The lipid and lipoprotein profiles of apo B-carrying particles in remnant-like lipoproteins isolated from plasma in sudden cardiac death cases

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Objective: Remnant-like lipoprotein particles (RLP) are known as a subset of TG-rich lipoproteins which possess proatherogenic properties. In this study we have elucidated the characteristics of apoB carrying lipoprotein particles in RLP isolated from plasma in sudden cardiac death (SCD) cases and in coronary artery disease (CAD) patients.
Methods: Lipids and lipoproteins, especially apoB-48 and apoB-100, were analyzed in plasma and in RLP from SCD and CAD cases with full stomach.
Results: SDS-PAGE and ELISA showed the predominance of apoB-100 in RLP compared with apoB-48. The main peak of RLP fractionated by HPLC and monitored by TC and TG revealed the particle size at VLDL or IDL, similar to those from CAD patients. The main peak of RLP monitored by apoB-48 revealed the particle size smaller than those monitored by apoB-100 in both cases. RLP-TG/total TG ratio was shown to be significantly higher in SCD cases than in control death cases ($P < 0.001$) in spite of the same fatty acid composition in RLP between the cases.
Conclusion: These results suggested that the major subset of apoB particles in RLP isolated from plasma in SCD cases was apoB-100 carrying VLDL remnants, not apoB-48 CM remnants, which was identical with RLP in CAD patients.

Key word: sudden cardiac death (SCD), coronary artery disease (CAD), remnant-like lipoprotein particles (RLP), RLP-cholesterol (RLP-C), RLP-TG, apoB-48, apoB-100

INTRODUCTION

Triglyceride-rich lipoproteins (TRL) in plasma are well known to contain chylomicrons (CM), very low-density lipoproteins (VLDL) and their remnants. CM is secreted from the intestine carrying apoB-48 known as exogeneous lipoproteins, while VLDL is secreted from the liver carrying apoB-100 known as endogeneous lipoproteins. Remnant-like lipoprotein particles (RLP) in plasma of hyperlipidemic patients have been well characterized and are known to contain both apoB-48 (CM remnants) and apoB-100 particles (VLDL remnants) as a subset of TG-rich lipoproteins (TRL) with the characteristics of apoE, apoCs and cholesterylester-rich [1-8]. Many in vitro and in vivo studies have revealed the characteristics of proatherogenic and pro-inflammatory properties of RLP, which are strikingly similar to the characteristics of oxidized LDL [9, 10].

We have investigated the cause of sudden cardiac death (SCD) or "Pokkuri death syndrome" during the last decade and found the strong association between SCD and the high plasma concentration of RLP. During this study we have found that the postmortem alteration of lipoproteins in plasma was significantly little compared with that of other plasma components such as enzymes, etc [11]. In this study we have examined the more details of characteristics of RLP, especially on apoB-48 and apoB-100 carrying lipoprotein particles in SCD cases and compared with RLP isolated from CAD patients.

As highly sensitive apoB-48 ELISA was developed recently by Kinoshita *et al*. [12], we have analyzed the concentration and size of apoB-48 carrying lipoprotein particles in plasma and in RLP from SCD cases, and compared with those of apoB-100 carrying lipoprotein particles using the HPLC separation method [13, 14] in this study.

The ratio of apoB-100/apoB-48 in RLP has been previously reported by Cohn *et al*. [7] and Campos *et al*. [8] by SDS-PAGE method using isolated RLP from living subjects. In this study we have used the highly sensitive ELISA method for both apoB-48 and apoB-100 measurement and tried to identify the major subset of apoB carrying lipoprotein particles in RLP isolated from SCD cases with full stomach (indicative of postprandial state). Because 2/3 of the SCD cases observed at autopsy in our laboratory had full stomach [15, 16] and often concurrent alcohol intake.

The subset of lipoprotein mainly composing RLP in TRL may play a pivotal role for the risk of sudden cardiac death [17]. This is the first report to describe the quantitative analysis of apoB-48 lipoprotein particles in RLP isolated from SCD and CAD patients and compared with apoB-100 carrying lipoprotein particles in RLP. Further, we have compared the RLP-TG/total TG ratio in plasma and the fatty acid composition of

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Table 1 Demographic data of sudden cardiac death (SCD) and control cases with full stomach.

	Control (n = 54)	SCD (n = 114)	
	mean ± SD	mean ± SD	P value
Age in years	47.3 ± 18.3	49.6 ± 13.5	NS
Male/Female	47/7	93/21	< 0.05
Heart weight (g)	366 ± 87	404 ± 93	< 0.05
Body weight (kg)	60.3 ± 10.3	64.4 ± 12.3	NS
Body height (cm)	164 ± 8.4	164 ± 9.7	NS
BMI	22.2 ± 3.3	23.8 ± 3.4	NS
Postmortem time (h)	8.5 ± 3.2	8.5 ± 3.4	NS
Incidence of severe coronary atherosclerosis (%)	22.2 (12/54)	60.5 (69/114)	< 0.01

RLP between SCD and control death cases to elucidate the biochemical characteristics of RLP in plasma of SCD cases.

MATERIALS AND METHODS

Subjects

The study population comprised 168 sudden death cases, all with full stomach, indicative of postprandial state. They had all died suddenly and unexpectedly between September 1994 and July 2005, were aged 20-69 years old and came from the western part of Kanagawa prefecture in Japan. Most of the subjects had no significant history of medical conditions including cardiac symptoms, and had not taken medications prior to death, based on their medical records. An autopsy was performed on all subjects within 12 hr after death under the protocol of the Department of Forensic Medicine, Tokai University School of Medicine. The autopsy included pathological examination and biological, toxicological, bacteriological and virological testing of body fluids. The study group consisted of subjects with full stomach who had died from SCD (n = 114) and the control death group (n = 54) consisted of subjects with full stomach who had died from non-cardiac causes in the same geographical area during the same period of time. The SCD group was limited to the cases of acute ischemic changes diagnosed by microscopic examination. Subjects with congenital anomalies, acquired valvular deformities, idiopathic cardiomyopathy (dilated cardiomyopathy and hypertrophic cardiomyopathy), infective endo-or myocarditis, alcoholic cardiomyopathy, fibromusclar dysplasia, congestive heart failure, chronic cor pulmonale were excluded from this study. Among the non-cardiac death subjects, most had died in traffic accidents and from suicide. Subjects with fatty liver and renal failures were excluded from both groups because they were known to have high plasma RLP-C and RLP-TG levels [15, 18].

Grading of severity of coronary atherosclerosis was determined according to the postmortem protocol of Department of Forensic Medicine, Tokai University School of Medicine. Briefly, no sclerotic lesion was classified as grade (−), presence of fatty flecks or streaks in any coronary arteries as (±), presence of atheromatous plaque such as focal thickening of lipid nature as (+), moderate atherosclerotic changes with slight or without ulceration or calcification as (2+), presence of extensive atherosclerosis with marked ulceration and/or calcification as (3+), and narrowing of the coronary artery as (4+). The presence of severe coronary atherosclerosis in Table 1 was the cases above the grade (2+). The aliquots of plasma in CAD patients previously reported [11] which had kept frozen at-80. C was used for this study.

This study was conducted under the approval of the Ethical Committee of Tokai University School of Medicine.

Isolation of RLP from plasma

RLP were isolated using an immunoseparation method developed by Nakajima and colleagues [2]. Briefly, 0.2 ml of plasma samples were applied to columns containing 2 ml (1.0 × 2.5 cm) of immunoaffinity mixed gel containing two clones of monoclonal antibodies, Mab JI-H raised against human apoB100 and MabH-12 raised against human apoAI (Japan Immunoresearch Laboratories, Takasaki, Japan). The plasma samples were incubated with immunoaffinity mixed gels at room temperature for 30 min. In this process, more than half of apoB carrying VLDL bound to the gels, but RLP did not bind to the gel because the Mab JI-H binding site (apoB-51) was most probably masked by apoE [5]. Lipoproteins unbound to the gel (containing primarily CM and VLDL remnants) were eluted with 3.5 ml of 10 mM sodium phosphate buffer (pH 7.2). In addition, total VLDL (d < 1.006) was isolated from plasma by ultracentrifugation for 16 hr at a peak density of 1.006 kg/l. RLP used for HPLC analysis was not ultracenrifuged (d < 1.006), but concentrated the immunoaffinity unbound fraction by Amicon filter.

Characterization of RLP isolated from plasma

RLP were characterized by SDS-polyacrylamide gel electrophoresis (SDS-PAGE) [1], and by high performance liquid chromatography (HPLC) [14]. Slab gel electrophoresis was carried out in a 3-15% linear gradient of polyacrylamide in Tris-HCl buffer (pH 6.8). The gel was visualized with silver reagent (Daiichi Chemicals, Tokyo). HPLC analysis was carried out using an HPLC system equipped with gel permeation columns (TSK Lipopropak XL, Toso, Tokyo). RLP was fractionated by HPLC and determined cholesterol (modified Determiner L-TC, Kyowa Medex, Tokyo), TG (modified Determiner L-TG, Kyowa Medex), apoB-48 (Otsuka, Japan) and ApoB-100 (JIMRO, Japan) in RLP.

Table 2 Plasma lipid and lipoprotein levels in cases of sudden cardiac death (SCD) and control death with full stomach.

Gastric contents	Control (n = 54) Presence		SCD (n = 114) Presence		
	median	25-75% tile	median	25-75% tile	P value[a]
Total cholesterol (mg/dl)	173	144-207	218	185-252	< 0.0001
Triglycerides (mg/dl)	113	80-161	169	110-259	< 0.0001
VLDL-C (mg/dl)	20	7.2-36.9	29	17-51	< 0.005
LDL-C (mg/dl)	92	69-122	140	103-175	< 0.0001
HDL-C (mg/dl)	45	28-64	42	33-53	NS
apo B-100 (mg/dl)	66	55-93	88	71-121	< 0.005
apo B-48 (μg/dl)	365	230-818	390	205-675	NS
RLP apo B-100 (μg/dl)	1179	803-1849	1356	877-2489	< 0.001
RLP apo B-100/B48 ratio	5.92	1.57-18.44	7.06	1.97-19.40	NS
RLP-C (mg/dl)	9.9	5.3-19.1	17.6	12.0-25.9	< 0.0001
RLP-TG (mg/dl)	48.8	39.2-80.4	86.5	59.6-150.7	< 0.0001
RLP-TG/RLP-C ratio	5.07	2.55-8.35	4.83	3.91-6.86	NS
RLP-TG/totalTG ratio	0.44	0.36-0.54	0.58	0.50-0.76	< 0.001

Analysis of lipids, lipoproteins and fatty acid compositions in RLP

Blood was removed from the heart and centrifuged to obtain plasma, which was stored at −80℃ and never thawed before subsequent analyse. Plasma TC and TG concentrations were determined enzymatically by Determiner L-TC and Determiner L-TG (Kyowa Medex, Tokyo) on a Toshiba Auto-Analyser TBA-20R (Toshiba, Tokyo). VLDL-C, LDL-C and HDL-C were determined by ultracentrifugation. RLP-C and RLP-TG were determined as described previously [2]. Cholesterol and TG concentrations in the RLP fractions were determined by sensitive enzymatic assays (Determiner L-TC and Determiner L-TG, respectively; Kyowa Medex, Tokyo).

ApoB-100 (JIMRO, Takasaki) and apoB-48 (Otsuka, Tokyo) in RLP fractionated by HPLC were measured by ELISA. Glycerol (highly purified) was purchased from Wako Pure Chemicals (Osaka, Japan). Fatty acid composition was determined by gas chromatography method [19].

Statistical methods

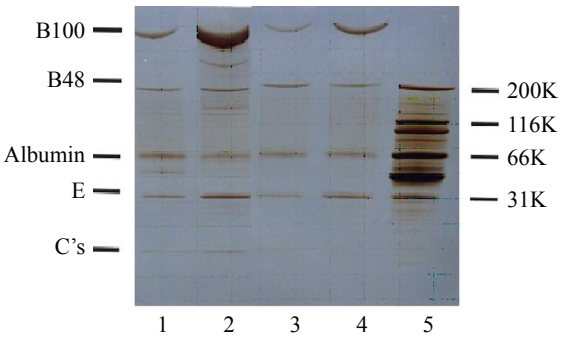
Student's *t*-test, Mann-Whitney U test and the chi-squared test were used to estimate the statistical significance of differences between SCD cases and control death cases. The former was used in variables with Gaussian distribution and the latter was used in variables with non-Gaussian distribution such as plasma RLP-C and RLP-TG concentrations. $P < 0.05$ was considered significant.

RESULTS

Plasma lipid and lipoprotein levels in SCD and control death cases

The demographic characteristics of the 168 study subjects with full stomach were shown in Table 1. The SCD group (n = 114) and control death group (n = 54) had a similar post-mortem interval, age, body height, weight and BMI. Heart weight was statistically different between the two groups ($P < 0.05$). Significant difference was observed between the two groups in the incidence of severe coronary atherosclerosis ($P < 0.01$).

The median concentrations of plasma TC ($P < 0.001$), TG ($P < 0.001$), VLDL-C ($P < 0.005$), LDL-C ($P < 0.0001$), apoB ($P < 0.0001$), RLP-C ($P < 0.001$) and RLP-TG ($P < 0.001$) were significantly elevated in SCD compared with control death cases (Table 2). RLPapoB-100 was significantly elevated in SCD cases ($P < 0.001$), while RLPapoB-48 (identical to plasma apoB-48, because all apoB-48 were measured in RLP [2]) was not different between the two groups.



Lane 1: RLP (d<1.006) (No.1)
Lane 2: Total VLDL (No.1)
Lane 3: RLP (d<1.006) (No.2)
Lane 4: Total VLDL (No.2)
Lane 5: Molecular Standards

Fig. 1 Characterization of apolipoproteins in RLP (d < 1.006) and total VLDL isolated from post-mortem plasma in two SCD cases by SDS-PAGE. RLP (lanes 1 and 3) isolated from the plasma of two SCD cases within 12 hr after death were found to be rich in apo E, Apo B-100 and apo B-48, detected at similar mobility to RLP (d < 1.006) isolated from living patients. ApoB-100 was shown to be significantly greater than apoB-48 in RLP.

Further analysis of RLP was performed using plasma with typical lipid and lipoprotein profiles of SCD cases [11] randomly selected from this study group.

Characteristics of RLP analyzed by SDS-PAGE and HPLC

RLP isolated from plasma of two SCD cases by the immunoaffinity mixed gel method followed by ultracentrifugation for 16 hr (d = 1.006 kg/l) was

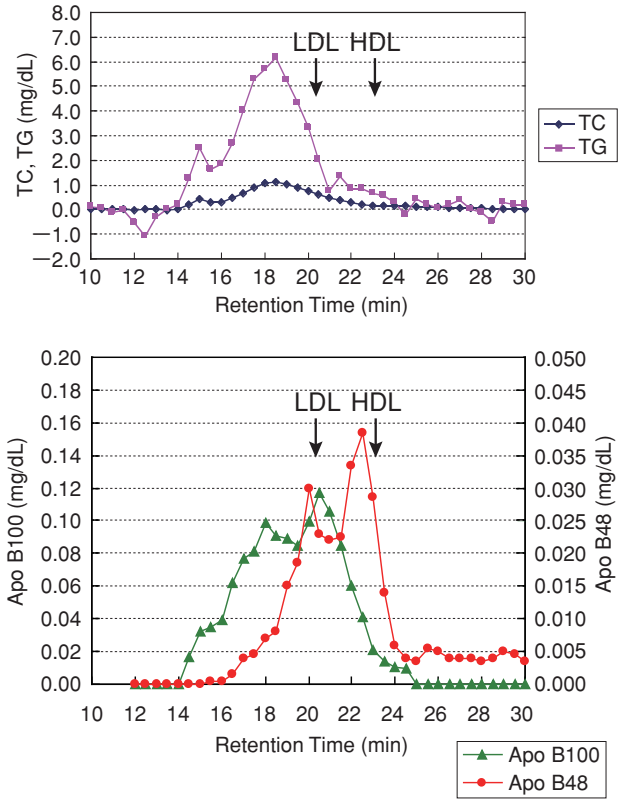


Fig. 2 (a) HPLC profiles of RLP isolated from plasma from a CAD patient. (A) HPLC profile of RLP was monitored by measuring both total cholesterol (TC) and triglycerides (TG) (top) and monitored by apoB-48 and apoB-100 (bottom). This is a typical profile of CAD patient monitored by lipids and lipoproteins. The major particles in RLP were VLDL size, monitored by lipids, but smaller particles were detected by apoB-48 rather than those by apoB-100.

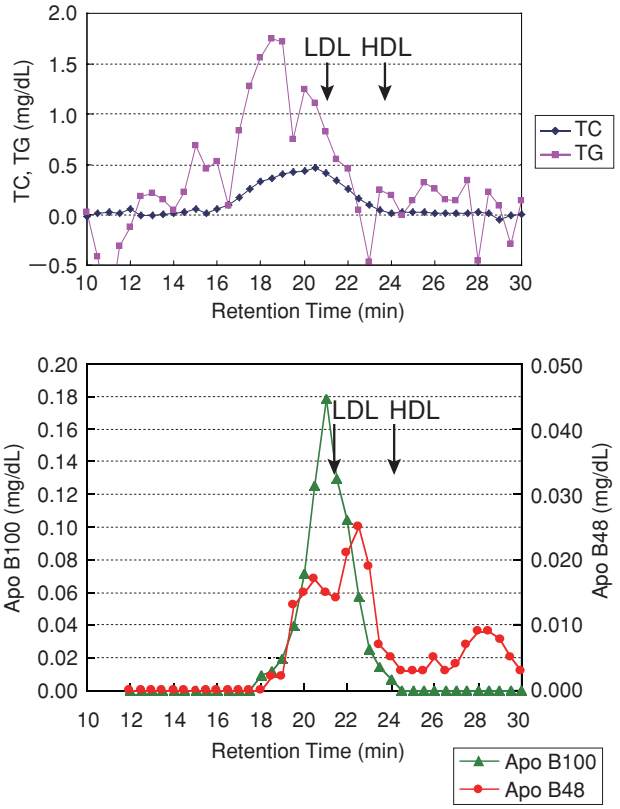


Fig. 2 (b) HPLC profile of RLP isolated from the plasma of a SCD case. HPLC profile of RLP was monitored by measuring both total cholesterol (TC) and triglycerides (TG) (top) and monitored by apoB-48 and apoB-100 (bottom). This is a typical profile of SCD case monitored by lipid and lipoprotein. The major particle size in RLP was VLDL size, monitored by lipids, but smaller particles were observed when monitored by apoB-48 rather than those by apoB-100, as shown in Fig. 2 (a).

shown to be rich in apo E and contained less apoB48 than apoB100 (Fig. 1; lanes 1 and 3), indicating a very similar electropheretic SDS-PAGE profile to the RLP profile in CAD patients previously reported [1, 2].

Figure 2 showed typical lipid and lipoprotein profiles of RLP in CAD and SCD cases. RLP (SCD #8983) fractionated by HPLC and monitored by TC, TG, apoB-48 and apoB-100 showed lipoproteins mainly with particle sizes in the range of VLDL to LDL, very similar profile to RLP in CAD patient (CAD #4) (Fig. 2(a) and (b)) [1, 2]. The main peak of RLP fractionated by HPLC and monitored by TC and TG revealed the particle size at VLDL to LDL, similar to those from CAD patients. The main peak of RLP monitored by apoB-100 and apoB-48 revealed the smaller particle size than VLDL to LDL.

Figure 3 showed the ratio of RLP-TG/total TG in SCD and control death plasma. The RLP-TG/TG ratio was significantly higher in SCD than in control death case, although the controls were heavily depend upon 3 outliners. The median RLP-TG/total TG ratio was 0.44 in controls and 0.58 in SCD ($P < 0.001$) (Table 2), indicating the significantly higher ratio of RLP-TG in total TG as shown in Fig. 3. This indicated the possibility that RLP-TG in SCD was more resistant to lipolysis than RLP-TG in control death cases.

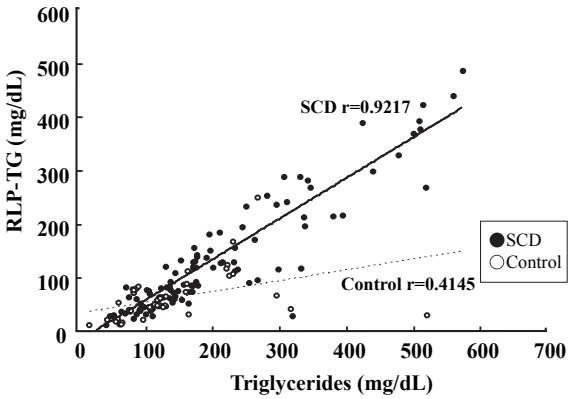


Fig 3. Plasma RLP-TG/total TG ratios in SCD (closed circle) and control death (open circle) cases. The ratio was significantly high in SCD cases compared with control death cases. But three cases of control death heavily affected on lowering the slop of control group, but these cases were sometimes observed in postmortem plasma. Table 2 indicated the median RLP-TG/total TG ratio which showed parallel with Fig. 3.

Table 3 Free fatty acid composition in RLP isolated from plasma of sudden cardiac death (SCD) and control death cases.

	Control (n = 12)		SCD (n = 34)		
	median	25-75% tile	median	25-75% tile	P value[a]
EPA/AA ratio	0.38	0.19-0.80	0.37	0.23-0.56	NS
Lauric acid (weight%) C12: 0	0.28	0.24-0.43	0.27	0.21-0.36	NS
Myristic acid (weight%) C14: 0	1.70	1.53-2.27	1.87	1.53-2.09	NS
Myristoleic acid (weight%) C14: 1ω5	0.0	0.0-0.0	0.0	0.0-0.0	NS
Palmitic acid (weight%) C16: 0	27.9	25.8-29.7	27.9	26.7-29.8	NS
Palmitoleic acid (weight%) C16: 1ω7	2.66	2.29-3.13	2.47	2.07-3.33	NS
Stearic acid (weight%) C18: 0	9.0	7.3-10.0	8.7	7.7-10.0	NS
Oleic acid (weight%) C18: 1ω9	25.3	24.2-27.1	26.2	22.3-29.1	NS
Linoleic acid (weight%) C18: 2ω6	20.0	18-23	20.7	17.8-22.8	NS
γ-linolenic acid (weight%) C18: 3ω6	0.0	0.0-0.2	0.11	0.0-0.24	NS
Linolenic acid (weight%) C18: 3ω3	1.32	1.12-1.78	1.12	0.84-1.62	NS
Arachidic acid (weight%) C20: 0	0.0	0.0-0.25	0.15	0-0.28	NS
Eicosenoic acid (weight%) C20: 1ω9	0.0	0.0-0.26	0.08	0.0-0.32	NS
Eicosadienoic acid (weight%) C20: 2ω6	0.0	0.0-0.06	0.0	0.0-0.22	NS
5-8-11 Eicosatrienoic acid (weight%) C20: 3ω9	0.0	0.0-0.0	0.0	0.0-0.0	NS
Dihomo-γ-Linolenic Acid (weight%) C20: 3ω6	0.45	0.0-0.57	0.53	0.39-0.61	NS
Archidonic acid (weight%) C20: 4ω6	3.1	2.5-4.1	2.7	2.2-4.1	NS
Eicosapentaenoic acid (weight%) C20: 5ω3	1.5	0.6-2.4	1.1	0.67-1.6	NS
Behenic acid (weight%) C22: 0	0.0	0.0-0.55	0.48	0.27-0.71	<0.05
Erudic acid (weight%) C22: 1ω9	0.0	0.0-0.0	0.0	0.0-0.0	NS
Docosatetraenoic acid (weight%) C22: 4ω6	0.0	0.0-0.0	0.0	0.0-0.0	NS
Docosapentaenoic acid (weight%) C22: 5ω3	0.0	0.0-0.64	0.32	0.0-0.51	NS
Lignoceric acid (weight%) C24: 0	0.0	0.0-0.37	0.21	0-0.44	NS
Docosahexaenoic acid (weight%) C22: 6ω3	2.7	2.0-4.8	2.9	2.3-4.0	NS
Nervonic acid (weight%) C24: 1ω9	0.22	0.0-0.64	0.37	0.0-0.54	NS

Fatty acid composition in RLP

Fatty acids in plasma mainly originate from triglyceride-rich lipoproteins. The composition of fatty acids in RLP was analyzed and no significant differences were found between RLP isolated from the plasma of SCD and control death cases. They are rich in palmitic acid, oleic acid and linoleic acid. The fatty acid composition characteristics of RLP were shown in Table 3. No significant difference was observed in eicosapentaenoic acid/arachidonic acid ratio (EPA/AA ratio) in SCD and control death cases.

DISCUSSION

The principal objective of this study was to assess the predominance of apoB-100 carrying lipoprotein particles in RLP, not apoB-48 carrying particles in SCD and CAD cases with full stomach, indicative of postprandial state. The major lipoproteins in RLP may play a pivotal role for the risk of coronary artery events as reported by Oi *et al*. [20] from an *in vivo* study. RLP isolated from SCD cases induced severe spasm in healthy pig coronary arteries. Furthermore, RLP isolated from the plasma of SCD cases showed very similar properties to RLP isolated from the plasma of CAD patients, indicating minimal post-mortem alterations in bioactive properties of RLP. Saniabadi *et al*. [21] described the biological activities of RLP isolated from post-mortem plasma and demonstrated that RLP promoted human platelet aggregation in whole blood as did RLP isolated from living subjects [22]. In addition, RLP isolated from post-mortem plasma in SCD cases demonstrated strong inhibition of endothelium-dependent vasorelaxation and NO synthesis in endothelial cells [23] as did RLP isolated from hyperlipidemic patients [10].

There were no apparent discrepancies in our studies between post-mortem plasma levels and clinically observed plasma levels of total cholesterol, TG and cholesterol concentrations in VLDL, LDL and HDL [15, 16, 24] as shown in Table 1. Similar elevated levels of plasma lipids and lipoproteins, including RLP-C and RLP-TG, in SCD cases and in CAD patients were determined in the fasting and postprandial states [16, 25]. The distribution of plasma apoB-48 levels in this study group was consistent with the levels reported by Sakai *et al*. [26] in the Japanese population. HDL-C levels detected in this study were not significantly different between SCD and control death groups, different from the well-established results between CAD patients and healthy controls. Based on our previous observations of patients receiving hemodialysis [18], plasma RLP soon after death appeared to be in similar condition with patients whose lipoprotein lipase activity was elevated by heparin infusion, showing no significant change in RLP-C but significant reduction of RLP-TG levels.

RLP isolated from plasma of SCD cases is shown in Fig. 1 to be rich in apo E and to contain less amount of apoB48 than that of apoB100, indicating a very similar electrophoretic profile to the RLP profile in CAD patients previously reported [1, 2, 11].

HPLC profile of RLP isolated from a typical SCD post-mortem plasma were compared with those from a typical CAD patients. HPLC profile of RLP monitored by TC and TG were very similar at Fig. 2(a) and (b). HPLC profile monitored by apoB-48 and apoB-100

showed also very similar particle size in both CAD and SCD, but RLP isolated from SCD seemed to be slightly smaller size compared with RLP from CAD. As we have experienced the similar trend in SCD cases, this might be in a part associated with inevitable post-mortem alteration in plasma. Smaller particles detected by apoB-48 may be reflecting the presence of apoB-48 particles varying to very small size particles, although the possibility of destruction of chylomicrons during the frozen storage could not be entirely neglected. Recently, apoB-48 particles even smaller than LDL are reported to exist before freezing the plasma [27], supporting the former possibility.

We have previously reported that RLP-TG was the strongest risk factor among the parameters tested in SCD plasma [15, 16], and these results were highly correlated with high plasma TG levels in CAD mortality [28, 29].

Increased TG in the postprandial state was composed of both CM and VLDL remnants [30], but in SCD cases with full stomach, namely reflecting the postprandial state, the significance of CM or CM remnants in RLP as a risk for clinical events were not well studied. Therefore we analyzed the plasma levels of apoB-48 in SCD and control death cases (Table 2). Unexpectedly, apoB-48 levels were not elevated in the plasma of neither SCD nor control death cases compared with the fasting state [31] and no statistical difference between the two groups was observed. RLP-apoB-100 was significantly elevated in the plasma of SCD cases ($P < 0.001$) compared with control death cases, indicating apoB-100 carrying lipoproteins as a major bioactive subset in RLP. More than 7-fold concentration of apoB-100 was detected in SCD when compared with apoB-48 in Table 2. Although apoB-48 particles and apoB-100 particles in RLP have a similar composition and particle structure, the biological properties of RLP may reflect the nature of the major subset [10].

In order to identify other composition of RLP, we analyzed the fatty acid composition in RLP. The fatty acid composition including EPA/AA ratio (known as an atherogenic index) was not different between RLP isolated from SCD and control death cases. As Oi *et al.* [20] reported, the severity of the biological activity (such as spasm) induced by RLP in coronary arteries of healthy pigs was highly correlated with the amount of RLP treated, but might not owe to specific components contained in RLP. So far we have not found any specific difference of bioactive properties between CM and VLDL remnants [10], although RLP-TG/RLP-C ratio seemed to be slightly different between SCD and control death groups. Furthermore, RLP-TG/total TG ratio shown at Fig. 3 was significantly different between the two groups ($P < 0.001$) in spite of the same composition of fatty acids in RLP. Similarly, RLP-TG/total TG ratio has been reported to be significantly elevated in diabetes and CAD cases [13, 32]. These may be strongly associated with suppressed LPL activities on the lipolysis of RLP-TG. The LPL inhibitor such as angiopoetin-like protein 3 [33] may affect the suppression of RLP-TG lipolysis. Further studies are required to clarify this mechanism.

In conclusion, RLP isolated from post-mortem plasma of SCD cases was mainly composed of VLDL remnants and was almost identical with RLP in CAD patient. ApoB-100 carrying VLDL remnants may play a pivotal role as a bioactive subset in RLP, while apoB-48 carrying CM remnants may not even in the postprandial state.

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