Clin Genet 2006: 69: 455–457
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CLINICAL GENETICS
doi: 10.1111/j.1399-0004.2006.00612.x

Letter to the Editor

Cholesteryl ester transfer protein gene mutations in Brazilian hyperalphalipoproteinemia

To the Editor:

Cholesteryl ester transfer protein (CETP) plays a central role in high-density lipoprotein (HDL) metabolism and is a key protein in the reverse cholesterol transport (1, 2). CETP facilitates the transfer of cholesteryl ester from HDL to apolipoprotein B-containing lipoproteins, and its deficiency is associated with hyperalphalipoproteinemia (HALP) (3, 4). Although the inverse association between HDL-cholesterol (HDL-C) concentrations and cardiovascular disease (CVD) is well established (5), the role of CETP in atherosclerosis remains controversial (6–8). Several mutations at the CETP gene locus have been described, which cause depletion of CETP activity and consequently high HDL-C in plasma (4, 9). HALP patients due to plasma CETP deficiency have been reported, mostly from Japan (3, 4, 8, 10), but there are some reports of CETP deficiency from German, Caucasian, and Asian populations (9).

In this study, we investigated the prevalence of the most studied CETP gene mutations (intron 14 splicing defect, Int14A, and exon 15 missense mutation, D442G) in Brazilian HALP subjects (152 HALP and 139 controls, CTL). In addition, we evaluated the impact of each genetic mutation on the degree of carotid atherosclerosis, the concentrations of lipoproteins, the activities of CETP, phospholipid transfer protein (PLTP), and lipases in the plasma.

For the identification of the CETP mutations, the genomic DNAs were extracted from peripheral leukocytes and analyzed by the polymerase chain reaction-restriction fragment length polymorphism method, as described previously (11–13).

The Brazilian population is ethnically diverse, with a predominance of Afro-descendents. The frequency of the Int14A and D442G alleles in the HALP population was 0.023 and 0.0033, respectively. The prevalence of Int14A mutation was 4%, which was lower than that observed in the Japanese HALP population (32%) (14) but higher than that in the North-American HALP (0.7%) (9) and Japanese-American HALP (0.5%) subjects (6). The prevalence of the D442G mutation was far lower (0.7%) than that reported for the HALP (above 22%) and Japanese general population (4.5–7%) (4, 12) and for Japanese-American subjects (5.1%) (6).

Among the six Int14A mutation carriers (Table 1), we found one homozygote, a 61-year-old white woman, born from a non-consanguineous marriage, with family history of coronary artery disease (CAD), but no clinical cardiovascular damage. This is the first description of a homozygote Int14A CETP mutation outside Japan. Among the heterozygotes for Int14A, a 29-year-old male presented corneal arcus with established CAD and a 46-year-old female presented a carotid atheroma with no other manifestations of CVD. Both were whites, with no biochemical characteristics distinct from other mutation carriers. Three individuals had positive family histories of CAD. A 73-year-old male, from Asian origin and heterozygote for the D442G, presented results similar to the CTL group. Besides no personal or family register of CAD, he was the only one who presented increased intima-media thickness (IMT), possibly because he was the oldest.

When we considered all mutation carriers together, higher HDL-C concentration (83%), lipoprotein lipase (LPL, 11%) and PLTP (60%) and lower CETP (36%) and hepatic lipase (HL, 26%) activities were observed. While the D442G carrier presented CETP, LPL, HL, and PLTP activities closer to the values from CTL group, the homozygote for Int14A mutation had an HL activity below the reference interval (2.5 and 97.5 percentiles of CTL), suggesting a double gene defect as described by Hirano et al. (7). The double deficiency of CETP and HL is

Table 1. Clinical and biochemical characteristics of CETP gene mutation carriers and controls

Parameters	Int14A (HO) ($n = 1$)	Int14A (HE)					All Int14A (HE)[a] ($n = 5$)	D442G (HE) ($n = 1$)	All carriers ($n = 7$)	Controls ($n = 81$–139)
Age (years)	61	46	52	68	33	29	46 ± 5	73	52 ± 15	42 ± 15
Sex (F/M)	F	F	F	M	M	M	–	M	–	84/55
Origin	Br*	Br*	Eu	As	Ar	Br*	–	As	–	–
Cholesterol (mmol/l)	9.32	5.44	4.87	6.22	6.16	6.5	5.83 ± 0.65	5.02	6.21 ± 1.50	4.82 ± 1.24
Triglycerides (mmol/l)	2.49	1.06	0.70	0.88	1.72	2.04	1.28 ± 0.56	0.74	1.38 ± 0.70	1.05 ± 0.52
LDL-C (mmol/l)	2.72	3.29	2.36	3.55	3.26	3.26	3.13 ± 0.44	2.72	3.03 ± 0.41	2.98 ± 1.04
HDL-C (mmol/l)	5.46	1.76	2.05	2.25	2.02	2.30	2.05 ± 0.26	1.94	2.51 ± 1.29	1.37 ± 0.26
HDL_2-C (mmol/l)	0.93	0.54	0.63	0.63	0.45	0.45	0.54 ± 0.10	0.36	0.44 ± 0.18	0.31 ± 0.15
HDL_3-C (mmol/l)	4.53	1.50	1.80	1.62	1.58	1.69	1.50 ± 0.13	1.47	2.09 ± 1.19	1.03 ± 0.26
CETP (%)	0	6.6	16.4	13.3	6.2	5.1	10 ± 5	18	9 ± 5	14 ± 8
PLTP (%)	40	22	43	24	13	19	20 ± 15	5	24 ± 14	15 ± 10
LPL (nmol FFA/ml/h)	745	1247	3331	3290	2936	2957	2752 ± 861	3646	2593 ± 112	2344 ± 1,201
HL (nmol FFA/ml/h)	237	1081	1923	1322	2952	2913	2038 ± 872	2278	1815 ± 995	2466 ± 1,470
Carotid IMT (mm)	0.61	0.60	0.76	0.84	0.66	0.56	0.69 ± 0.09	0.97	0.72 ± 0.13	0.65 ± 0.16

As, Asian; Br*, Brazilian (60–70% mestizos and afro-descendents); C, cholesterol; CETP, cholesteryl ester transfer protein; Eu, European; F, female; HDL-C, cholesterol in high-density lipoprotein; HE, heterozygote; HL, hepatic lipase; HO, homozygote; IMT, intima-media thickness; LDL-C, cholesterol in low-density lipoprotein; LPL, lipoprotein lipase; M, male; PLTP, phospholipid transfer protein; TG, triglyceride.
[a]Data as mean ± SD.

characterized by the absence of increased triglycerides (TGs) and accumulation of remnant lipoproteins. The Int14A mutation carrier presented elevated TG levels and complete deficiency of CETP activity. The presence of a slight reduction of CETP and HL activities and the increase of LPL in the other mutation carriers may explain why HALP group presented TG-enriched HDL coexisting with normal total TG levels.

We previously demonstrated that genetic components were probably the primary cause of HDL modulation in Brazilian HALP population (15). In this study, we showed that HALP phenotype was not associated with the CETP gene mutations probably due to a small number of analyzed mutants. Further genetic studies should be performed to investigate other mutations/polymorphisms in CETP, HL, and LPL genes to enable a better genetic characterization of the Brazilian HALP population.

Acknowledgements

The authors are grateful for the excellent technical assistance of Aparecida Pereira and Mirian Danelon as well as the support from Dr Edna Nakandakare and the Laboratory of Lipids, University of São Paulo Medical School. This work was supported in part by grants from the Fundação de Amparo à Pesquisa do Estado de São Paulo (FAPESP), Fundo de Apoio ao Ensino e Pesquisa da Universidade de Campinas (FAEP), and Coordenadoria de Pessoal de Ensino Superior (CAPES).

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