DOI: 10.7860/JCDR/2020/43462.13632

Original Article

Biochemistry Section

Lipid Profile and C-Reactive Protein Levels in Healthy, Overweight and Obese Adults: A Hospital-based Observational Study

VANEET KAUR[1], HIMANSHU MADAAN[2], MEENAKSHI PURI[3], PAWAN KUMAR KARE[4]

ABSTRACT

Introduction: Numerous biomarkers involved in inflammation have been associated with cardiovascular events, out of which high sensitivity-C-Reactive Protein (hs-CRP), an acute-phase protein, appears to be the most promising. The association between dyslipidaemia and obesity is well established in literature and has been found to be risk factor for Cardiovascular Disease (CVD).

Aim: To study the levels of lipid profile and hs-CRP and also find out the relationship between hs-CRP and Body Mass Index (BMI) in obesity.

Materials and Methods: The present observational study was carried out from June 2018 to August 2018 in Kalpana Chawla Government Medical College, Haryana, India. A total of 100 apparently healthy volunteers, aged 21-60 years were enrolled for this study. The participants were divided on the basis of BMI into three groups; Healthy (normal): 18.5-22.9 kg/m^2, overweight: 23-24.9 kg/m^2, and obese ≥25 kg/m^2. Anthropometric measurements and biochemical investigations were conceded in all the study participants. Lipid profile and hs-CRP levels were estimated on fully automated clinical chemistry analyser. Statistical analysis was conducted using IBM SPSS statistics (version 22.0). A $p<0.05$ was considered as significant level.

Results: The median levels of HDL were the lowest in the obese group and a statistically significant difference was observed in HDL levels between healthy and obese group ($z=3.190$, $p=0.001$) and between overweight and obese group ($z=2.760$, $p=0.006$). The median hs-CRP levels were highest in the obese group and statistically significant difference was observed between healthy and overweight group ($z=2.009$, $p=0.044$) and between healthy and obese group ($z=2.849$, $p=0.004$). A significant positive correlation was observed between BMI and hs-CRP levels ($r=0.302$, $p<0.002$). It was further observed that 17 subjects of obese group had hs-CRP levels greater than 3 mg/L as compared to eight of healthy group and nine of overweight group.

Conclusion: The subjects of the obese group are at the highest risk of CVD. Hence, they need to be considered for future development. However, abnormalities of lipid metabolism were not observed in the obese study subjects.

Keywords: Acute phase protein, Body mass index, Cardiovascular disease, Inflammation

INTRODUCTION

Obesity is an enduring ailment established in both developed and developing countries [1]. Accumulation of fat may be due to excess calorie intake, lack of exercise, and interaction of various socioeconomic, environmental and genetic factors [2]. More than half of the world's population is considered overweight and being overweight is associated with several co-morbidities such as Type 2 Diabetes Mellitus (T2DM), CVD, hypertension, dyslipidaemia, respiratory disease, osteoarthritis, and depression [1]. Among these, dyslipidaemia associated with obesity leads to inflammatory changes in vessels which also contribute to cardiovascular diseases [3,4]. CVD involves inflammatory processes and serum inflammatory markers and thus considered to be important for the evaluation of cardiovascular risk [5]. Adipose tissue produces numerous pro-inflammatory cytokines such as Interleukin-1 (IL-1), Tumour Necrosis Factor-α (TNF-α) and Interleukin-6 (IL-6) which are involved in synthesis of hs-CRP in liver [6]. Hs-CRP, an acute phase protein, which is markedly increased in inflammatory conditions and has been considered as an important indicator of cardiovascular risk [7,8].

A significant correlation has been observed between increased hs-CRP levels and adiposity, insulin resistance and low High Density Lipoprotein Cholesterol (HDL-C) by various studies [9-12]. Although there is no elaborate description linking the two physiological mechanisms and further an association partly mediated by adipose tissue, the main source of inflammatory cytokines is also possible [13]. Epidemiologic studies also reported strong associations between hs-CRP and obesity through anthropometric indexes [9-12]. Body mass index (BMI) is frequently used mode for relating obesity and assessing the risk for obesity-related disorders [14].

It has been stated by World Health Organisation (WHO) that Asians have a higher percentage of body fat than white people of same age, sex and BMI with the increased proportion of risk factors for T2DM and CVD. Therefore, for the Asian population, the current WHO cut-off point of 25 kg/m^2 cannot provide adequate information on risks related to overweight and obesity [15]. To resolve such issue, guidelines for obesity and overweight based on BMI for Asian Indians were revised based on consensus developed through discussions by a Prevention and Management of Obesity and Metabolic Syndrome group [16]. According to these revised guidelines, Asian Indians are categorised as overweight (BMI 23.0-24.9 kg/m^2) [17] and obesity (BMI ≥25 kg/m^2) [16]. Therefore, the present study was designed to study the levels of lipid profile parameters and hs-CRP levels in all the three groups as categorised based on the BMI as per the Asian Indian guidelines and finding out the association between hs-CRP and BMI among study subjects.

MATERIALS AND METHODS

The present observational cross-sectional study was carried out between June 2018 to August 2018 in the Department of Biochemistry, Kalpana Chawla Government Medical College, Karnal (Haryana), India. A total of 100 apparently healthy volunteers (attendant of patients and working hospital staff) participated in this study. As it was a hospital-based study and the duration for the study was only 2 months, therefore 100 healthy volunteers screened randomly and recruited for this study. The age of participants ranged

between 21 to 60 years. The inclusion criteria for participants were normotensive and normoglycaemic. The participants having Diabetes Mellitus (DM), hypertension, Chronic Obstructive Pulmonary Disease (COPD), bronchial asthma, malignancy, renal disease and cardiovascular disease were excluded from the study. All enrolled participants were divided into three groups on the basis of BMI as per the Asian guidelines [16] criteria; Healthy Normal: (BMI 18.5-22.9 kg/m^2), overweight: (23-24.9 kg/m^2), and obese (BMI ≥25 kg/m^2). The study was approved by Institutional Ethical Research Committee (IERC) of Kalpana Chawla Government Medical College, Karnal (Letter Number-KCGMC/IEC/2018/05/06, Dated-24/5/2018). Written consent of the study subjects was obtained before the commencement of the study.

Anthropometric Measurements

Anthropometric measurements such as body weight was measured by making the subject stand in the centre of the weighing machine and height by making the study subject stand against an upright surface. The BMI was calculated as weight (kg) divided by height (m^2). Waist circumference (cm) was measured in the middle of the lower margin of ribs and upper border of iliac crest at the completion of regular expiration. Hip circumference (cm) was measured at the level of greater trochanter and Waist: Hip Ratio (WHR) was also calculated [18].

Biochemical Investigations

Five millilitre fasting blood sample (5 mL) was obtained for biochemical investigations. All biochemical investigations such as Fasting Blood Sugar (FBS), total cholesterol, HDL-C, LDL-C, triglyceride and hs-CRP were carried out on Fully Automated Clinical Chemistry Analyser (Cobas-c501, Roche Diagnostics) in the Department of Biochemistry, KCGMCH, Karnal, India. FBS was estimated by hexokinase method [19], total cholesterol in serum was measured by enzymatic colourimetric method [20], serum triglycerides were estimated by enzymatic colourimetric method [21], HDL-Cholesterol (HDL-C) was measured by homogenous enzymatic colourimetric method [22] and LDL-Cholesterol (LDL-C) was carried out by homogenous enzymatic colourimetric assay [23]. Quantitative estimation of hs-CRP was done by particle enhanced immunoturbidimetric assay [24].

STATISTICAL ANALYSIS

Continuous data were given as mean±SD and range or median as appropriate. Normality of quantitative data were checked by measures of Kolmogorov Smirnov tests of Normality. Group (BMI category) comparisons of values of skewed data were made with Kruskall Wallis test followed by Mann-Whitney test for two groups. Group comparisons were made with the Chi-Square test. Spearman's correlation coefficient was calculated to observe the relationship between BMI and hs-CRP. p-value <0.05 was considered as significant level.

RESULTS

The study subjects were divided on the basis of BMI (kg/m^2) into healthy (n=35), overweight (n=22) and obese groups (n=43). The distribution of study subjects into males and females based on BMI groups was found insignificant (p=0.415) as shown in [Table/Fig-1].

Gender	Total (n=100)	Healthy (n=35)	Overweight (n=22)	Obese (n=43)	p-value
Males	48	19	8	21	0.415
Females	52	16	14	22	

[Table/Fig-1]: Gender wise distribution of study subjects among groups.
n: Number; p<0.05 was significant level

The [Table/Fig-2] shows that there was a significant difference in the waist and hip circumference found between all the study groups (p<0.001). Median HDL-cholesterol level was the lowest in the obese group (37.9 mg/dL). The difference in the levels of waist and hip circumference and HDL-cholesterol levels between healthy and obese group were statistically significant (p<0.001, z=4.524), (p<0.001, z=4.660) and (p=0.001, z=3.190), respectively. The difference in the levels of waist and hip circumference and HDL-cholesterol levels between overweight and obese group were also statistically significant (p<0.001, z=3.899) (p=0.002, z=3.024) (p=0.006, z=2.760), respectively.

The median level of hs-CRP was the highest in the obese group (2.5 mg/L) followed by overweight (2.0 mg/L) and lowest in the healthy group (1.1 mg/L). hs-CRP was found statistically significant (p<0.013) between all the study groups. The difference in the levels of hs-CRP between healthy and overweight group was statistically significant (p=0.044, z=2.009) and between healthy and obese group was also statistically significant (p=0.004, z=2.849).

In [Table/Fig-3], subjects were divided into three subcategories based on their hs-CRP levels. A statistically significant difference (p=0.032) was found in the distribution of subjects between all the study groups.

BMI showed a positive correlation with hs-CRP levels (r=0.302, p<0.002) in the study subjects as shown in [Table/Fig-4].

Parameter (s)	Healthy (n=35)		Overweight (n=22)		Obese (n=43)		p-value
	Mean±SD	Median	Mean±SD	Median	Mean±SD	Median	
Waist circumference (cm)	87.22±22.4	87.0	86.1±12.9	86.5	97.1±12	100	<0.001[a] <0.001[c] <0.001[d]
Hip circumference (cm)	93.25±2.9	97.0	95.6±14.4	100	104.2±13.0	105.5	<0.001[a] <0.001[c] 0.002[d]
WHR	0.9±0.007	0.92	0.91±0.06	0.93	0.93±0.04	0.94	0.249[a]
FBS (mg/dL)	83.9±14.6	85.5	88.5±9.65	90.0	90.3±8.9	90.0	0.131[a]
Total cholesterol (mg/dL)	165.0±32.1	162	156.3±28.7	151.5	153.0±32.7	150	0.156[a]
Triglycerides (mg/dL)	125.8±52.8	119	130.3±75.6	112.5	138±59.6	122	0.497[a]
HDL-C (mg/dL)	45.8±11.4	44	45.0±10.8	43.5	37.8±7.1	37.9	0.002[a] 0.001[c] 0.006[d]
LDL-C (mg/dL)	107.5±26.6	103.5	103.8±25.7	98.5	102.1±6.4	102	0.677[a]
hs-CRP (mg/L)	2.39±3.28	1.1	3.44±3.08	2.0	3.3±2.6	2.5	0.013[a] 0.044[b] 0.004[c]

[Table/Fig-2]: Anthropometric and biochemical characteristics of the study subjects.
WHR: Waist hip ratio; FBS: Fasting blood glucose; p<0.05 was significant level; [a]between among groups; [b]between healthy and overweight group; [c]between healthy and obese group; [d]between overweight and obese group

hs-CRP (mg/L)	Total (n=100)	Healthy (n=35)	Overweight (n=22)	Obese (n=43)	*p-value
<1 mg/L	29	17	5	7	0.032
1-3 mg/L	37	10	8	19	
>3 mg/L	34	8	9	17	

[Table/Fig-3]: Distribution of study subjects among groups on the basis of hs-CRP levels.
n: number, p<0.05 was significant level; *Chi square test

BMI vs. hs-CRP	Total subjects (n=100)
Correlation coefficient (ρ)	0.302
p-value	<0.002

[Table/Fig-4]: Correlation between BMI and hs-CRP levels.
ρ: Spearman's rho correlation coefficient; correlation was significant at 0.01 level (2-tailed)

DISCUSSION

The present study shows that the difference between the levels of waist circumference and hip circumference between the healthy and obese group was statistically significant and between the overweight and obese group was also statistically significant. However, WHR did not reflect significant change among obese group and healthy group. The study findings are similar to the study of Dev N and Marcus CR, [25].

In the present study, the fasting blood sugar levels were within normal range in obese group. However, fasting blood sugar levels were slightly higher in obese subjects when compared to healthy subjects.

The levels of lipid profile parameters such as total cholesterol, HDL-C, LDL-C and triglycerides were found in normal ranges. The levels of HDL-C were lowest in the obese group and a statistically significant difference was observed between the levels of HDL-C between healthy and obese group and between overweight and obese groups. Nagila A et al., has also reported decreased levels of HDL-C in the obese group as compared to control group [26].

There is a low-grade inflammation as a result of expansion of adipose tissue, due to increase in production of pro-inflammatory molecules. Inflammatory biomarkers have been associated with cardiovascular events, out of which hs-CRP, an acute phase reactant appears to be the most promising. In the present study, a statistically significant difference was observed in the levels of hs-CRP among the three study groups. Overweight adults had significantly greater hs-CRP levels as compared to healthy adults. The elevated levels of hs-CRP indicate a state of low-grade inflammation in obese adults [27]. Previous studies have also shown similar results in overweight and obese adults. Gayathri B and Vinodhini VM, showed that hs-CRP levels were higher in overweight and obese individuals when compared with age and sex-matched controls [28]. Dev N and Marcus SR, also reported an increase in the levels of hs-CRP in the obese group [25]. To assess the cardiovascular risk, three hs-CRP cut-off points have been recommended by the American Heart Association and the Centres for Disease Control and Prevention. As per this recommendation, low risk if hs-CRP levels <1 mg/L, average risk if hs-CRP levels between 1 to 3 mg/L and high risk if hs-CRP levels >3 mg/L [29]. The present study revealed that 17 subjects of obese group had hs-CRP levels >3 mg/L as compared to eight subjects of healthy group and nine subjects of overweight group. On comparison of groups, a statistically significant difference was found in the distribution of subjects between all groups. On the basis of this finding, the present study suggests higher risk of cardiovascular diseases in obese subjects.

Observational analysis showed a strong, positive association between circulating CRP levels and BMI in the present study. Similar finding has been also reported by Gayathri B and Vinodhini VM, Lavanya K et al., Aronoson D et al., [28,30,31]. Another study conducted by Musso C et al., in obese and overweight adolescents and reported that high hs-CRP levels positively correlated with BMI [32].

Limitation(s)

The small sample size was the limitation of this study. Moreover, study of other pro-inflammatory molecules along with hs-CRP would have been more helpful in revealing the role of inflammation in obesity.

CONCLUSION(S)

In the present study, hs-CRP behaves as an independent marker which rises in response to inflammation seen in obesity. The increased levels of hs-CRP in overweight and obese group show that they are at the highest risk of CVD. Hence, they need to be followed for its future development. The lipid profile parameters were within normal range indicating that obese subjects in this study were not dyslipidaemic however, serum HDL level was significantly decreased in obese persons. A positive correlation was observed between BMI and hs-CRP levels indicating that serum CRP levels increase with rise in BMI in adults.

Declaration: I (Dr. Vaneet Kaur) declare that a part of this study was presented by me in the form of a poster during the APFCB Congress 2019 from 17-20 November, 2019, Jaipur, India.

REFERENCES

[1] Golay A, Yabarra J. Link between obesity and Type 2 diabetes. Best Pract Res Clin Endocrinol Metab. 2005;19:649-63.
[2] Ranjani H, Mehreen TS, Pradeepa R, Anjana RM, Garg R, Arvind K, et al. Epidemiology of childhood overweight and obesity in India: A systematic review. Indian J Med Res. 2016;143(2):160-74.
[3] Yudkin JS, Stehouwer C, Emeis J, Coppack S. C-reactive protein in healthy subjects: associations with obesity, insulin resistance and endothelial dysfunction: A potential role for cytokines originating from adipose tissue? Arteriosclerosis, Thromb Vascular Biol. 1999;19(4):972-78.
[4] Mohamed-Ali V, Goodrick S, Rawesh A, Katz DR, Miles JM, Yudkin JS, et al. Subcutaneous adipose tissue releases interlukin-6, but not tumor necrosis factor-α, in vivo. J Clin Endocrinol Metab. 1997;82(12):4196-200.
[5] Rizzo M, Rini G, Berneis K. Inflammation and atherosclerosis: Recent insights and future perspectives. Antinflamm Antiallergy Agents Med Chem. 2008;7:150-51.
[6] Rodriguez-Hernanandez H, Simental Mendia LE, Rodriguez-Ramirez G, Reyes-Romero MA. Obesity and inflammation. Epidemiology, risk factors, and markers of inflammation. Int J Endocrinol. 2013;01-11.
[7] Jialal I, Devaraj S. Role of C-reactive protein in the assessment of cardiovascular risk. Am J Cardiol. 2003;91:200-02.
[8] Dayal D, Jain H, Attri SV, Bharti B, Bhalla AK. Relationship of high sensitivity C-reactive protein levels to anthropometric and other metabolic parameters in Indian children with simple overweight and obesity. JCDR. 2014;8(8):PC05-08.
[9] Frohlich 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;23:1835-39.
[10] Hak AE, Stehouwer CD, Bots ML, Polderman KH, Schalkwijk CG, Westendorp IC, et al. Associations of C-reactive protein with measures of obesity, insulin resistance, and subclinical atherosclerosis in healthy, middle-aged women. Arterioscler Thromb Vasc Biol. 1999;19:1986-91.
[11] Chambers JC, Eda S, Bassett P, Karim Y, Thompson SG, Gallimore JR, et al. C-reactive protein, insulin resistance, central obesity, and coronary heart disease risk in Indian Asians from the United Kingdom compared with European whites. Circulation. 2001;104:145-50.
[12] Mendall MA, Patel P, Ballam L, Strachan D, Northfield TC. C reactive protein and its relation to cardiovascular risk factors: A population based cross sectional study. BMJ. 1996;312:1061-65.
[13] Lin CC, Kardia SLR, Li CI, Liu CS, Lai MM, Wen-Yuan Lin WY, et al. The relationship of high sensitivity C-reactive protein to percent body fat mass, body mass index, waist-to-hip ratio, and waist circumference in a Taiwanese population. BMC Public Health. 2010;10:579.
[14] Rothman KJ. BMI related errors in the measurement of obesity. Int J Obes (Lond). 2008;32 Suppl 3:S56-59.
[15] World Health Organisation (WHO). WHO expert consultation. Appropriate body-mass index for Asian populations and its implications for policy and intervention strategies. The Lancet. 2004;363:157-63.
[16] Mishra A, Chowbey P, Makkar BM, Vikram NK, Wasir JS, Chadha D, et al. Consensus statement for diagnosis of obesity, abdominal obesity and the metabolic syndrome for Asian Indians and recommendations for physical activity, medical and surgical management. J Assoc Physicians India. 2009;57:163-70.
[17] Raatikainen K, Heiskanen N, Heinonen S. Transition from overweight to obesity worsens pregnancy outcome in a BMI-dependent manner. Obesity (Silver Spring). 2006;14(1):165-71.
[18] Krakauer NY, Krakauer JC. An anthropometric risk index based on combining height, weight, waist, and hip measurements. Journal of Obesity. 2016;01-09.
[19] Wu A. Tietz Clinical Guide to Laboratory Tests. 4th ed. San Francisco CA (USA): Saunders Elsevier; 2006. 1856p.

[20] Allan CC, Poon LS, Chan CS, Richmond W, Fu PC. Enzymatic determination of total serum cholesterol. Clin Chem. 1974;20(4):470-75.
[21] Siedel J, Schmuck R, Staepels J. Long term stable liquid ready to use monoreagent for the enzymatic assay of serum or plasma triglycerides (GPO-PAP) method. AACC Meeting, Abstract 34. Clin Chem. 1993;39:1137.
[22] Matsuzaki Y, Kawaguchi E, Morita Y. Evaluation of two kinds of reagents for direct determination of HDL-cholesterol. J Anal Bio-Sc. 1996;9:419-27.
[23] Freidwald WT, Lev RI, Fredrickson DS. Estimation of the concentration of low density lipoproteins cholesterol in plasma, without the use of the preparative ultracentrifuge. Clin Chem. 1972;18(6):499-502.
[24] Price CP, Trull AK, Berry D, Gorman EG. Development and validation of a particle enhanced turbidimetric immunoassay for C- reactive protein. J Immunol Methods. 1987;99:205-11.
[25] Dev N, Marcus SR. High sensitive C- reactive protein, an independent and early novel inflammatory marker in healthy obese women. Biomed Res. 2012;23(1):73-77.
[26] Nagila A, Bhatt M, Podel B, Mahato P, Gurung D, Prajapati S, et al. Thyroid stimulating hormone and its correlation with lipid profile in obese Nepalese population. J Clin Diagn Res. 2008;8(2):932-37.
[27] Sartipy P, Loshkutoff DJ. Monocyte chemoattractant protein-1 in obesity and insulin resistance. Proc Natl Acad Sci USA. 2003;100(12):7265-70.
[28] Gayathri B, Vinodhini VM. High Sensitive C-reactive protein and its relationship with other cardiovascular risk variables in obese, overweight and healthy individuals. AJP Res. 2018;11(8):194-98.
[29] Pearson TA, Mensah GA, Alexander RW, Anderson JL, Cannon RO 3rd, Criqui M, et al. Markers of inflammation and cardiovascular disease. Application to clinical and public health practice. A statement for healthcare professionals from the centres for disease control and prevention and the American heart association. Circulation. 2003;107:499-511.
[30] Lavanya K, Ramamoorthi K, Acharya RV, Madhyasatha SP. Association between overweight, obesity in relation to serum hs- CRP levels in adults 20-70 years. J Clin Diagn Res. 2017;11(12):OC32-35.
[31] Aronoson D, Bartha P, Zinder O, Kerner A, Markiewicz W, Avizohar O, et al. Obesity is the major determinant of elevated C-reactive protein in subjects with the metabolic syndrome. Int J Obes Relat Metab Disord. 2004;28(5):674-79.
[32] Musso C, Graffigna M, Soutelo J, Honfi M, Ledesma L, Mikstztowicz V, et al. Cardometabolic risk factors as Apo lipoprotein B, triglyceride/HDL-cholesterol ratio and C-reactive protein, in adolescents with and without obesity: Cross-sectional study in middle class suburban children. Pediatr Diabetes. 2011;12:229-34.

PARTICULARS OF CONTRIBUTORS:
1. Associate Professor, Department of Biochemistry, Kalpana Chawla Government Medical College, Karnal, Haryana, India.
2. Professor, Department of Biochemistry, Kalpana Chawla Government Medical College, Karnal, Haryana, India.
3. Assistant Professor, Department of Biochemistry, Kalpana Chawla Government Medical College, Karnal, Haryana, India.
4. Demonstrator, Department of Medical Biochemistry, Gandhi Medical College, Bhopal, Madhya Pradesh, India.

NAME, ADDRESS, E-MAIL ID OF THE CORRESPONDING AUTHOR:
Dr. Pawan Kumar Kare,
Demonstrator, Department of Medical Biochemistry, Gandhi Medical College,
Bhopal, Madhya Pradesh, India.
E-mail: pawankare4@gmail.com

PLAGIARISM CHECKING METHODS: [Jain H et al.]
- Plagiarism X-checker: Dec 21, 2019
- Manual Googling: Feb 20, 2020
- iThenticate Software: Mar 20, 2020 (15%)

ETYMOLOGY: Author Origin

AUTHOR DECLARATION:
- Financial or Other Competing Interests: None
- Was Ethics Committee Approval obtained for this study? Yes
- Was informed consent obtained from the subjects involved in the study? Yes
- For any images presented appropriate consent has been obtained from the subjects. NA

Date of Submission: **Dec 18, 2019**
Date of Peer Review: **Feb 01, 2020**
Date of Acceptance: **Mar 05, 2020**
Date of Publishing: **Apr 01, 2020**