

Effect of Body Mass Index on Serum Cystatin C Level in Healthy Subjects

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ABSTRACT

Background with aims and objectives: In last two decades, a multitude of clinical studies have investigated the role of cystatin C as a marker of kidney function. Cystatin C belongs to family 2 of the super family of cysteine protease inhibitors. It is produced in all the nucleated cells of the human body and its production rate is constant. Cystatin C is present in all human body fluids. The study was conducted to investigate the effect of Body Mass Index (BMI) on serum cystatin C in healthy population.

Study Design: Analytical cross-sectional.

Materials and Methods: Eighty five healthy subjects both males and female ages 18-60 years were included in this study group. These subjects were divided into three groups based on BMI as control, over weight and obese. Serum cystatin C was measured by ELISA.

Results: The serum cystatin C level was significantly high in over weight and obese groups as compared to control group in both males and females.

Conclusion: The present study suggests that BMI affects serum cystatin C level.

Key words: BMI, ELISA, GFR, Serum Cystatin C.

INTRODUCTION

Human Cystatin C is a cationic, low molecular weight (13,359 kd) cysteine protease inhibitor¹. It is encoded by the CST3 gene that is being ubiquitously expressed at moderate levels². Earlier reports revealed that primarily cells of the neuroendocrine system synthesize Cystatin C. Later on, cystatin C production from many other cells including macrophages and fibroblasts has also been reported. Therefore, the synthesis of cystatin C does not seem to be tissue-specific and all nucleated cells constitutively express and constantly secrete cystatin C³⁻⁵. However, few invitro studies reported that cystatin C production varies in response to various factors⁶. Consequently, it is expected that such endogenous conditions may also alter the synthesis of cystatin C among tissues⁷.

Cystatin C is an important extra- and transcellular cysteine protease inhibitor and its monomeric form is present in all human body fluids. It is especially abundant in Cerebrospinal fluid, seminal

plasma, milk, synovial fluid, saliva, tears, urine and blood plasma⁷. The concentration of serum cystatin C in healthy adult individuals ranges around 0.8-1.2 mg/l depending upon the analytical method⁸⁻¹².

Body mass index (BMI) is a measure of adiposity obtained from the weight in kilograms divided by the square of the height in meters. Obesity is accompanied by hypertrophy and hyperplasia of adipocytes.¹³ The adipocytes not only serve as a site for storage of triacylglycerol, but are also capable of producing and secreting a number of proteins including cystatin C, that affect diverse physiological processes^{13,14}.

In last two decades, a multitude of clinical studies have evaluated the role of cystatin C as a marker of kidney function and proposed that increased serum levels are almost exclusively associated with a reduction in Glomerular filtration rate.^{11, 15-17} A number of studies reported that Cystatin C does not appear to be affected by age, gender, diet, lean body mass, body fat content, inflammatory processes, liver functions or malignancies. Consequently, it was claimed that serum cystatin C levels as renal function marker is superior to serum creatinine and close to inulin clearance^{8, 17-20}.

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As there have been inconsistent reports regarding the effect of BMI on serum cystatin C levels, the present study was focused on the effect of BMI on serum cystatin C levels in healthy individuals in order to avoid interference by other factors.

MATERIALS AND METHODS

This is analytical cross sectional study. Eighty five (85) healthy subjects, both males and female (ages 18-60 years) were included in the study. All healthy subjects with serum creatinine <1.5mg/dl in males and <1.3mg/dl in females were included in the study. We excluded pregnant women, patients suffering from renal diseases, hypertension, diabetes mellitus and other systemic diseases.

Data collection procedure: Informed consent of all the study subjects was obtained and relevant history was recorded in proformas. Height was measured in centimetre (cm) on a standard height scale and weight was measured in kilograms on Camry weight scale. Body mass index was calculated as weight (kilogram) divided by square of height (meters squared).

The study subjects were divided into three groups according to their BMI:¹³

1. Controls: BMI 20-24.9 kg/m²
2. Over weight: BMI 25-29.9 kg/m²
3. Obese: BMI >30 kg/m²

The human cystatin C enzyme linked immunosorbant assay (ELISA) test kit by BioVendor Germany, was used for the quantitative measurement of human cystatin C in serum.

Statistical Analysis: Mean (X), Standard deviation (SD), Standard error of mean (SEM) were calculated manually by using scientific calculator. Student “t” test was used for the comparison of group means. *p*-value less than 0.05 (5%) was regarded as significant.

RESULTS

Distribution of males and females in different groups is given in figure 1. Mean serum cystatin C levels in male control, overweight and obese group was 1.39±0.10, 2.42±0.08 and 3.15±0.08 mg/L respectively and was highly significantly (*p* <0.01) higher in overweight and obese group as compared to controls. In obese group cystatin C level was highly significantly (*p* <0.01) higher than overweight groups as well (Table 1).

Serum cystatin C level in control, overweight and obese female groups was 1.19±0.10, 2.27±0.07 and 2.85±0.05 mg/L respectively and was highly significantly (*p* <0.01) higher in overweight and obese groups as compared to control. In obese group

serum cystatin C level was highly significantly (*p* <0.01) higher than overweight group (Table 1).

Figure 1: Distribution of males and females in different groups

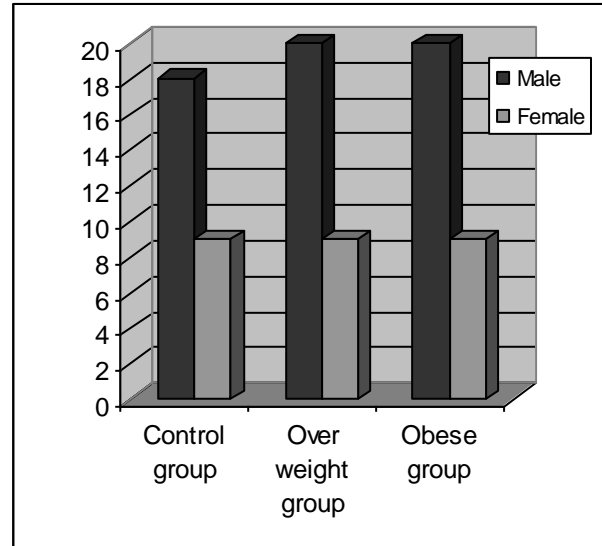


Table 1: Mean ± SEM of Serum Cystatin C levels (mg/L) is given separately for male and female subgroups.

Group	Male (Mean±SEM) S. Cystatin C (mg/L)	Female Mean ±SEM S. Cystatin C (mg/L)
Control	*1.39± 0.10	*1.19± 0.10
Overweight	2.42± 0.08	2.27± 0.07
Obese	3.15**± 0.08	2.85** ± 0.05

* *P*<0.01 highly significantly lower as compared to overweight and obese

***P*<0.01 highly significantly higher as compared to overweight

DISCUSSION

Although the validity of serum cystatin C as a more sensitive GFR marker has been extensively studied, the effect of obesity on serum cystatin C levels needs further research.

Schuck et al. reported that serum Cystatin C is independent of BMI. The major limitation of this study was inclusion of obese and non obese chronic renal disease (CRD) patients and relative contribution of CRD and higher BMI to raised serum cystatin C in individual subjects could not be distinguished²⁰.

However, there are a number of contrary reports that serum cystatin C levels do appear to be influenced by multiple factors including, male sex, greater weight and height, smoking and higher C-reactive protein levels^{22,23}.

The present study evaluated the effect of BMI on serum cystatin C levels in healthy individuals. As there have been studies reporting elevated levels of serum cystatin C in various pathological conditions like renal disease, hypertension and diabetes, we excluded subjects suffering from these conditions. Moreover, we also excluded pregnant women as there have been studies reporting alteration of serum cystatin C in normal and complicated pregnancies⁷.

The results of the present study reveal that the serum cystatin C level is significantly high in over weight and obese groups as compared to control group, both in males and females, suggesting an effect of BMI. As cystatin C is known to be produced in all nucleated cells including adipocytes, overweight and obese individuals are likely to have higher serum cystatin C level.^{3-5,14} A moderate but biologically insignificant correlation between BMI and cystatin C has been reported.²³ Our results are generally in agreement with Knight et al. who have reported that serum cystatin C levels do appear to be influenced by multiple factors including greater weight and height.²² Recently, Al Wakeel JS et al. and Muntner P et al. have also reported a significant correlation between serum cystatin C and BMI.^{24,25}

CONCLUSION

The present study suggests that BMI affects serum cystatin C level.

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REFERENCES

1. Grubb A. Diagnostic value of analysis of cystatin C and protein HC in biological fluids. *Clin Nephrol* 1992; 38: S20-7.
2. Abrahamson M, Olafsson I, Palsdottir A, Ulvsback M, Lundwall A, Jensson O et al. Structure and expression of the human cystatin C gene. *Biochem J* 1990; 268: 287-94.
3. Grubb A, Lofberg H. Human gamma trace, a basic microprotein: Aminoacid sequence and presence in the adenohypophysis. *Proc Natl Acad Sci USA* 1982; 79: 3024-27.
4. Abrahamson M. Molecular basis for amyloidosis related to hereditary brain hemorrhage. *Scand J Clin Lab Invest Suppl* 1996;226:47-56.
5. Chapman HA Jr, Reilly JJ Jr, Yee R, Grubb A. Identification of cystatin C, a cysteine proteinase inhibitor, as a major secretory product of human alveolar macrophages in vitro. *Am Rev Respir Dis* 1990;141:698-705.
6. Solem M, Rawson C, Lindburg K, Barnes D. Transforming growth factor beta regulates cystatin C in serum-free mouse embryo (SFME) cells. *Biochem Biophys Res Commun* 1990;172:945-51.
7. Mussap M, Plebani M. Biochemistry and clinical role of human cystatin C. *Crit Rev Clin Lab Sci* 2004; 41: 467-550.
8. Norlund L, Fex G, Lanke J, VonSchenck H, Nilsson JE, Leksell H, Grubb A. Reference intervals for the glomerular filtration rate and cell proliferation markers: serum cystatin C and serum beta 2-microglobulin/cystatin C ratio. *Scand J Clin Lab Invest* 1997; 57: 463-70.
9. Finney H, Newman DJ, Thakkar H, Fell JME, Price CP. Reference ranges for plasma cystatin C and creatinine measurements in premature infants, neonates and older children. *Arch Dis Child* 2000; 16: 463-64.
10. Tian S, Kusano E, Ohara T et al. Cystatin C measurement and its practical use in patients with various renal diseases. *Clin Nephrol* 1997; 48: 104-8.
11. Kyhse-Andersen J, Schmedt C, Nordin G, Andersson B, Nilsson ehle P, Lindstrom V et al. Serum cystatin C, determined by a rapid, automated particle-enhanced turbidimetric method, is a better marker than serum creatinine for glomerular filtration rate. *Clin Chem* 1994; 40: 1921-26.
12. Uhlmann E, Hock KG, Issitt C, Sneeringer MR, Groman RT et al. Reference intervals for plasma cystatin C in healthy volunteers and renal patients, as measured by the Dade Behring BN II system, and correlation with creatinine. *Clin Chem* 2001; 47: 2031-33.
13. Devlin TM, editor. Text book of biochemistry with clinical correlations. 6th ed. USA: Wiley Liss;2006.
14. Kratchmarova I, Kalume DE, Blagoev B, Scherer PE, Podtelejnikov AV, Molina H, et al. A Proteomic Approach for Identification of Secreted Proteins During the Differentiation of 3T3-L1 Preadipocytes to Adipocytes. *Mol Cell Proteomics* 2002;1:213-222.
15. Newman DJ, Thakkar H, Edwards RG et al. Serum cystatin C measured by automated immunoassay: A more sensitive marker of changes in GFR than serum creatinine. *Kidney Int* 1995; 47: 312-18.
16. Grubb A, Simonsen O, Sturfelt G, Truedsson L, Thysell H. Serum concentration of cystatin C, factor D and beta 2-microglobulin as a measure of glomerular filtration rate. *Acta med Scand* 1985; 218: 499-503.
17. Randers E, Kristensen JH, Erlandsen EJ, Danielsen H. Serum cystatin C as a marker of the renal function. *Scand J Clin Lab Invest* 1998; 58: 585-92.
18. Simonsen O, Grubb A, Thysell H. The blood serum concentration of cystatin C (gamma - trace) as a measure of the glomerular filtration rate. *Scand J Clin Lab Invest* 1985; 45: 97-101.
19. Vinge E, Lindergard B, Nilsson-Ehle P, Grubb A. Relationships among serum cystatin C, serum creatinine, lean tissue mass and glomerular filtration

- rate in healthy adults. *Scand J Clin Lab Invest* 1999; 59: 587-92.
20. Schuck O, Teplan V, Stolova M, Skibova J. Estimation of glomerular filtration rate in obese patients with chronic renal impairment based on serum cystatin C levels. *Clin Nephrol* 2004; 62: 92-96.
 21. Burtis CA, Ashwood ER, Bruns DE, editors. *Tietz text book of clinical chemistry and molecular diagnostics*. 4th ed. India; Saunders: 2006.
 22. Knight EL, Verhane JC, Spiegelman D, Hillege HL, deZeeuw D, Curhan GC, deJong PE. Factors influencing serum cystatin C levels other than renal function and the impact on renal function measurement. *Kidney Int* 2004; 37: 210-216.
 23. Galteau MM, Guyon M, Gueguen R, Siest G. Determination of Serum Cystatin C: biological variation and reference values. *Clin Chem Lab Med* 2001; 39: 850-857.
 24. Al Wakeel JS, Memon NA, Chaudhary AR, Mitwali AH, Tarif N, Isnani A, et al. Normal Reference Level of Serum Cystatin C in Saudi Adults. *Saudi J Kidney Dis Transpl* 2008; 19(3):361-370.
 25. Muntner P, Winston J, Uribarri J, Mann D, Fox CS. Overweight, Obesity, and Elevated Serum Cystatin C Levels in Adults in the United States. *Am J Med* 2008;121(4):341-348.