

INTRODUCTION & OBJECTIVES

Type 2 diabetes (T2D) is an emerging disease in pediatric population. The association between T2D and non-alcoholic fatty liver disease (NAFLD) has been described; this association increases the risk of coronary heart disease (CHD). Recent evidence suggests that sizes and composition of HDL may be more important than HDL-C levels in predicting CHD. There is not data regarding the HDL subclasses distribution and composition in T2D youths with hepatic steatosis (HE).

This study was designed to evaluate the association between the sizes and composition of HDL with HE in T2D youths.

METHODS

The protocol was approved by the local Ethics and Research Committees. This Cross-sectional study included a total of 70 adolescents, 47 adolescents with T2D and 23 healthy adolescents. The characteristics of the study were explained to all the participants; a complete clinical history, anthropometry and physical examination were performed. The presence of HE was determined by magnetic resonance by spectroscopy (MRS). In a venous blood sample (12 hours fasting); glucose, HbA1c and lipid profile were determined. The size and composition of the HDL subpopulations were analyzed by polyacrylamide gel electrophoresis (PAGE) and a by enzymatic assays on a Hitachi 902 analyzer.

RESULTS

TABLE 1. Clinical and anthropometric characteristics in healthy adolescents and patients with T2D

	HEALTHY ADOLESCENTS n = 23	T2D WITHOUT HEPATIC STEATOSIS n = 16	T2D WITH HEPATIC STEATOSIS n = 31	P Value
Sex (M/F)	11 / 12	6 / 10	6 / 25	0.080 [§]
Age (years)	13.4 ± 2.4	15.1 ± 1.8	15.9 ± 1.6 ^β	<0.001*
Weight (kg)	51.6 ± 10.4	66.6 ± 15.3 ^α	64.6 ± 14.4 ^β	<0.001*
Height (m)	1.58 ± 0.08	1.64 ± 0.12	1.58 ± 0.08	0.106*
BMI (Kg/m ²)	20.3 ± 2.9	24.1 ± 3.4 ^α	25.6 ± 5.0 ^β	<0.001*
BMI (score z)	0.4 (-1.9 - 1.7)	1.1 (-1.2 - 2.1) ^α	1.2 (-1.0 - 4.1) ^β	0.015 ^δ
Waist circumference (cm)	71.2 ± 6.8	84.0 ± 10.5 ^α	86.6 ± 13.7 ^β	<0.001*
Waist/height ratio	0.44(0.37-0.50)	0.48(0.40-0.61) ^α	0.66(0.43-0.81) ^β	<0.001 ^δ
SBP (mmHg)	103.9 ± 6.0	102.5 ± 10.6	105.2 ± 9.0	0.617 [§]
DBP (mmHg)	63.1 ± 3.1	65.5 ± 9.0	68.8 ± 7.3 ^β	0.011*
Tanner 3-5 (%)	74.0	88.0	96.0	0.046 [§]
Duration of diabetes (months)	N/A	40.0 ± 24.1	54.6 ± 28.6	0.001 ^μ
Metformin treatment (%)	N/A	100	100	0.999 [§]
Insulin treatment (%)	N/A	64.8	66.7	0.892 [§]
Insulin dose (U/Kg/day)	N/A	0.42 ± 0.26	0.64 ± 0.25	0.028 ^μ

* ANOVA ^αP < 0.05 Control vs T2D without EH ^βP < 0.05 Control vs T2D with EH (Bonferroni-Dunn's Test)
^δ Kruskal-Wallis Test [§] x² ^μ U de Mann-Whitney

TABLE 2. Metabolic characteristics in healthy adolescent and patients with T2DM

	HEALTHY ADOLESCENTS n = 23	T2D WITHOUT HEPATIC STEATOSIS n = 16	T2D WITH HEPATIC STEATOSIS n = 31	P Value
HbA1c average (%)	N/D	7.3 ± 1.0 ^γ	8.2 ± 2.2	0.032 ^ξ
Glucose (mg/dL)	80.6 ± 4.4	190.0 ± 121.6 ^α	203.0 ± 97.0 ^β	<0.001*
Total Cholesterol (mg/dL)	152.1 ± 20.4	168.7 ± 36.7	171.2 ± 45.5	0.157*
Triglycerides (mg/dL)	86.4 (43 - 184)	139.6 (47 - 317)	189.5 (55 - 1041) ^β	0.001 ^δ
C-HDL (mg/dL)	51.9 ± 10.9	43.3 ± 6.9 ^{α,γ}	38.5 ± 9.1 ^β	<0.001*
C-LDL (mg/dL)	86.6 ± 18.0	103.0 ± 32.9	102.4 ± 26.9 ^β	0.005*
ApoB (mg/dL)	78.5 ± 13.5	99.3 ± 28.1 ^α	107.0 ± 32.2 ^β	0.001*
ApoA (mg/dL)	141.6 ± 25.5	136.0 ± 13.8	130.7 ± 19.4	0.160*
Uric Acid (mg/dL)	5.3 (3 - 7.5)	5.5 (2.5 - 8.6)	4.3 (2.4 - 8.7) ^β	0.037 ^δ
Creatinine (mg/dL)	0.6 (0.4 - 1.1)	0.6 (0.4 - 1.0)	0.6 (0.4 - 0.8)	0.234 ^δ
GOT (U/L)	19.6 (13.2 - 28.5)	19.7 (9.0 - 40.5)	24.3 (9.2 - 115.1)	0.108 ^δ
GPT (U/L)	14.4 (6.5 - 30.8)	21.4 (7.1 - 73.3)	26.4 (3.8 - 143.4)	0.747 ^δ
ALP (U/L)	207.7 (57.2 - 401.0)	139.6 (56.9 - 227.3)	132.0 (69.4 - 377.9)	0.070 ^δ
GGT (U/L)	16.17 (7.8 - 39.3)	27.45 (7.0 - 84.5)	34.0 (8.7 - 177.7) ^β	0.015 ^δ

* ANOVA ^αP < 0.05 Control vs T2D without EH ^βP < 0.05 Control vs T2D with EH (Bonferroni-Dunn's Test)
^γP < 0.05 T2D without EH vs T2D with EH ^δ Kruskal-Wallis Test ^ξ t de Student

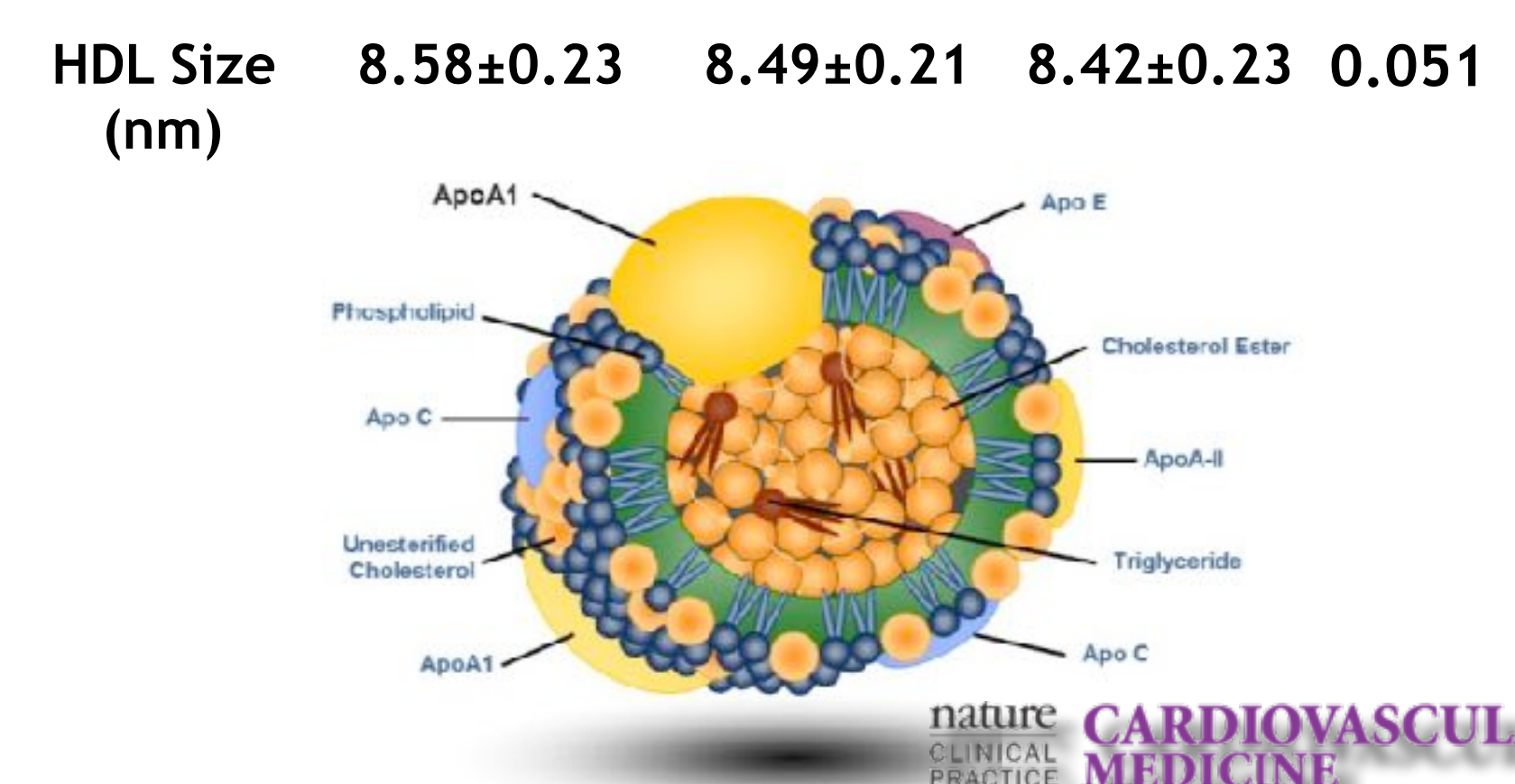
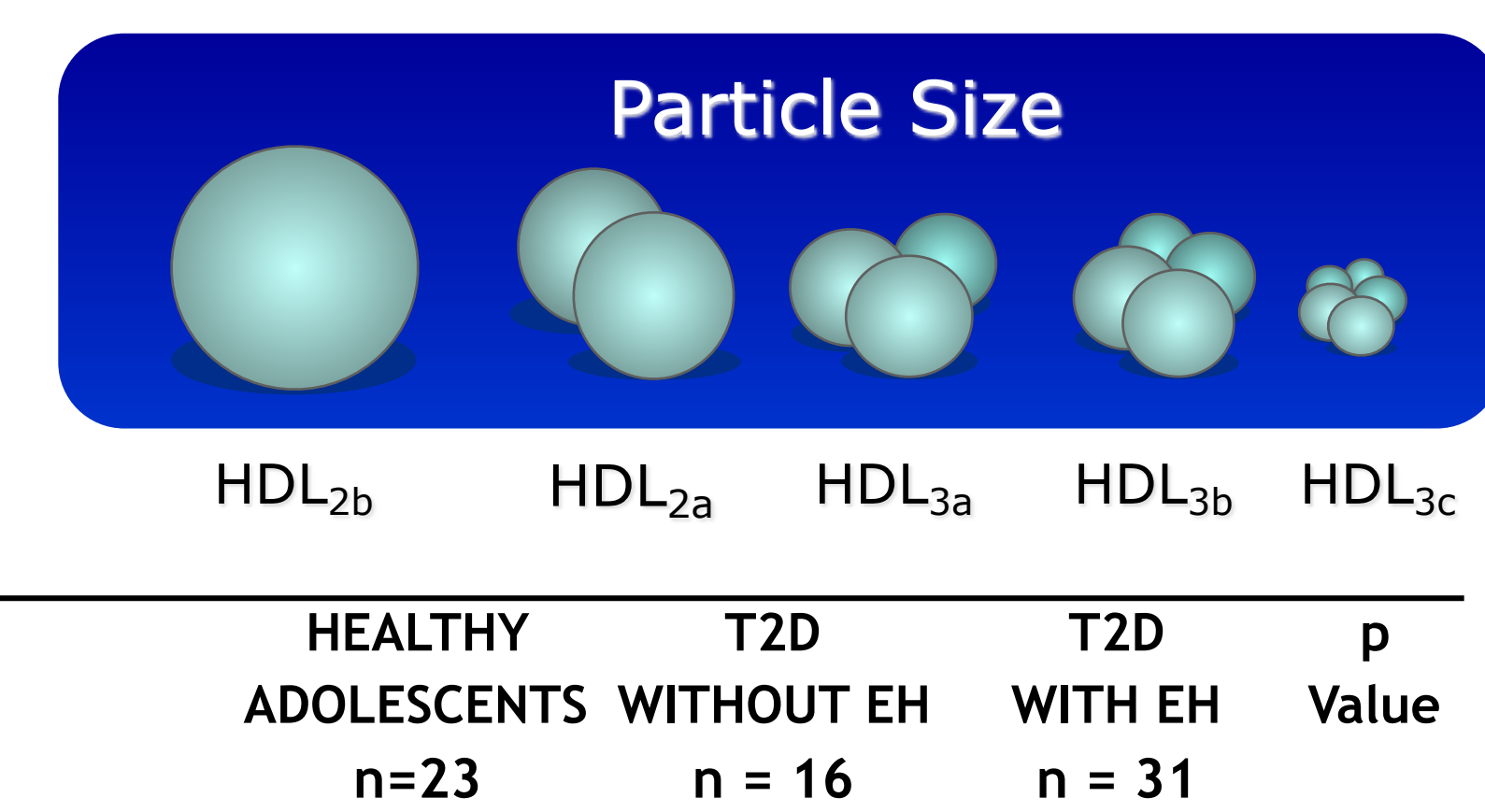
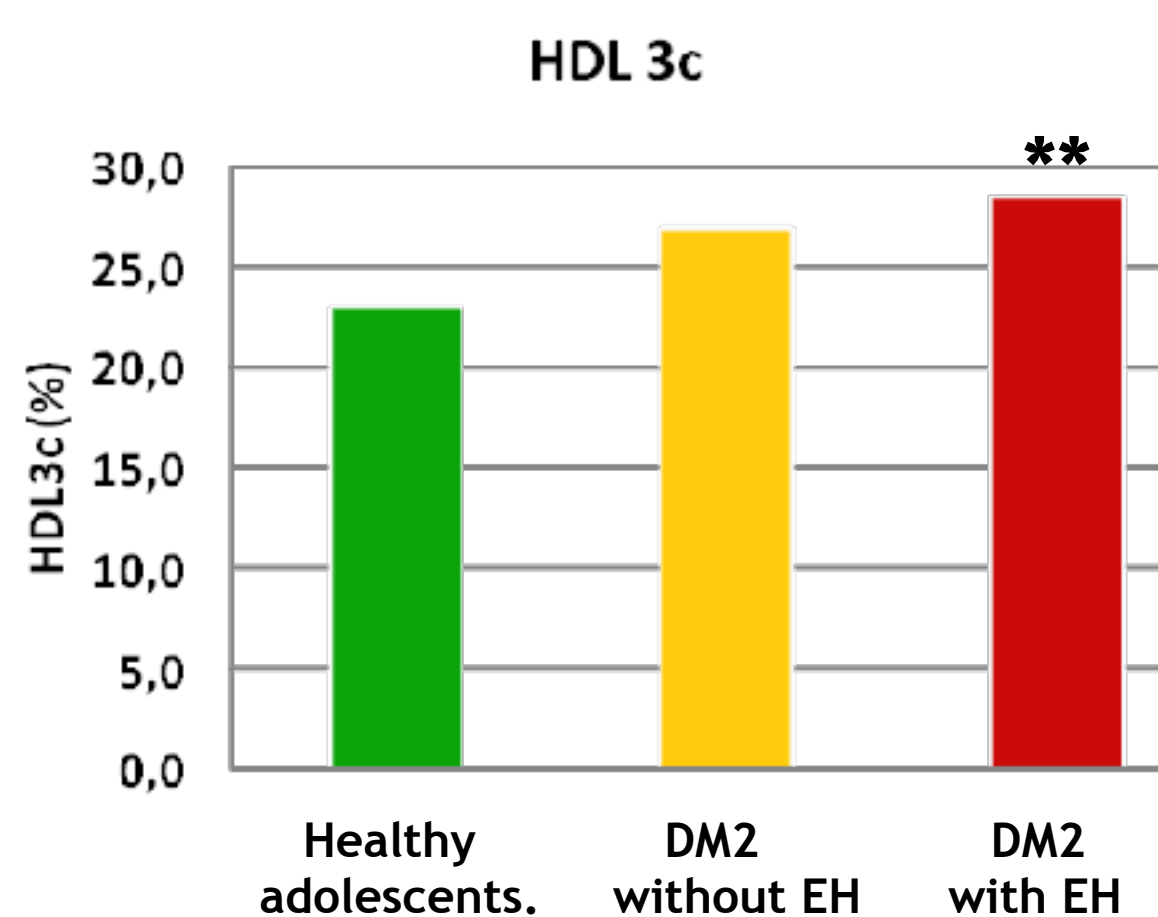
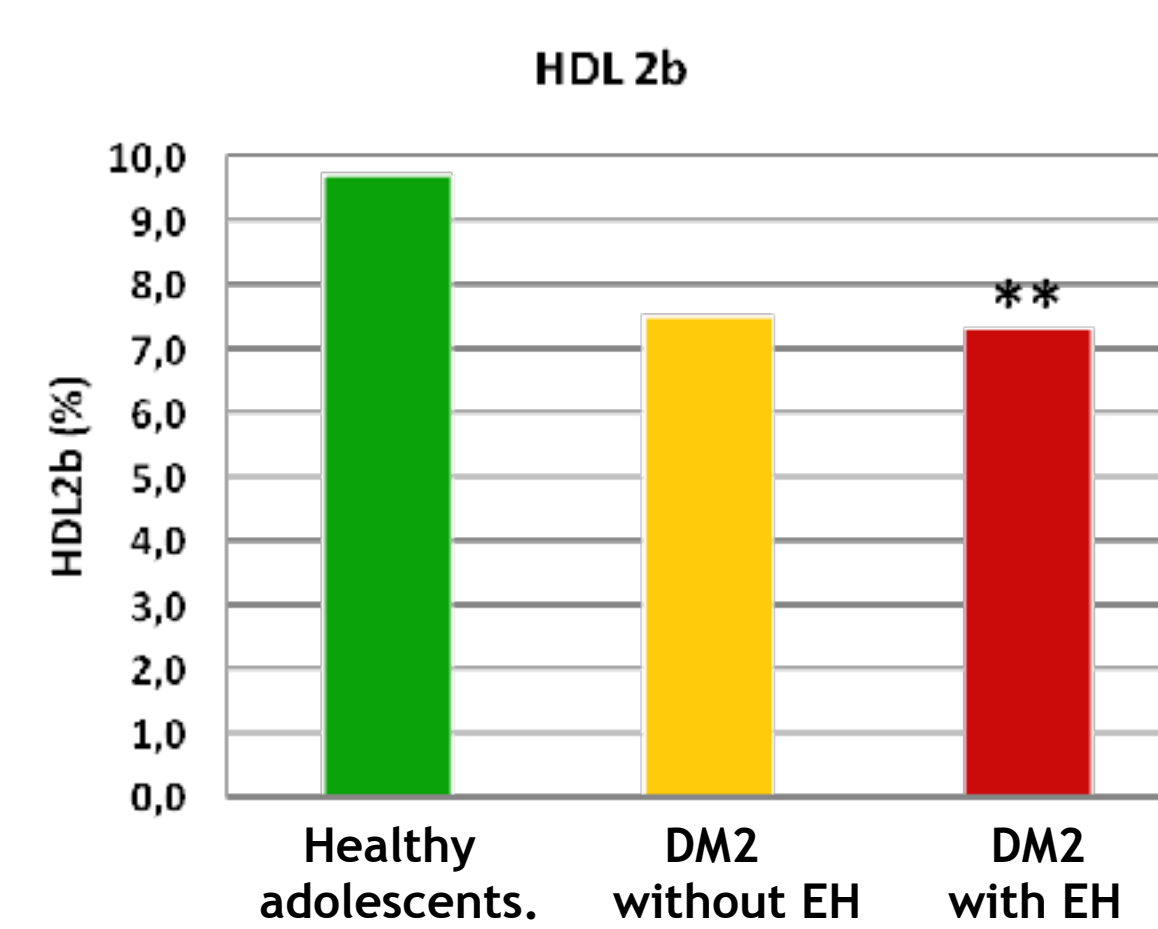


TABLE 3. HDL Composition

	HEALTHY ADOLESCENTS n = 23	T2D WITHOUT HE n = 16	T2D WITH HE n = 31	P Value
Total Proteins (%)	50.4 ± 3.2	52.9 ± 2.8	52.1 ± 4.3	0.085
Phospholipid (%)	19.5 ± 2.4	19.5 ± 2.6	19.7 ± 2.8	0.963
Triglycerides (%)	4.2 ± 1.6	4.7 ± 1.5	5.7 ± 2.4 ^β	0.032
Cholesteryl Ester (%)	22.5 ± 2.7	20.4 ± 2.0	19.3 ± 3.7 ^β	0.002
Free Cholesterol (%)	3.1 ± 0.5	2.3 ± 0.4 ^α	2.1 ± 0.4 ^β	<0.001

TABLE 4. Association of the sizes and composition of HDL with %PDF in adolescents T2D

HDL Subpopulations	PROTON DENSITY FAT FRACTION (%PDF) n = 45			
	r	p Value ^α	r*	p Value ^β
HDL Subpopulations				
HDL2b (%)	-0.237	0.026	-0.341	0.004
HDL2a (%)	-0.269	0.013	-0.249	0.094
HDL3a (%)	-0.169	0.084	0.060	0.718
HDL3b (%)	0.113	0.179	0.166	0.344
HDL 3c (%)	0.276	0.011	0.327	0.015
HDL Size (nm)	-0.285	0.009	-0.327	0.051
HDL Composition				
Total Protein (%)	0.092	0.227	0.044	0.804
Phospholipid (%)	0.058	0.319	0.019	0.919
Triglycerides (%)	0.299	0.007	0.299	0.013
Cholesteryl Ester (%)	-0.281	0.010	-0.265	0.030
Free Cholesterol (%)	-0.372	<0.001	-0.064	0.820

^αThe values were estimated by Pearson correlation analysis. ^{r*} Multiple linear regression analysis. ^β The values were obtained adjusting for age, sex, Tanner stage, BMI, HbA1c levels, insulin doses and time of evolution of T2D.

CONCLUSIONS

In adolescents with T2D, the presence of HE is associated with abnormalities in the distribution of HDL subpopulations (highest proportions of large HDL2b and lowest proportions of small HDL3c subpopulations), as well as in the lipid composition of the particles (highest proportion of TG and lowest proportions of cholesterol esters).