Original Article

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Correlation between HDL Level with Clinical and Biochemical Markers of Atherogenesis

Internal Medicine Section

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ABSTRACT

Introduction: HDL cholesterol (HDL-C) level is an independent predictor of risk for cardiovascular events in both genders, low levels of which have been demonstrated to have an inverse association with cardiovascular disease and mortality. Anthropometric measurements serve as surrogate markers for atherogenic burden. Atherosclerosis represents a chronic inflammatory process and low HDL being an inflammatory state, inflammatory markers such as high-sensitivity C Reactive Protein (hs-CRP) may provide an adjunctive method for global assessment of cardiovascular risk. TG/HDL ratio has been proved to have a high association with prevalence of metabolic syndrome and with insulin resistance.

Aim: To find out the correlation between High Density Lipoprotein (HDL) level with clinical and biochemical markers of atherogenesis.

Materials and Methods: This cross-sectional study was conducted with a study population of 200 individuals, who were either in-patients or outpatients in the Department of Medicine at Kasturba Medical College (KMC), Manipal Karnataka, India. (Study period July 2012-July 2014). Individuals aged 18-70 years being investigated for dyslipidemia for the first time were included. Data was collected by in person interview by specific questionnaire. Height, weight, waist circumference and hip circumference were measured as per standard protocol. Laboratory reports were noted for the estimation of Fasting Blood Sugar (FBS), HDL and Triglycerides (TG), hs-CRP, fasting insulin. Serum LDL was calculated. Statistical data analysis was done using SPSS 16.0 version. Mean and median values were calculated corresponding to the data. Results were analysed using Pearson's and Spearman's correlation coefficient. Independent t-test and Mann-Whitney's tests were used for calculation of significance of correlation.

Results: Among the studied individuals 53% (106) were males and 47% (94) were females. A total of 77%(154) of the individuals with low HDL and 70%(140) individuals with normal HDL had abnormal waist circumference. Mean Waist Hip Ratio (WHR) was same across groups with low and normal HDL. Mean Body Mass Index (BMI) was similar in both the groups among males; however, a statistically significant difference was found among females. Abnormal waist circumference was found similar across both the groups. Median hs-CRP showed a statistically significant higher values among individuals with lower HDL. Fasting insulin and HOMA-IR was higher among individuals with low HDL (vs normal HDL), but it was statistically insignificant. Median TG/HDL ratio among 41 individuals were found to be 3.08, which was higher than the cut-off to signify insulin resistance. HOMA-IR and TG/HDL ratio showed a statistically significant positive correlation suggesting that TG/HDL ratio can be used as a marker of insulin resistance.

Conclusion: This study revealed that anthropometric indices showed an inverse correlation with HDL levels, with BMI being a better predictor of HDL-C changes. HDL was lower in individuals with insulin resistance, with TG/HDL ratio being a significant marker of insulin resistance.

Keywords: Anthropometric indices, Homeostasis model assessment-insulin resistance, Insulin resistance

INTRODUCTION

Cardiovascular Diseases (CVD) are increasing due to over utilisation of fats or due to genetic causes. It is a leading cause of morbidity and mortality from infancy to old age. Atherosclerosis is an insidious and vulnerable disease [1]. Though conventional risk prediction algorithms are made available on presence of major cardiovascular risk factors identified in diseased individuals, authentic and exact biomarkers of CVDs are lacking. Lipid abnormalities have been pragmatic in obese individuals, including elevated cholesterol, triglycerides, and lower HDL cholesterol levels [2]. Atherogenic dyslipidemia is normally seen in individuals with obesity, metabolic syndrome, insulin resistance and type 2 Diabetes Mellitus (DM). This compendium has emerged as an important marker for increased cardiovascular risk observed in these populations [3]. Insulin resistance plays a vital role in pathophysiology of type 2 diabetes and is tightly related with major public health problems, including obesity, hypertension, coronary artery disease, dyslipidemia, and a cluster of metabolic and cardiovascular abnormalities that define the metabolic syndrome [4].

High Density Lipoprotein Cholesterol (HDL-C) level is an independent predictor of risk for cardiovascular events in both genders. High serum levels of HDL is associated with reduced risk for atherosclerosis and its clinical sequelae [5]. The molecular basis for the apparent vascular protection afforded by elevated HDL-C is widely attributed to the ability of HDL particles to drive reverse cholesterol transport. HDL particles also seem to have anti-inflammatory, antioxidant and antithrombotic properties [6]. Anthropometric measurements serve as surrogate markers for atherogenic burden. Atherosclerosis represents a chronic inflammatory process and low HDL being an inflammatory state. Inflammatory markers such as hs-CRP may provide an adjunctive method for global assessment of cardiovascular risk [7]. Low HDL levels have been demonstrated to have an inverse association with cardiovascular disease and mortality. TG/HDL ratio has been proved to have a high association with prevalence of metabolic syndrome and with insulin resistance [8].

In the present study, we have tried to correlate HDL level with clinical and biochemical markers of atherogenesis. Anthropometric measurements, measurement of hs-CRP levels and correlation with HDL levels may help us predict low HDL which is a part of metabolic syndrome and thus predict the future risk of atherosclerosis early and define the at risk population. This study focused on considering inclusion of all anthropometric and biochemical parameters in

one single study. Even though there have been similar studies in literature, all parameters have not been included in a single study.

MATERIALS AND METHODS

Study Population

This cross-sectional study was conducted with a study population of 200 individuals, who were either inpatients or outpatients in the Department of Medicine at Kasturba Medical College (KMC), Manipal, Karnataka, India. The study period extended from July 2012 to July 2014. Individuals being investigated for dyslipidemia for the first time were included. Individuals aged 18-70 years were included and they underwent complete lipid profile testing.

Sample Size

Sample size was calculated as 198 using the below mentioned formula.

$$n=\frac{\left[Z_{1-\frac{\alpha}{2}}+Z_{1-\beta}\right]^2}{\{1/_2 log\frac{[1+r_1]}{1-r_1}-1/_2 log\frac{[1+r_0]}{1-r_0}\}^2}$$

Where:

n-sample size

At 5% level of significance

$Z_{1-\alpha/2}$=1.96

For 80% power=(1-β)

$Z_{1-\beta}$=0.84

r_1=-0.5 (hypothesised value of correlation coefficient)

r_0=-0.335

200 subjects were included in the study.

The exclusion criteria were as follows: Individuals on weight losing diet; long term lipid lowering agents use (for last three months); patients on aspirin or NSAIDs; pregnant women; women on hormone replacement therapy; patients with febrile illness in the past one month suggested of infection (bacterial, fungal, mycobacterial, viral); active diseases like rheumatoid arthritis, ankylosing spondylitis, psoriatic arthritis, systemic vasculitis etc., acute pancreatitis, recent MI (in the last three months), tumour embolism; Malignancy-lymphoma, carcinoma, sarcoma; Trauma-surgery, burns, fracture.

Methodology

Data was collected by in person interview using a specific questionnaire. Height, weight, waist circumference and hip circumference were measured as per standard protocol. BMI was calculated using the formula weight/(height in meters)^2. Laboratory reports were noted on the estimation of FBS; estimation of total cholesterol (CHOD-POD enzymatic method); estimation of triglycerides (GPO-PAP method); estimation of HDL-cholesterol was done by direct homogenous method. LDL Cholesterol was calculated using the formula-Total cholesterol-(HDL-C+Triglycerides)=LDL cholesterol. hs-CRP was measured using the Immunoturbidimetric method. Fasting insulin was estimated using the chemiluminescence method. All the biochemical tests were done using ROCHE diagnostics machine, Germany except for HbA1c levels that was done using Biorad-D-10, Germany.

The Institutional ethics committee approval was obtained for the study (IEC345/2011).

STATISTICAL ANALYSIS

Statistical data analysis was done using SPSS 16.0 version. Mean and median values were calculated corresponding to the data. Results were analysed using Pearson's and Spearman's correlation coefficient. Independent t-test and Mann Whitney's tests were used for calculation of significance of correlation.

RESULTS

Two hundred individuals were enrolled in the study; 53% (106) were males and 47% (94) were females. Mean age of the study population was 48.91±10.59 years, with mean age of males being 50.25±10.97 years and that of females being 47.39±9.99 years. Age distribution showed similar pattern among males and females [Table/Fig-1].

Anthropometry Measurements	Low HDL (n=100)	Normal HDL (n=100)
Mean BMI	26.65±4.90	25.08±4.28
Male	25.45±3.73	25.62±3.98
Female	27.52±5.56	24.23±4.64
Waist Circumference	93.22±11.08	91.98±10.28
Waist Hip Ratio	0.96±0.06	0.96±0.04
Male	0.98±0.05	0.97±0.04
Female	0.94±0.06	0.96±0.04

[Table/Fig-1]: Descriptives of anthropometric measurements in the study population.

Distribution of Regular Physical Activity

Assessment of physical activity was done arbitrarily based on the below mentioned criteria, recommended by American College of Sports Medicine and the American Heart Association. To promote and maintain health, all healthy adults aged 18 to 65 year need moderate-intensity aerobic (endurance) physical activity for a minimum of 30 minute on five days each week or vigorous-intensity aerobic physical activity for a minimum of 20 minute on three days each week.

According to this, 56 individuals (28%) satisfied the criteria for regular physical activity, and 144 individuals (72%) did not perform regular physical activity.

Distribution of Comorbidities

Among the study group, 26.5% (53) were found to be diabetic and 73.5% (147) were non diabetic. A 20% (40) were hypertensive and 6% (12) were found to be smokers. Among 200 patients, 1% (2) was detected to have Ischemic Heart Disease (IHD). Both the patients with ischemic heart disease were found to have low HDL.

Biochemical and Anthropometric Indices

Mean HDL was found to be 46.2±12.63. Mean HDL was higher among females (50.24±13.90) as compared to males (42.62±10.18).

Mean BMI was similar in both the groups among males; however, a statistically significant difference was found in females. Abnormal waist circumference was found in similar numbers across both the groups.

hsCRP among Low HDL and Normal HDL

Median hsCRP were higher among individual with lower HDL, which demonstrated a statistically significant difference [Table/Fig-2]. (p=0.015). A negative correlation was found between hsCRP and HDL with r=-0.197.

	Low HDL (n=35)			Normal HDL (n=31)			p-value
	Median	25th percentile	75th percentile	Median	25th percentile	75th percentile	
hsCRP	2.00	1.2	5.3	1.1	0.70	2.3	0.015

[Table/Fig-2]: Distribution of hsCRP among low HDL and normal HDL individuals. hsCRP: High sensitivity C reactive protein; HDL: High density lipoprotein

Assessment of Insulin Resistance

Fasting insulin estimation and Homeostatic Model Assessment-Insulin Resistance (HOMA-IR) calculation was done in 41 subjects. A negative correlation was found between HDL and fasting insulin

(r=-0.201) and HOMA IR (r=-0.223), a measure of insulin resistance. Fasting insulin and HOMA-IR were found higher among individual with low HDL (13.15 and 3.39 respectively) compared with individual with normal HDL (8.40 and 2.57 respectively), but it was statistically insignificant [Table/Fig-3].

	Low HDL (n=24)			Normal HDL (n=17)			p-value
	Median	25th percentile	75th percentile	Median	25th percentile	75th percentile	
Fasting Insulin	13.15	7.30	16.70	8.40	6.80	12.05	0.138
HOMA-IR	3.39	1.79	5.85	2.57	1.55	3.54	0.122

[Table/Fig-3]: Parameters for assessment of insulin resistance.
HOMA-IR: Homeostatic model assessment-insulin resistance; HDL: High density lipoprotein

TG/HDL and Insulin Resistance

Median TG/HDL ratio among 41 individuals were found to be 3.08, which is higher than the cut off to signify insulin resistance [Table/Fig-4].

Median TG/HDL Ratio (n=41)	Interquartile Range	
	25th percentile	75th percentile
3.08	2.42	4.38

[Table/Fig-4]: Median TG/HDL ratio in the study population.
TG: Triglyceride; HDL: High density lipoprotein

Correlation Coefficients

Out of the 41 individuals, 21 patients were found to have TG/HDL ≥3. HOMA-IR found higher among individuals with higher TG/HDL ratio (median=4.34 v/s median=2.42) which is statistically significant (p=0.041). HOMA-IR and TG/HDL ratio showed a statistically significant positive correlation (p=0.009) suggesting that TG/HDL ratio can be used as a marker of insulin resistance [Table/Fig-5].

TG/HDL v/s	Correlation Coefficient	p-value
HOMA-IR	0.402	0.009

[Table/Fig-5]: Correlation Coefficients.
TG: Triglyceride; HDL: High density lipoprotein; HOMA-IR: Homeostatic model assessment-insulin resistance

DISCUSSION

In the current study, higher prevalence of low HDL were seen among female (58.5%) as compared to male (42.5%). Mean HDL were higher among female as expected for their gender. Similar results were demonstrated by Mohan V et al., in CURES-34 study with female having higher prevalence of lower HDL. The prevalence of metabolic syndrome has been shown to be higher among female with the increased propensity of women to have central obesity. The gender difference may be due to different cut-off points for HDL-C [9-13].

Mean BMI in the study population were 25.86 kg/m^2 which was higher than the cut-off value for Asian standards (BMI >23 kg/m^2) [14]. Female with low HDL had a higher BMI as compared to those with normal HDL, which was statistically significant. Thus, higher BMI in female could be used to predict lower HDL and atherogenic dyslipidemia.

HDL and BMI showed a statistically significant inverse correlation in the current study (r=-0.192, p=0.006). Similar results were demonstrated by Schröder H et al., in 2003 in Southern Europe (r = -0.19, p<0.001) and Seidell et al., in 2001 in the Quebec Family Study (p<0.05) [15,16].

An abnormal waist circumference could also be used as a marker to predict low HDL in female. This is well in line with Indian obesity which is central. A 82% of females and 65% of males were found to have abnormal waist circumference. The number was higher among females because of the lower cut-off among them [9,10].

Mean Waist Hip Ratio (WHR) were similar but higher than the cut-off values (0.88 and 0.81 for men and women respectively) in both the groups [17]. WHR also showed an inverse association with HDL, thus showing a trend though not statistically significant.

This study demonstrated a negative correlation between HDL and hsCRP which was statistically non-significant. Fröhlich M et al., and Kim KI et al., have demonstrated a low level of negative correlation between HDL and hsCRP which was statistically significant [18,19]. Thus, low HDL may be considered as an inflammatory marker.

Study showed that individuals with triglyceride-HDL ratio ≥3 had a higher HOMA-IR value compared to those with a ratio of <3 which was statistically significant (p=0.041) suggesting a higher TG to HDL ratio is a better predictor of insulin resistance. HOMA-IR and TG/HDL showed a statistically significant positive correlation strengthening the fact that TG/HDL ratio can be used a marker of insulin resistant state. Study conducted by Sayantan et al., in Kolkata demonstrated similar results [20]. Various other studies have shown TG/HDL ratio to be a better marker of insulin resistance [21-23].

Hence, while dealing with atherogenic Indians, clinicians should consider combination of anthropometric parameters like WHR, BMI, HDL hs-CRP, and triglyceride-HDL ratio.

LIMITATION

The sample size was smaller with respect to estimation of hs-CRP and fasting insulin levels. Physical activity was assessed arbitrarily with no questionnaire being administered for assessment.

CONCLUSION

This study revealed that anthropometric indices showed an inverse correlation with HDL levels, with BMI being a better predictor of HDL-C changes. Waist circumference demonstrated a trend of inverse association with HDL-C, but did not show a significant negative correlation. HDL and hs-CRP demonstrated an inverse relation showing statistically non-significant. HDL was found to be lower in individuals with insulin resistance, with TG/HDL ratio being a significant marker of insulin resistance.

REFERENCES

[1] Pepys MB, Hirschfield GM. C-reactive protein: A critical update. J Clin Invest. 2003;111(12):1805-12.
[2] Austin MA, King MC, Vranizan KM, Krauss RM. Atherogenic lipoprotein phenotype. A proposed genetic marker for coronary heart disease risk. Circulation. 1990;82(2):495-506.
[3] Musunuru K. Atherogenic dyslipidemia: cardiovascular risk and dietary intervention. Lipids. 2010;45(10):907-14.
[4] Muniyappa R, Iantorno M, Quon MJ. An integrated view of insulin resistance and endothelial dysfunction. Endocrinol Metab Clin North Am. 2008;37(3):685-711.
[5] Toth PP, Davidson MH. High-density lipoproteins: marker of cardiovascular risk and therapeutic target. J Clin Lipidol. 2010;4(5):359-64.
[6] Nicholls SJ RK, Barter PJ. High-density lipoproteins as therapeutic targets. Curr Opin Lipidol. [review]. 2005;16(3):345-49.
[7] Ridker PM. High-Sensitivity C-Reactive Protein: potential adjunct for global risk assessment in the primary prevention of cardiovascular disease. Circulation. 2001;103(13):1813-18.
[8] Grundy SM. Small LDL, atherogenic dyslipidemia, and the metabolic syndrome. Circulation. 1997;95(1):1-4.
[9] Mabry RM, Reeves MM, Eakin EG, Owen N. Gender differences in prevalence of the metabolic syndrome in Gulf Cooperation Council Countries: a systematic review. Diabet Med. 2010;27(5):593-97.
[10] Beigh SH, Jain S. Prevalence of metabolic syndrome and gender differences. Bioinformation. 2012;8(13):613-16.
[11] Park HS, Oh SW, Cho S-I, Choi WH, Kim YS. The metabolic syndrome and associated lifestyle factors among South Korean adults. International Journal of Epidemiology. 2004;33(2):328-36.
[12] Njelekela MA, Mpembeni R, Muhihi A, Mligiliche NL, Spiegelman D, Hertzmark E, et al. Gender-related differences in the prevalence of cardiovascular disease risk factors and their correlates in urban Tanzania. BMC Cardiovasc Disord. 2009;9:30.
[13] Park HS, Lee SY, Kim SM, Han JH, Kim DJ. Prevalence of the metabolic syndrome among Korean adults according to the criteria of the International Diabetes Federation. Diabetes Care. 2006;29(4):933-34.
[14] Appropriate body-mass index for Asian populations and its implications for policy and intervention strategies. The Lancet. 2004;363(9403):157-63.

[15] Schröder H, Marrugat J, Elosua R, Covas MI. Relationship between body mass index, serum cholesterol, leisure-time physical activity, and diet in a Mediterranean Southern-Europe population. The British Journal of Nutrition. 2003;90(2):431-39.
[16] Seidell JC, Pérusse L, Després JP, Bouchard C. Waist and hip circumferences have independent and opposite effects on cardiovascular disease risk factors: the Quebec Family Study. The American Journal of Clinical Nutrition. 2001;74(3):315-21.
[17] Snehalatha C, Viswanathan V, Ramachandran A. Cutoff values for normal anthropometric variables in Asian Indian adults. Diabetes Care. 2003;26(5):1380-84.
[18] Fröhlich M, Imhof A, Berg G, Hutchinson WL, Pepys MB, Boeing H, et al. Association between C-reactive protein and features of the metabolic syndrome: a population-based study. Diabetes Care. 2000;23(12):1835-39.
[19] Kim KI, Oh SW, Ahn S, Heo NJ, Kim S, Chin HJ, et al. CRP level and HDL cholesterol concentration jointly predict mortality in a Korean population. The American Journal of Medicine. 2012;125(8):787-95.
[20] Ray S, Bairagi AK, Guha S, Ganguly S, Ray D, Basu AK, et al. A simple way to identify insulin resistance in non-diabetic acute coronary syndrome patients with impaired fasting glucose. Indian J Endocrinol Metab. 2012;16(Suppl 2):S460-64.
[21] Chiang JK, Lai NS, Chang JK, Koo M. Predicting insulin resistance using the triglyceride-to-high-density lipoprotein cholesterol ratio in Taiwanese adults. Cardiovascular Diabetology. 2011;10(1):1-6.
[22] Giannini C, Santoro N, Caprio S, Kim G, Lartaud D, Shaw M, et al. The Triglyceride-to-HDL Cholesterol Ratio: Association with insulin resistance in obese youths of different ethnic backgrounds. Diabetes Care. 2011;34(8):1869-74.
[23] Salazar MR, Carbajal HA, Espeche WG, Leiva Sisnieguez CE, March CE, Balbín E, et al. Comparison of the abilities of the plasma triglyceride/high-density lipoprotein cholesterol ratio and the metabolic syndrome to identify insulin resistance. Diabetes and Vascular Disease Research. 2013;10(4):346-52.

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