ORIGINAL ARTICLE

Role of C-peptide in Altered Lipid Profile among Apparently Healthy Adults of Vijayapura City, Karnataka

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Abstract:

Background: C-peptide is produced in equimolar concentration during insulin production as inactive molecule by beta islet cells of Langerhans. C-peptide is most useful biomarker of endogenous insulin production. *Aim and Objectives*: To predict metabolic syndrome in advance by estimation of C-peptide and lipid profile in healthy adults. *Material and Methods:* Serum C-peptide, fasting blood glucose and lipid profile of 128 healthy individuals were estimated. Adults in the age group of 18 to 60 years of both sexes were included in study. *Results*: C-peptide levels were increased in 27%, Serum cholesterol in 30%, LDL Cholesterol in 55% and triglyceride levels in 21% of healthy individuals. Significant correlation was observed between C peptide, age, serum cholesterol, LDL and cholesterol LDL ratio in male subjects only. In our study group most of the subjects (both males and females) fell in overweight group. *Conclusion*: C-peptide level and lipid profile may be considered as useful biomarkers to predict type 2 diabetes mellitus in advance, possibly due to insulin resistance.

Keywords: C-peptide, Insulin, Lipid Profile, Insulin Resistance

Introduction:

Normal insulin secretary function is essential for maintaining normal glucose tolerance and abnormal insulin secretion is invariably present in patients with Type 2 Diabetes Mellitus (T2DM). Quantification of beta cell function: measurement of peripheral insulin concentration by radio immune assay is still the most widely used method for quantifying beta cell function in vivo. Although this approach provides valuable information it is limited because 50-60% of insulin produced by pancreas is excreted by the liver without ever reaching systemic circulation [1, 2]. The standard radio immunoassay method is also unable to distinguish between endogenous and exogenous insulin making it ineffective as the measure of endogenous beta cell reserve in insulin treated diabetic patients. Anti-insulin antibodies that may be present in patients treated with insulin interfere with insulin radio immune assay making insulin measurements in insulin treated patients inaccurate. Conventional insulin radio immuno assays are also unable to distinguish between levels of circulating proinsulin and true levels of circulating insulin.

Insulin is derived from a single chain precursor proinsulin with Golgi apparatus of beta cell. Proinsulin is cleaved by convertases enzyme to form insulin, C-peptide and 2 pairs of basic amino acids. Insulin is subsequently released into circulation at concentration equimolar with those of C-peptide [3-5]. Unlike insulin C-peptide is not extracted by the liver and is excreted almost by the kidneys. Its plasma half life is approximately 30 minutes in contrast with that of insulin which has approximately 4 minutes [4, 6-8].

Because C-peptide is secreted in equimolar concentration with insulin and not excreted by the liver, many investigators have used C-peptide as a

marker of beta cell function. The concentration of plasma C-peptide as an index of beta cell function depends on critical assumptions that mean clearance rate of C-peptide is constant over the ranges of C-peptide levels under normal physiological conditions. This has been shown to be valid for dogs and humans [9, 10].

Association of obesity with Type 2 Diabetes Mellitus (T2DM) has been recognized for decades. Its close association between Insulin Resistance (IR) and obesity is seen in all ethnic groups and found across full range of body weights, all ages and both sexes. IR rises as body fat content increases. However central (intra abdominal) adiposity is more strongly linked to IR and to number of important metabolic variants that is Plasma Glucose (PG), Plasma Cholesterols, Triglycerides (TG) and decreased High Density Lipoproteins (HDL) than total adiposity [11-17].

In T2DM due to insulin resistance, compensatory increase and release of insulin can lead to beta cell damage. C-peptide test and lipid profile estimation may be used to evaluate persons having metabolic syndrome. It can be used in apparently healthy people to predict in advance about T2DM and IR. Hence current study was undertaken to evaluate relationship between C-peptide and lipid profile among healthy male and female adults.

Material and Methods:

Cross sectional study was conducted in 2013-14. One hundred and twenty eight apparently healthy subjects of age group between 18-60 years (teaching and nonteaching staff) of Shri B M Patil Medical College, BLDEU, Vijayapura were selected for the study. After explaining details of the study, informed written consent was obtained from each of the subject. With prevalence rate of diabetes mellitus especially T2DM 6-9% in Vijayapur, 95% confidence interval and acceptable error of ±5 margin calculated sample size was 125. Institutional Ethics Committee clearance was obtained. All apparently healthy adult individuals were included in the study and subjects giving history of DM, Ischemic Heart Disease (IHD), Chronic Obstructive Pulmonary Disease (COPD), Hypertension (HTN), tuberculosis, congenital heart diseases were excluded.

Estimation of Biochemical Parameters:

For lipid profile study, 3 ml of blood was collected from each subject after overnight fasting of 12 hours. Serum values of Total Cholesterol (TC), HDL, and TG were measured. TC, HDL and LDL were estimated by enzymatic cholesterol oxidase-CHOD PAP method. Estimation of TG was done by enzymatic GPO- POD method. Concentration was expressed in mg/dl. Estimation of FBS was done by using medical device namely Easy Glucometer. C-peptide estimation was done using chemiluminiscence assay method by kits.

Statistical Analysis:

Statistical analysis was done using SPSS. The results were expressed as Mean ± SD. $P<0.05$ was considered as statistically significant. ANOVA was done to see the intergroup difference in mean values of parameters. Correlation was done with Pearson correlation coefficient.

Results:

One hundred and twenty eight apparently healthy subjects were included in the study in which 71 were male and 57 were female subjects. Table 1 shows significant difference in C-peptide levels in overweight and obese subjects (p-0.04), also significant difference in parameters i.e. serum triglycerides, HDL cholesterol, VLDL cholesterol and cholesterol/HDL ratio. It also shows highly

significant in LDL cholesterol levels (0.009). Table 2 shows mean distribution in female subjects and no significant difference in all three (3) groups. Table 3 shows correlation study in male subjects and all three groups. It shows highly significant correlation between C-peptide and serum cholesterol, LDL cholesterol, cholesterol/ HDL ratio, LDL/HDL ratio, fasting blood glucose especially in overweight group. Table 4 shows correlation study in female subjects and all three groups. It shows no significant correlation among all three groups. Female subjects failed to show correlation.

Table 1: Mean distribution and ANOVA of Biochemical Parameters in Different Categories of Males

Characteristics	N (n=15)	OW(n=43)	OB(n=13)	p value
C-peptide (ng/mL)	1.4±0.8	1.7±0.8	2.2±0.7	0.04*
Serum Cholesterol (mg /dL)	166.8±38.3	181.2±32.3	194.2±25.3	0.092
Serum Triglycerides (mg/dL)	107.1±24.1	157.0±71.5	129.2±41.9	0.019*
HDL Cholesterol (mg/dL)	55.3±7.1	50.9±5.8	49.7±4.6	0.027*
LDL Cholesterol (mg/dL)	90.1±35	98.8±28.4	141.7±91.7	0.009**
VLDL Cholesterol (mg/dL)	21.4±4.8	31.4±14.3	25.8±8.3	0.019*
Cholesterol/HDL Ratio	3.1±0.6	3.5±0.7	3.8±0.9	0.02*
LDL/HDL Ratio	1.6±0.6	1.9±0.6	2.6±1.1	0.002**
Total Lipids (mg/dL)	440.1±84.9	518.7±119.4	488.3±127.6	0.078
FBS (mg/dL)	81.6±11.4	91.7±14	94.6±12.3	0.019*

*** Significant at the 0.01 level (2-tailed), * significant at the 0.05 level (2-tailed)*

N- Normal BMI; OW- Over Weight; OB- Obese

Table 2: Mean Distribution and ANOVA of Biochemical Parameters in Different Categories of Females

Characteristics	N (N=9)	OW(N=33)	OB(N=15)	p value
C-peptide (ng/mL)	1.5±0.5	1.5±0.7	1.4±0.6	0.815
Serum Cholesterol (mg /dL)	180.1±37.6	187.3±38.2	184.8±36.7	0.871
Serum Triglycerides (mg/dL)	136.5±77.7	134.2±38.6	133.6±37.4	0.988
HDL Cholesterol (mg/dL)	55.6±6.3	53.6±7.8	50.7±4	0.205
LDL Cholesterol (mg/dL)	97.1±31.2	106.8±35.3	107.3±36.2	0.735
VLDL Cholesterol (mg/dL)	27.3±15.5	26.8±7.7	26.7±7.4	0.988
Cholesterol/HDL Ratio	3.2±0.8	3.5±0.8	3.6±0.8	0.509
LDL/HDL Ratio	1.7±0.6	2.1±0.7	2.1±0.7	0.554
Total Lipids (mg/dL)	495.8±142.5	508.3±97	502.6±91.5	0.945
FBS (mg/dL)	88.3±21	88.4±10.2	087.3±7.8	0.953

N- Normal BMI; OW- Over Weight; OB- Obese

Table 3: Pearson Correlation(r) between C-peptide (ng/mL) and Other Biochemical Parameters among males

Characteristics	N (N=15)		OW(N=43)		OB(N=13)	
	r	p	r	p	r	p
Serum Cholesterol (mg /dL)	0.164	0.559	0.408	0.007**	0.369	0.214
Serum Triglycerides (mg/dL)	0.358	0.19	-0.073	0.642	0.075	0.807
HDL Cholesterol (mg/dL)	0.099	0.727	0.063	0.69	-0.492	0.087
LDL Cholesterol (mg/dL)	0.11	0.696	.489	0.001**	0.14	0.648
VLDL Cholesterol (mg/dL)	0.358	0.19	-0.073	0.642	0.075	0.807
Cholesterol/HDL Ratio	0.12	0.669	.303	0.048*	0.503	0.08
LDL/HDL Ratio	0.085	0.762	.369	0.015*	0.197	0.518
Total Lipids (mg/dL)	0.253	0.363	0.178	0.254	0.315	0.294
FBS (mg/dL)	.793**	0	.360	0.018*	0.128	0.677

***Correlation is significant at the 0.01 level (2-tailed), * Correlation is significant at the 0.05 level (2-tailed).*

N- Normal BMI; OW- Over Weight; OB- Obese

Table 4: Pearson Correlation(r) between C-peptide (ng/mL) and Other Biochemical Parameters among Females

Characteristics	N (N=9)		OW(N=33)		OB(N=15)	
	r	p	r	p	r	p
Serum Cholesterol (mg /dL)	0.221	0.568	0.274	0.123	-0.288	0.298
Serum Triglycerides (mg/dL)	-0.224	0.563	-0.103	0.568	-0.446	0.096
HDL Cholesterol (mg/dL)	0.038	0.922	0.154	0.394	0.451	0.091
LDL Cholesterol (mg/dL)	0.37	0.327	0.285	0.108	-0.251	0.367
VLDL Cholesterol (mg/dL)	-0.224	0.563	-0.103	0.568	-0.446	0.096
Cholesterol/HDL Ratio	0.208	0.592	0.198	0.269	-0.412	0.127
LDL/HDL Ratio	0.327	0.39	0.223	0.213	-0.333	0.225
Total Lipids (mg/dL)	-0.005	0.989	0.174	0.332	-0.413	0.126
FBS (mg/dL)	0.402	0.283	0.242	0.175	-0.251	0.368

***Correlation is significant at the 0.01 level (2-tailed), * Correlation is significant at the 0.05 level (2-tailed).*

N, normal BMI; OW, over weight; OB, obese

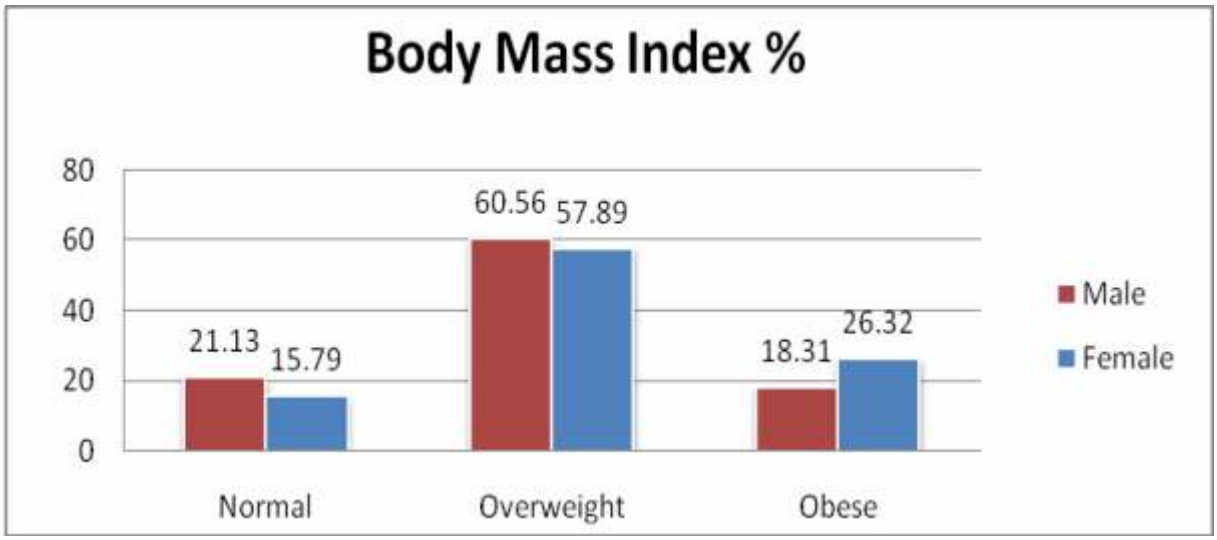

Fig.1 shows BMI % in Both Male and Female Groups

Figure 1 suggests that most of the subjects both male and female were in overweight group i.e. 60.56% males and 57.9% females. In normal range males predominate marginally. In obesity group, the female group predominated over male group.

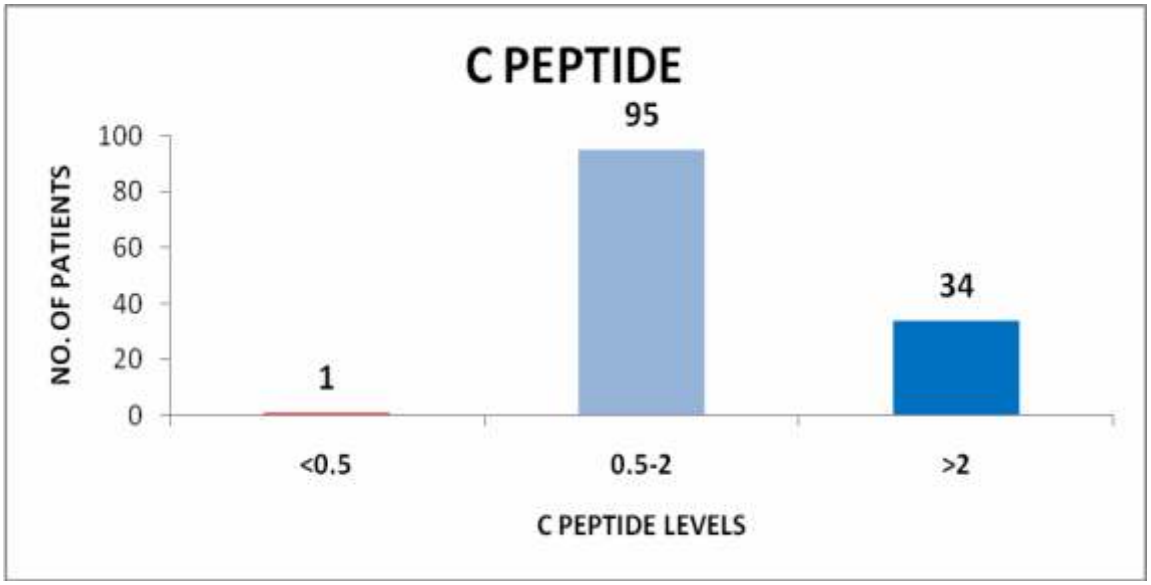

Fig. 2 shows Increased C-peptide Levels in 34 Subjects of Total 128 Subjects

Discussion:

Obesity is associated with many complications most important is being T2DM. In the present study attempt is made to predict possibility of developing T2DM in later life in healthy obese and non obese individuals. In our study we have observed significant dyslipidemia and increased serum C-peptide levels in apparently healthy individuals which suggest possible IR. As the age advances with modern life style people are proned to develop T2DM. Even in healthy subjects different biochemical parameters have shown changes. C-peptide levels also have shown increase suggestive of increased insulin secretion. These subjects are proned to develop T2DM in future due to IR.

Galiamov *et al* have found increased C-peptide biomarker of insulin suggestive of IR in 86 subjects (64 males and 22 females) with abdominal obesity. They have also concluded that IR could be one of the causes in pathogenesis of Chronic Kidney Disease (CKD) in obese people [18]. Lezhenko and Gladun observed increase levels of C-peptide in obese adolescents without rise in arterial blood pressure [19]. Svendsen *et al* in their study have shown that very low calorie diet (VLCD) for 8 weeks resulted in significant weight decrease and improvement in metabolic parameters including C-peptide. VLCD can improve insulin sensitivity, beta cell function and insulin clearance [20]. Further study reveals that insulin resistance is more in obese people. Abdominal fat had stronger relationship with insulin sensitivity than peripheral non abdominal fat even for those with BMI less than 25kg/m [21]. In the study of Mason *et al* observed that exercise with dietary weight loss can lead to decrease in insulin resistance with concomitant decrease in C-peptide levels of healthy postmenopausal women [22]. In our study insignificant change in serum C-peptide levels and lipid profile in female subjects might be due to hormonal support and postmenopausal healthy life style. In our study most of subjects were in overweight group and correlation was found in this group.

Limitations of the study:

This is a hospital based study. Systematic random sampling procedure was adapted to draw the study sample. A more representative random sample can be collected from community by taking equal number of normal, overweight and obese people in sample from the community.

Conclusion:

C-peptide is released along with insulin in equimolar concentration by beta cells of Langerhans. Unlike insulin molecule C-peptide is stable and half life is 30 minutes. It is excreted by kidneys and normal serum levels are 0.5-2.0ng/ml. Estimation of serum C-peptide may be used as a biomarker of endogenous insulin secretion and to predict the development of T2DM in future even in healthy subjects.

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References

1. Williams TB. Endocrinology, 12th edn. Saunders, VIII /31, 1392-93.
2. Yalow R, Berson S. Immunoassay of endogenous plasma insulin in man. *J Clin Invest* 1960; 39:1157-1175.
3. Steiner DF, James DE. Cellular and molecular biology of the beta cell. *Diabetologia* 1992; 35(suppl2):s41-s48
4. Melani F, Ryan WG, Rubenstein AH, Steiner DF. Proinsulin secretion by a pancreatic beta-cell adenoma. Proinsulin and C-peptide secretion. *N Engl J Med* 1970; 283(14):713-9.
5. Horwitz DL, Starr JI, Mako ME, Blackard WG, Rubenstein AH. Proinsulin, insulin, and C-peptide concentrations in human portal and peripheral blood. *J Clin Invest* 1975; 55(6):1278-83.
6. Polonsky KS, Pugh W, Jaspan JB, Cohen DM, Karrison T, Tager HS, *et al*. C-peptide and insulin secretion. Relationship between peripheral concentrations of C-peptide and insulin and their secretion rates in the dog. *J Clin Invest* 1984; 74(5):1821-9.
7. Bratusch-Marrain P, Wardhausl WK, Gasic S. Hepatic disposal of biosynthetic human insulin and porcine C-peptide in humans. *Metabolism* 1984; 33:151-157
8. Faber OK, Hagen C, Binder C, Markussen J, Naithani VK, Blix P Metal. Kinetics of human connecting peptide in normal and diabetic subjects. *J Clin Invest* 1978; 62(1):197-203.
9. Polonsky K, Jaspan J, Pugh W, Cohen D, Schneider M, Schwartz T. Metabolism of C-peptide in the dog. In vivo demonstration of the absence of hepatic extraction. *J Clin Invest* 1983; 72(3):1114-23.
10. Polonsky KS, Licinio-Paixao J, Given BD, Pugh W, Rue P, Galloway Jetal. Use of biosynthetic human C-peptide in the measurement of insulin secretion rates in normal volunteers and type I diabetic patients. *J Clin Invest* 1986; 77(1): 98-105.
11. Cefalu WT, Werbel S, Bell-Farrow AD, Terry JG, Wang ZQ, Opara EC. 3rd. Insulin resistance and fat patterning with aging: relationship to metabolic risk factors for cardiovascular disease. *Metabolism* 1998; 47(4):401-8.
12. Larsson B, Svärdsudd K, Welin L, Wilhelmsen L, Björntorp P, Tibblin G. Abdominal adipose tissue distribution, obesity, and risk of cardiovascular disease and death: 13 year follow up of participants in the study of men born in 1913. *Br Med J (Clin Res Ed)* 1984; 288(6428):1401-4.
13. Despres JP, Tremblay A, Perusse L. Abdominal adipose tissue, serum HDL-cholesterol: association independent from obesity and serum triglyceride concentration. *Int J Obes* 1988; 12:1-13.

14. Landin K, Krotkiewski M, Smith U. Importance of obesity for metabolic abnormalities associated with abdominal fat distribution. *Metabolism* 1989; 38:572-76.
15. Heitmann BL. The variation in blood lipid levels described by various measures of and abdominal obesity in Danish men and women aged 35-65 years. *Eur J Clin Nutr* 1992; 46:597-605
16. Reeder BA, Senthilselvan A, Després JP, Angel A, Liu L, Wang H *et al*. The association of cardiovascular disease risk factors with abdominal obesity in Canada. Canadian Heart Health Surveys Research Group. *CMAJ* 1997; 157 Suppl 1:S39-45.
17. Lamouche B, Abdominal obesity and its metabolic complications: implications for risk of IHD. *Coron Artery Dis* 1998; 9:473-481.
18. Galiamov MG, Saginova EA, Severova MM, Samokhodskaia LM, Krasnova TN, Sholomova VIetal. [Significance of the factors of hypoxia and endothelial dysfunction in kidney injury in the presence of obesity]. *Ter Arkh* 2013; 85(6):31-7.
19. Lazhenko G, Gladun E. The role of osteopontin and adoponectin in the development of insulin resistance and hypertension in obese adolescents. *Georgian Med News* 2013; (214): 43-8.
20. Svendsen PF, Jensen FK, Holst JJ, Hougaard SB, Nilas L, Madsabad S. The effect of a very low calorie diet, beta cell function, insulin clearance, incretin hormone secretion, androgen levels and body composition in young obese women. *Scand J Clin Lab Invest* 2012; 72(5):410-9.
21. David G, Arthur B Jenkins, Lesley V Campbell, Judith Freund, Donald J Chisholm. Abdominal fat and insulin resistance in normal and overweight women: Direct measurements reveal a strong relationship in subjects at both low and high risk of NIDDM. *Am J Prev Med* 2011;41(4):366-75
22. Mason C, Foster-Schubert KE, Imayama I, KongA, Xiao L, Bain C, *et al*. Dietary weight loss and exercise Carey effects on insulin resistance in postmenopausal women. *Nestle Nutr Inst Workshop Ser* 2013; 71:11-27.

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