

# Comparison of reference ranges for small, dense LDL-cholesterol between the US population and Japanese population

Asako Minagawa<sup>1</sup>, Miki Fujimura<sup>1</sup>, Yasuki Ito<sup>1</sup>, Maiko Higuchi<sup>1</sup>, Kunio Kamata<sup>1</sup>, Kimber L. Stanhope<sup>2</sup>, Peter J. Havel<sup>2</sup>, Katalin Horvath<sup>3</sup>, Bela F. Asztalos<sup>3</sup>, and Ernst J. Schaefer<sup>3</sup>  
<sup>1</sup> Denka Seiken Co., Ltd., Tokyo, Japan, <sup>2</sup> UC Davis, Sacramento, CA, <sup>3</sup> Tufts University, Boston, MA

## OBJECTIVE:

Small, dense LDL (sd LDL) is a subfraction of low-density lipoprotein (LDL) with smaller particle size and higher density than larger, more buoyant LDL. It is known as an atherogenic lipoprotein, and many recent epidemiological and pathological studies suggest that there is a positive relationship between sd LDL-cholesterol (sd LDL-C) level and CHD occurrence. Several different methods based on ultracentrifugation, electrophoresis, and nuclear magnetic resonance (NMR) have been employed for the determination of LDL particle size. These methods require special equipment and are labor- and time-intensive, making them unfeasible for general clinical use. We recently developed a novel homogeneous assay for quantification of sd LDL-C which utilizes phospholipase and unique detergents. The new assay enables us to test numerous samples in a very short period of time. The objective of this study was to determine and compare the sd LDL-C reference ranges of the Japanese population and US population using the new assay method.

## Study Design :

The participants were recruited in two US regions (Sacramento, CA and Boston, MA, n=651) and in Japan (n=620). The inclusion criteria were applied according to the ATP III on the US population and the Japan Atherosclerosis Society on the Japanese population, respectively:  
 US: LDL-C < 160 mg/dL, HDL-C ≥ 40 mg/dL, triglycerides < 200 mg/dL, and fasting glucose < 126 mg/dL  
 Japan: LDL-C < 140 mg/dL, HDL-C ≥ 40 mg/dL, and triglycerides < 150 mg/dL  
 There were 460 of US subjects and 459 of Japanese subjects who were eligible for the study.

## Data Analysis:

Data from eligible subjects were analyzed for their means and standard deviations. Student's t-test was applied to log-transformed values (if necessary) in order to compare the distribution of the variables. Nominal p values of <0.05 were considered statistically significant. Statistical analysis was carried out for all subject data to obtain the 2.5<sup>th</sup>, 25<sup>th</sup>, 50<sup>th</sup>, 75<sup>th</sup>, and 97.5<sup>th</sup> percentile values of sd LDL-C. The sd LDL-C reference ranges were calculated as the central 95% confidence interval.

Table 1 Demographic data and lipid profiles of Reference Range Study participants.

	US subjects (n=460)	Japanese subjects (n=459)	p-value
age (years)	46.9 ± 15.4	37.1 ± 11.9	< 0.001
gender			
male(%) / female(%)	220(47.8) / 240(52.2)	218(47.5) / 241(52.5)	-
Height (cm) <sup>1)</sup>	168.8 ± 9.9	164.6 ± 8.7	< 0.001
Weight (kg) <sup>1)</sup>	74.6 ± 17.3	58.0 ± 10.5	< 0.001
BMI (m <sup>2</sup> /kg) <sup>1)*</sup>	26.2 ± 5.7	21.3 ± 2.8	< 0.001
Systolic BP (mmHg) <sup>2)</sup>	121.9 ± 15.0	117.5 ± 12.5	< 0.001
Diastolic BP (mmHg) <sup>2)</sup>	76.2 ± 10.4	72.3 ± 9.6	< 0.001
Glucose (mg/dL)	89.3 ± 9.8	77.0 ± 10.4	< 0.001
TC (mg/dL)	188.8 ± 31.6	-	-
TG (mg/dL)*	78.0 [61.0, 107.0]	64.8 [49.3, 87.0]	< 0.001
LDL-C (mg/dL)	111.9 ± 27.4	100.2 ± 21.5	< 0.001
HDL-C (mg/dL)*	63.2 ± 16.2	64.4 ± 13.9	< 0.001
sd LDL-C (mg/dL)*	24.2 ± 9.1	18.9 ± 6.2	< 0.001

All data are expressed as mean ± SD, except TG being expressed as median [25<sup>th</sup>, 75<sup>th</sup>].  
 1) : Data was calculated from 445 US subjects and 453 Japanese subjects.  
 2) : Data was calculated from 440 US subjects and 453 Japanese subjects.  
 \*: p value was obtained by using log-transformed data.  
 BMI: Body Mass Index, BP: Blood pressure, TC: Total cholesterol, TG: Triglyceride

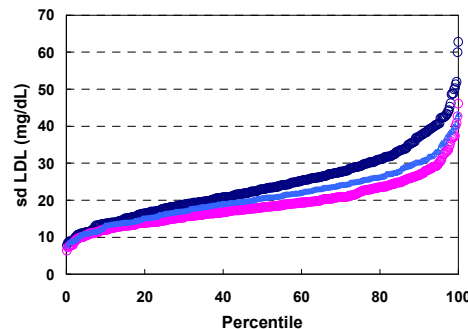


Figure 1 Comparison of distribution of the sd LDL-C concentrations between the US and Japanese population.

The sd LDL-C concentration results obtained for the US subjects (○, n=460) and Japanese subjects (△, n=459) were ranked in ascending order and plotted against its percentile. The data for the US subjects were also analyzed using the same inclusion criteria as was employed for Japanese subjects (△, n=366).

Table 2 Analysis of percentile values of sd LDL-C concentrations (mg/dL) in the US and Japanese subjects. The US data was also analyzed using the same criteria as was employed in the Japanese data.

Percentile	US subjects (n=460)	Japanese subjects (n=459)	US re-analysis (n=366)
97.5	44.8	34.7	38.7
75.0	29.3	22.4	25.6
50.0	23.0	18.0	20.7
25.0	17.8	14.5	16.4
2.5	10.2	9.3	9.5

The reference ranges (calculated as 95% confidence interval)

The US population :	10.2 - 44.8 mg/dL
Japanese population:	9.3 - 34.7 mg/dL

## RESULTS:

The distribution of demographic details and the lipid profiles for the subjects used in this analysis are outlined in Table 1. The Japanese subjects were younger by mean age of nearly 10 years than the US subjects. Basically, biological characteristics and lipid profiles were appeared to be significantly higher in the US subjects than those in the Japanese subjects in this study. The mean value of the sd LDL-C concentrations in the US subjects (24.2 ± 9.1 mg/dL) was higher than that in the Japanese subjects (18.9 ± 6.2 mg/dL) with statistical significance (p < 0.001). The reference ranges were calculated as 10.2 - 44.8 mg/dL for the US subjects and 9.3 - 34.7 mg/dL for the Japanese subjects (Table 2). Comparing distribution of the sd LDL-C concentrations, higher levels of sd LDL-C were observed in the US subjects than the Japanese subjects (Figure 1). This was especially revealing in the group with high levels of sd LDL-C, probably causing high upper limit of the US reference range. The US data was re-calculated using the same inclusion criteria as applied to the Japanese population (n=366) (Figure 1 and Table 2). In this re-analysis, the US reference range was lowered to 9.5 - 38.7 mg/dL, but the mean sd LDL-C was 21.4 ± 7.1 mg/dL still remaining significantly higher than that for the Japanese population (p < 0.05).

## CONCLUSION:

Racial difference in sd LDL-C was suggested by our study. Though the study might be lacking the comparability in its design, it was shown that the upper limit of sd LDL-C distribution in the US subjects tend to be higher than that in the Japanese subjects. Further investigations which use the subjects that match to each other may be needed to clarify whether differences in diet and/or genetic factors contribute to differences in sd LDL-C levels between the US and Japanese subjects.