p < 0.05$, $**p < 0.01$, $***p < 0.001$ *vs* values before OFTT cream load

change before and after fat load. However, levels of triglyceride (TG), RLP-C and RLP triglyceride (RLP-TG) increase after fat load, which shows that TG, RLP-C and RLP-TG are useful as the index of postprandial hyperlipidemia. Hypertriglyceridemia derives from an increase of chylomicron, VLDL or remnant lipoprotein. However, remnant lipoprotein is the most important of the three lipoproteins as the risk factor for atherosclerosis. Therefore, RLP is the most important as the index of fat load test or postprandial hyperlipidemia.

Study in patients with coronary artery disease

Figure 4 shows changes of RLP-C levels of healthy subjects and patients with coronary artery disease (CAD) in the fat load test. In healthy subjects, RLP-C levels slightly increase under the cut-off level. However, in patients with CAD, RLP-C levels increase obviously after fat load. And there are many patients with CAD whose RLP-C levels are below the cut-off level before fat load and over the cut-off level after fat load. In such cases, measurement of RLP-C is necessary not only before but also after fat load. Measurement of RLP-C in postprandial state is more sensitive and important for estimating the atherosclerotic risk.

Study in patients with diabetes mellitus

Figure 5 shows daily profile of RLP-C levels in 21 healthy subjects and 28 patients with type 2 diabetes mellitus. RLP-C levels before and 2 hours after breakfast, lunch and dinner, and 23 pm are measured. In healthy subjects, there are small fluctuations of RLP-C levels and short hours in which RLP-C levels exceed the cut-off value. In patients with diabetes mellitus, RLP-C levels keep high values all the day except a few hours before breakfast. This result shows that measurement of RLP-C levels only in fasting state is insufficient in sensitiveness and measurement of RLP-C levels in postprandial state is necessary for evaluating the coronary risk in patients with diabetes mellitus.

Generally, people frequently take snacks between meals; thus they may be in a postprandial state most of

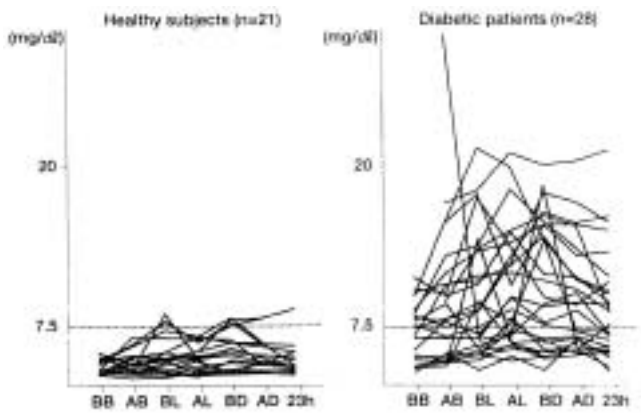

Fig. 5. Daily profile of RLP cholesterol levels in diabetic patients and healthy subjects.
RLP cholesterol levels before and 2 hours after each meal and 23 pm were measured.

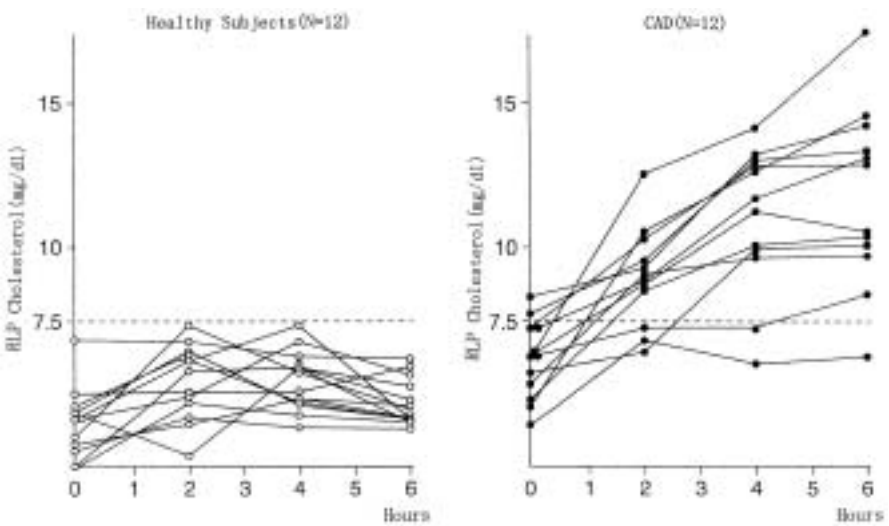

Fig. 4. Changes of RLP cholesterol levels before and after OFTT cream load in patients with coronary artery disease and healthy subjects.
Cut-off value is 7.5 mg/dl.

the day. Figure 3 shows that the RLP values 8 hours after fat load do not return to the RLP values before fat load. This result suggests that the fasting state seems to be only a few hours before breakfast in the day, showing that measurment of RLP is necessary for sensitive evaluation of the coronary risk.

Evaluation of postprandial hyperlipidemia

Fat loading test is useful for evaluation of postprandial hyperlipidemia. However, each researcher uses different type and amount of fat in the fat loading test. Therefore, each result can not be compared. The unification of type and amount of fat is necessary for establishment of the evaluation method for postprandial hyperlipidemia. We are using the OFTT cream (Table 1) developed for the fat load test and obtaining fundamental data of the fat load test using this cream for establishment of the evaluation method for postprandial hyperlipidemia.

The OFTT cream was used in the fat load test of 19 healthy subjects shown in Fig. 3. Results indicated that the more the amount of OFTT cream was, the latter the peak time of RLP values was and the higher the peak value of RLP was (17). The peak of RLP value appeared 4 hours after the load of fat 30 g/body surface (m^2) and 2 hours after the load of fat 17 g/body surface (m^2). Gastrointestinal side effects occurred in the test using much amount of fat. The side effects did not occur in the test using fat under 17 g/ body surface (m^2).

Apolipoprotein B48 and retinyl palmitate besides RLP may be considered as an index for evaluating postprandial hyperlipidemia. However, the method for measuring apolipoprotein B48 values has not been established yet and the reports regarding apolipoprotein B48 are few. And we found that the peak of retinyl palmitate concentration lagged behind the peaks of the RLP and TG concentrations in the study shown in Fig. 3 (17), indicating that retinyl palmitate may have different kinetics to RLP and TG. Accordingly, the retinyl palmitate level may not be a suitable index for evaluating postprandial hyperlipidemia.

Table 1. Composition of OFTT cream.

Water	56.9%
Lipid	32.9%
Protein	2.5%
Carbohydrate	7.4%
Composition of fatty acid	
Saturated fatty acid	64.3%
Mono unsaturated fatty acid	29.3%
Poly unsaturated fatty acid	3.5%
	(Weight %)

Coronary artery disease and postprandial hyperlipidemia

The OFTT cream load test was used in patients with coronary artery disease. We compared RLP-C and RLP-TG values before and after OFTT cream load (fat 30 g/m^2 body surface) between 33 patients with coronary artery disease and 24 normal subjects diagnosed by coronary angiography matching age, body mass index (BMI)and fasting values of TC, TG, HDL-C, RLP-C, RLP-TG and LDL-C in both. There were not significant differences in fasting values of RLP-C and RLP-TG between the two groups. However, RLP-C and RLP-TG values 2 and 4 hours after OFTT cream load were higher in patients with coronary artery disease than those in normal subjects (Fig. 6). This result suggests that the increase of RLP values in postprandial state is one of coronary risk factors. The results agreed with the report by Ikewaki (18).

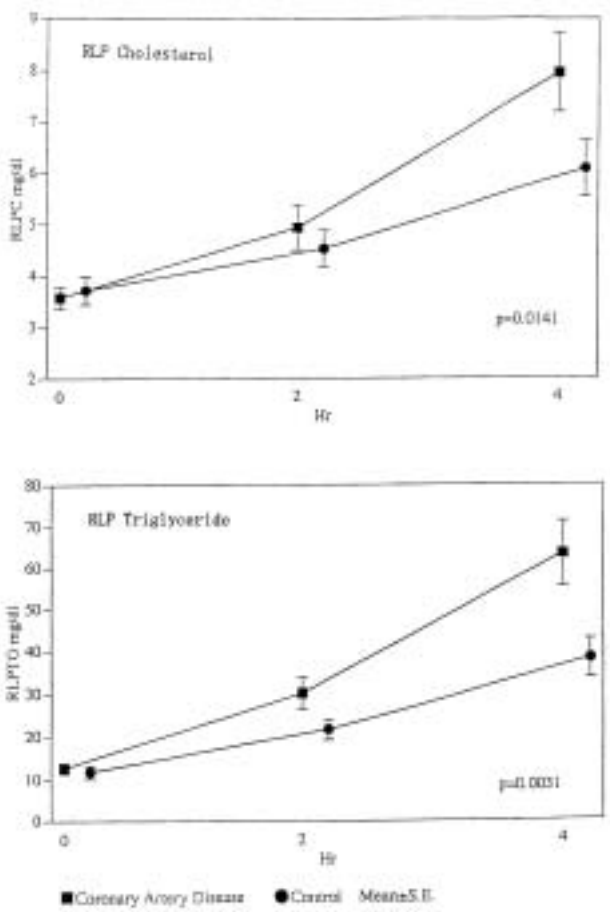

Fig. 6. Changes of RLP cholesterol and RLP triglyceride levels before and after OFTT cream load in 33 patients with coronary angiographically defined coronary artery disease and 24 controls.

Insulin resistance and postprandial hyperlipidemia

We showed the increase of RLP-C and RLP-TG values of diabetic patients with insulin resistance in a postprandial state by using the OFTT cream load test (19). Of 66 newly diagnosed type 2 diabetic patients by 75 g glucose tolerance test, 15 diabetic patients with 120 min insulin value over 80 μU/ml and ΣRI value (sum of 0, 30, 60, 90, 120, and 180 min insulin values) over 300 μU/ml were diagnosed as diabetes mellitus with insulin resistance, and 15 diabetic patients with 120 min insulin value under 60 μU/ml and ΣRI value under 250 μU/ml were diagnosed as diabetes mellitus without insulin resistance. Age, sex, and BMI between both groups were matched. The fat load test in both the diabetic groups and healthy group was undergone within 30 days after 75 g glucose tolerance test. Fat 17 g/m^2 body surface using the OFTT cream was loaded and values of RLP-C, RLP-TG and other lipids were measured before, and 2 and 4 hours after fat load. Though TC, HDL-C, glucose and insulin values did not change after fat load, TG, RLP-C and RLP-TG values obviously increased after fat load (Fig. 7). TG, RLP values peaked at 4 hours after fat load in both the diabetic groups, but peaked at 2 hours and decreased at 4 hours after fat load in the healthy group. And the sum of the amount of increase of 2 and 4 hour values was significantly higher in the diabetes group with insulin resistance than that in the diabetic group without insulin resistance. Reaven GM (20) showed an increase of postprandial RLP-C values even in healthy subjects with insulin resistance.

In diabetic patients with insulin resistance and patients with coronary artery disease, an increase of postprandial RLP values occurs and becomes a coronary risk factor.

Production of Remnant Lipoproteins

There are two kinds of remnant lipoproteins, one is chylomicron remnant converted from chylomicron, the other is VLDL remant converted from VLDL. Chylomicron is synthesized in the small intestine and VLDL is synthesized in the liver. In humans, apolipoprotein B100 is synthesized in the liver and is a principal component of VLDL, VLDL remnant and LDL, by which endogenously synthesized lipid is transported. Apolipoprotein B48 is synthesized in the small intestine and is a principal component of chylomicron and chylomicron remnant, by which dietary lipid is transported. We performed immunohistochemical staining using anti-apolipoprotein B100 monoclonal antibody, which reacts only with apolipoprotein B100 (21). Jejunal samples stained positive and granular staining was noted in the supranuclear region of epithelial cells. However, samples of esophagus, stomach and colon stained negative. We also identified apolipoprotein B100 expression in the epithelial cells of jejunum by immunoblotting and dot blotting of RCR-amplified cDNA (22). The results indicate that not only apolipoprotein B48, but also apolipoprotein B100 are expressed in the human small intestine. The expression of apolipoprotein B100 suggests that dietary VLDL may be synthesized in the human small intestine and converted into LDL, which might play an important role in atherosclerosis (Fig. 8).

Atherosclerosis Developing Mechanism of TG Rich Lipoproteins

It is reported that TG rich lipoproteins including remnant lipoproteins are easily taken into macrophages in the arterial wall and develops foam cell formation of mac-

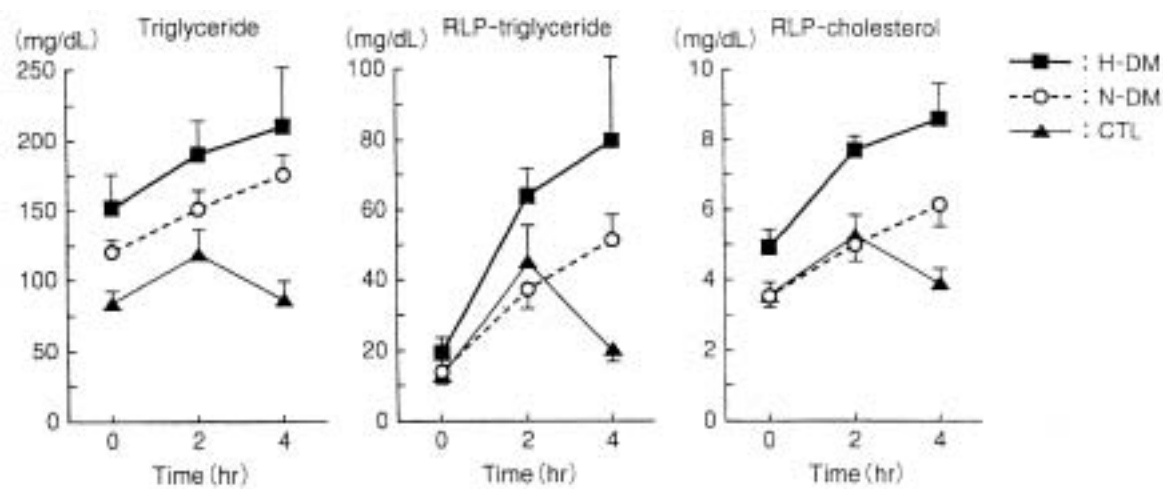

Fig. 7. Changes of triglyceride, RLP triglyceride and RLP cholesterol levels before and after OFTT cream load in type 2 diabetic patients with or without hyperinsulinemia and healthy controls.

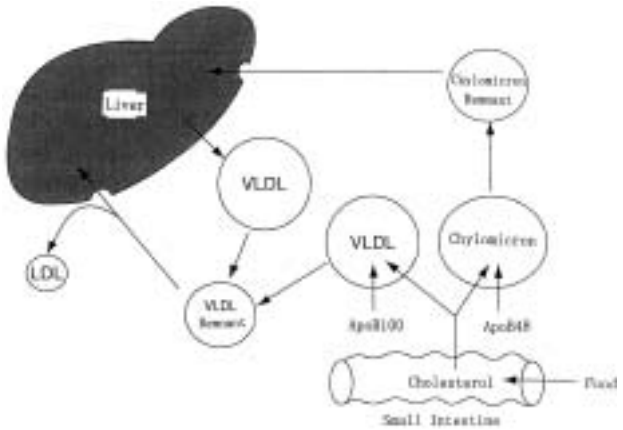

Fig. 8. Expression of apolipoprotein B100 in small Intestine and metabolism of VLDL.

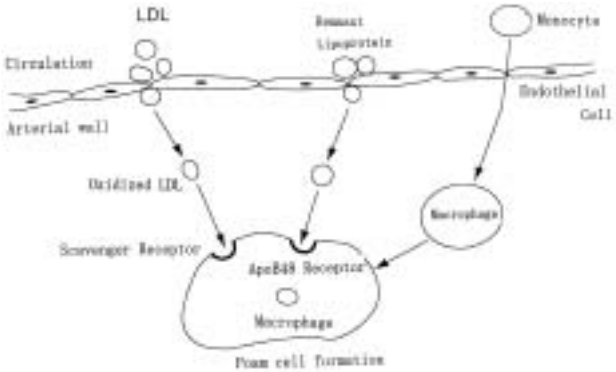

Fig. 9. Scavenger receptor and apolipoprotein B48 receptor of macrophages and athrosclerosis.

rophages and makes early atherosclerotic lesions. However, the mechanism has been unknown. We discovered a novel receptor: the apolipoprotein B48 receptor, which incorporated TG rich lipoproteins into macrophages derived from monocytes by recognizing the apolipoprotein B48 of TG rich lipoproteins, cooperating with Alabama University and Tsukuba University and clarified the cDNA structure of the receptor (23). (Fig. 9) The apolipoprotein B48 receptor was recognized to be the new receptor, which is different from the scavenger receptor incorporating modified LDL into macrophages and the LDL receptor.

It is also clarified that the gene of the apolipoprotein B48 receptor is composed of three introns and four exons and exists in the short arm of the 16th chromosome (24). The cDNA of the receptor consists of 1744 bp and the protein consists of 1188 amino acids (23). The expression of the apolipoprotein B48 receptor was not recognized in skeletal muscle but recognized in bone marrow, reticulo-endothelial system (spleen, lymph, appendix vermiformis and tonsilla), brain, heart, kidney, liver, lung, pancreas and placenta (23). The expression in placenta and lung was strong, the expression in heart and brain was weak.

Significance of the apolipoprotein B48 receptor in atherosclerosis

The distribution of the apolipoprotein B48 receptor suggests that the role of the receptor is to provide dietary lipids and vitamins for monocytes in circulation, reticuloendothelial system, placenta and bone marrow. However, in the states in which TG rich lipoproteins such as remnant lipoproteins increase, the apolipoprotein B48 receptor takes TG rich lipoproteins into macrophages and develops foam cell formation of macrophages and makes the early atherosclerotic lesion. We performed immunohistochemical staining using the anti-apolipoprotein B48 receptor antibody and recognized positive staining in the atherosclerotic lesions of cervical artery, aorta and coronary artery where macrophages existed (23).

Treatment for Increase of Remnant Lipoproteins

An increase of remnant lipoproteins is a risk factor for atherosclerosis. Diet, exercise and drugs are very useful for the increase of remnant lipoproteins. Fibrates are the first choice for the increase of remnant lipoproteins and decrease RLP-C and RLP-TG values by over 50% (25).

Recently we recognized that fibrates and thiazolidinediones such as troglitazone and pioglitazone inhibited the expression of mRNA and protein of the apolipoprotein B48 receptor (26). This result indicates that incorporation of TG rich lipoproteins including remnant lipoproteins into macrophages is inhibited by these drugs, which shows supression of atherosclerosis. Incorporation of TG rich lipoproteins into macrophages (THP-1 cells) is actually recognized.

An increase of remnant lipoproteins is easy to treatment. Therefore, it is very important to diagnose it early and treat it completely.

References

(1) Zilversmit DB: Atherosclerosis:a postprandial phenomenon. Circulation, 60: 473–485,1979

(2) Kane JP, Chen GC, Hamilton RL, Hardman DA, Malloy MJ, and Havel RJ: Remnants of lipoproteins of intestinal and hepatic origin in familial dysbetalipoproteinemia. Arteriosclerosis, 3: 47–56,

1983
(3) Compos E, Nakajima K, Tanaka A, and Havel RJ: Properties of apolipoprotein E-riched fraction of triglyceride-rich lipoproteins isolated from human blood plasma with a monoclonal antibody to apolipoprotein B-100. J Lipid Res, 33: 369–380, 1992
(4) Nakajima K, Okazaki M, Tanaka A, Pullinger CR, Wang T, Nakano T, Adachi M, and Havel RJ: Separation and determination of remnant-like particles in human serum using monoclonal antibodies to apo B100 and apoA-I. J Clin Ligand Assay, 19: 177–183, 1996
(5) Nakajima K, Saito T, Tamura A, Suzuki M, Nakano T, Adachi M, Tanaka A, Tada N, Nakamura H, and Murase T: A new approach for the detection of type III hyperlipoproteinemia by RLP-cholesterol assay. J Atheroscler Tromb, 1: 30–36, 1994
(6) Tomono S, kawazu S, Kato N, Ono T, Ishii C, Ito Y, Shimizu M, Shimoyama M, Nakano T, and Nakajima K: Uptake of remnant like particles (RLP) from diabetic patients from mouse peritoneal macrophages. J Atheroscler Tromb, 1: 98–102, 1994
(7) Saniabadi AR, Umemura K, and Shinoyama M: Aggregation of human blood platelets by remnant like lipoprotein particles of plasma chylomicrons and very low density lipoproteins. Tromb Haemost, 77: 996–1001, 1997
(8) Sakata K, Miho N, Ohtani S, Shirotani M, Yoshida H, and Takada A: Remnant like particle cholesterol in coronary artery disease: correlation with plasminogen activator inhibitor-1 activity. Fibrinolysis & Proteolysis, 12: 123–127, 1998
(9) Kugiyama K, Doi H, Motoyama T, Soejima H, Misumi K, Kawano H, Nakagawa O, Yoshimura M, Ogawa H, Matsumura T, Sugiyama S, Nakajima K, and Yasue H: Association of remnant lipoprotein levels with impairment of endothelium-dependent vasomotor function in human coronary arteries. Circulation, 97: 2519–2526, 1998
(10) Kawakami A, Tanaka A, Nakano T, Abby Saniabadi A, and Numano F: Stimulation of Arterial Smooth Muscle Cell Proliferation by Remnant Lipoprotein Particles isolated by Immunoaffinity Chromatography with Anti-apo A-I and Anti-apo B-100. Horm Metab Res, 33: 67–72, 2001
(11) Kawakami A, Tanaka A, Chiba T, Nakajima K, Shimokado K, and Yoshida M: Remnant lipoprotein-induced smooth muscle cell proliferation involves EG receptor transactivation. Circulation, 108: 2679–2688, 2003
(12) Kawakami A, Tanaka A, Nakajima K, Shimokado K, and Yoshida M: Atorvastatin attenuates remnant lipoprotein-induced monocyte adhesion to vascular endothelium under flow conditions. Circ Res, 91: 263–271, 2002
(13) Devaraj S, Vega G, Lange R, Grundy SM, and Jialal I: Remnant-like particle cholesterol levels in Patients with dysbetal ipoproteinemia or coronary artery disease. Am J Med, 104: 445–450, 1998
(14) Takeichi S, Yukawa N, Nakajima Y, Osawa M, Saito T, Seto Y, Nakano T, Saniabadi AR, Adachi M, Wang T, and Nakajima K: Association of plasma triglyceride-rich lipoprotein remnants with coronary atherosclerosis in cases of sudden cardiac death. Atherosclerosis, 142: 309–315, 1998
(15) McNamara JR, Shah PK, Nakajima K, Cupples LA, Wilson PW, Ordovas JM, and Schaefer EJ: Remnant-like particle (RLP) cholesterol is an independent cardiovascular disease risk factor in women: results from the Framingham Heart Stydy. Atherosclerosis, 154: 229–236, 2001
(16) Kugiyama K, Doi H, Takazoe K, Kawano H, Soejima H, Mizuno Y, Tsunoda R, Sakamoto T, Nakano T, Nakajima K, Ogawa S, Yoshimura M, and Yasue H: Remnant lipoprotein levels in fasting serum predict coronary events in patients with coronary artery disease. Circulation, 99: 2858–2860, 1999
(17) Tanaka A, Tomie N, Nakano T, Nakajima k, Yui K, Tamura M, and Numano F: Measurement of postprandial remnant-like particles (RLP) following a fat-loading test. Clin Chim Acta, 275: 43–52, 1998
(18) Ikewaki K: Postprandial lipemia in Patients with Coronary artery disease. J Jpn Atheroscler Soc, 18: 887–895, 1990 (in Japanese)
(19) Ai M, Tanaka A, Ogita K, Sekine M, Numano F, Numano F, and Reaven GM: Relationship between insulin concentration and plasma remnant lipoprotein response to an oral fat load in patients with type 2 diabetes. J Am Coll Cardiol, 38: 1628–1632, 2001
(20) Kim HS, Abbasi F, Lamendola C, McLaughlin T, and Reaven GM: Effect of insulin resisitance on postprandial elevations of remnant lipoprotein concentrations in postmenopausal women. Am J Clin Nutr, 74: 592–595, 2001
(21) Tamura M, Tanaka A, Yui K, Numano F, and Nakajima K: Apo B-100 existence in human intestinal epithelial cells. Ann NY Acad of Sci, 811: 488–492,1997
(22) Tamura M, Tanaka A, Kobayashi Y, Nihei Z, and Numano F: Expression of apolipoprotein B-100 in isolated human small intestinal epithelium. Horm Metab Res, 32: 343–349, 2000
(23) Brown ML, Ramprasad MP, Umeda PK, Tanaka A, Kobayashi Y, Watanabe T, Shimoyamada H, Pinkel D, Wu W Iin, Li R, Song R, Bradley WA, and Gianturco SH: A macrophage receptor for apolipoprotein B-48: Cloning, Expression and Atherosclerosis. Proc Natl Acad Sci U S A, 97: 7488–

7493, 2000
(24) Brown ML, Yui K, Smith JD, LeBoeuf RC, Weng W, Umeda PK, Li R, Song R, Gianturco SH, and Bradley WA: The murine macrophage apoB-48 receptor gene (Apob-48r): homology to the human receptor.J Lipid Res, 43: 1181–1191, 2002
(25) Tanaka A, Fujinuma Y, Numano F, Nakajima K, Saito T, and Nakano T: Remnant like particles decrease by bezafibrate administration in patients with hypertriglyceridemia. Diagnosis and New Drugs, 32: 179–183, 1995 (in Japanese)
(26) Haraguchi G, Kobayashi Y, Brown LB, Tanaka A, Isobe M, Gianturco SH, and Bradley WA: PPARα and PPARγ Activators Suppress the Monocyte-Macrophage Apolipoprotein B48 Receptor. J Lipid Res, 44: 1224–1231, 2003