Study of the Association of Serum Testosterone Levels With Metabolic Syndrome in Middle Aged Male Subjects

Brahmanandam Lingudu[1], Vivekanand Bongi[2], Subrahmanyam KAV[3], Mounika Anitha[4] and Johann Varghese[5]

[1]Associate Professor of endocrinology, government medical college, Ananthapuram, Andhra Pradesh India.
[2]Associate Professor of endocrinology, Andhra medical college, Visakhapatnam Andhra Pradesh India
[3]Professor of Endocrinology, Andhra medical college, Visakhapatnam Visakhapatnam Andhra Pradesh India
[4]Assistant Professor of Endocrinology, Andhra Medical College, Visakhapatnam, Andhra Pradesh India
[5]Junior Resident, Department Of Endocrinology,Andhra Medical College, Visakhapatnam, Andhra Pradesh India
Corresponding author email: brahmaendo2k2@gmail.com

ABSTRACT

Background: Lower testosterone levels and metabolic syndrome with central obesity have been shown to be independently associated with increased cardiovascular mortality in men.

Objective: to study the association of serum testosterone levels with MS and additionally to check for the association of the use ofstatins with serum testosterone levels.

Methodology: Observational, cross-sectional, single-centre study amongst men aged between 30 to 65 years. Consecutive male patients attending the endocrinology outpatient department were recruited during the study period after appropriate inclusion and exclusion criteria were satisfied. Physical examination with fasting serum samples were collected for the necessary biochemical evaluations.

Results: Serum testosterone levels were significantly associated with the presence of metabolic syndrome ($p=0.001$). Individual components of the metabolic syndrome like waist circumference ($p<0.01$), systolic Bp ($p<0.01$) and diastolic BP ($p<0.01$) were also significantly associated with serum testosterone levels with a negative correlation. Statin usage was also significantly associated with lower serum testosterone levels ($p<0.01$). Additionally, sedentary lifestyle ($p<0.01$) and erectile dysfunction ($p<0.01$) were also significantly associated with lower serum testosterone levels.

Conclusions: Lower serum testosterone levels are present in those with metabolic syndrome and could be considered as an additional risk-enhancing factor in atherosclerotic cardiovascular disease.

Keywords- Metabolic syndrome, testosterone, erectile dysfunction, statins, overweight, triglycerides

Introduction

Metabolic syndrome (MS) is an entity characterized by insulin resistance, abdominal obesity, dyslipidemia and hypertension. It has been proposed that serum testosterone may influence the development of MS.[1] Adverse metabolic effects on blood pressure, lipid and glucose metabolism are mediated by insulin resistance. Metabolic syndrome is ominously associated with increased cardiovascular morbidity and mortality. First described by Kylin in 1923and followed in 1988 by Reaven who described 'Syndrome X' as a constellation of hyperglycemia, hypertension, insulin resistance, low HDL and raised TG levels and VLDL. [2]

Table 1- Metabolic Syndrome currently is defined as [3]

Central obesity (defined as waist circumference with ethnicity specific values) **plus** any two of the following four factors:	
Raised triglycerides	≥ 150 mg/dL (1.7 mmol/L) **or specific treatment for this lipid abnormality**
Reduced HDL cholesterol	< 40 mg/dL (1.03 mmol/L) in males < 50 mg/dL (1.29 mmol/L) in females **or specific treatment for this lipid abnormality**
Raised blood pressure	systolic BP ≥ 130 or diastolic BP ≥ 85 mm Hg **or treatment of previously diagnosed hypertension**
Raised fasting plasma glucose	(FPG) ≥ 100 mg/dL (5.6 mmol/L), **or previously diagnosed type 2 diabetes** If above 5.6 mmol/L or 100 mg/dL, OGTT is strongly recommended but is not necessary to define presence of the syndrome.

Visceral adipocytes due to their location have a higher metabolic activity as compared to subcutaneous adipocytes and constitute a rich source of adipocytokines and free fatty acids to the liver, increasing hepatic insulin resistance and favoring gluconeogenesis. Thus, central obesity, measured clinically via the waist circumference is strongly implicated with insulin resistance and atherosclerotic cardiovascular disease.[4] Low testosterone has been demonstrated to be independently associated withthe development of metabolic syndrome, T2DM and also a risk factor for cardiovascular outcomes.[5] The hypothesis that greater visceral fat accumulation occurs in states of lower testosterone levels has been confirmed by CT and MRI.[4] Testosterone is known to facilitate myocyte but deters adipocyte development from stem

cells.[6] Via beta-adrenergic receptors it stimulates lipolysis but decreases fatty acid synthesis. [7] Further evidence on the relationship between testosterone levels and MS or its components in Asian populations is required. There are very few studies fromIndia which have studied the association of metabolic syndrome and serum testosterone levels. Hence this study was undertaken to study the association of serum testosterone levels with MS and additionally to check for the association of the use of statins with serum testosterone levels.

Materials and Methods

This is an observational, cross-sectional, single-centre study done at King George Hospital(KGH), Visakhapatnam from April 2020 – September 2021. Consecutive male patients attending the endocrinology outpatient department were recruited during the study period.

Inclusion Criteria: Men between 30 years to 65 years of age.

Exclusion Criteria:

No consent, Immunocomprimised individuals

Methodology-

Institutional ethics committee clearance from Andhra Medical College/KGH was taken. Information was collected through prepared proforma from each patient, who met the inclusion criteria, after receiving their consent and who were attending the outpatient department of endocrinology at KGH. Subjects were categorized into moderate/vigorous physical activity or sedentary activity depending on whether the subjects engaged in activities such as either walking for atleast 30 minutes per day, or other activities like jogging or running, swimming, gardening and lifting weights. Height, weight, BMI and waist circumference were measured using standard techniques. Blood pressure was measured in the supine position with the cuff applied to the right arm. Fasting(>8 hours) blood samples were obtained for assessment of routine parameters such as complete blood picture with ESR, blood urea, creatinine, liver function tests and metabolic parameters such as FPG and lipid profile. Serum was separated from the same blood sample after cold centrifugation at 2 degrees centigrade for 10 minutes at 2000 rpm and stored at -70 degrees centigrade for assessment of 8 am serum total testosterone levels. FPG was assessed using the glucose oxidase method. Total cholesterol, Tg, and HDL levels were measured using enzymatic methods via a chemistry autoanalyzer. Serum low-density lipoprotein level was calculated : LDL (mg/dl) =Total cholesterol — (Triglycerides /5) -HDL . Total testosterone levels were quantified using Chemiluminescence Immuno Assay kit (Roche e411).

Statistical analysis

Continuous variables were expressed as appropriate means ± standard deviations. Categorical variables were summarized as frequencies and percentages. To test the differences amongst the participants with and without MS, the Student t test for continuous variables was used and Chi-square test was used for qualitative variables. Pearson's correlation coefficients were calculated between the individual quantitative components of metabolic syndrome and testosterone levels. A value of $P< 0.05$ was considered statistically significant. The data was analyzed using IBM SPSS (Version 21.0)

Results

Table 1: Descriptive statistics of the study population

Parameter	Mean +/- SD	Range
Age (years)	50.33 +/- 9.4	30-65
BMI (kg/m2)	26.4 +/- 3.4	17-40.7
Waist circumference (cm)	90.17 +/- 5.5	78-105
Serum testosterone level (ng/dl)	463.25 +/- 92.2	193-700
Systolic BP (mm Hg)	130.5 +/- 14.7	90-100
Diastolic BP (mm Hg)	80.8 +/- 9	60-110
Fasting plasma glucose (mg/dl)	152.4 +/- 51.5	81-396
Total Cholesterol (mg/dl)	193.3 +/- 32.75	120-267

HDL (mg/dl)	39.9 +/- 7.53	21-56
LDL (mg/dl)	122+/- 25.87	36-184
Triglycerides (mg/dl)	155 +/- 53.6	53-335

As per table 1 demographic and clinical variable are studies. The mean age was 50.33 years, mean BMI 26.4, serum testosterone was 463, mean SBP/DBP was 130.5/80.8. Fasting plasma glucose was 152.4 with mean serum triglyceride was 155.

TABLE 3: COMPONENTS OF METABOLIC SYNDROME

Variable	Number of patients	Percentage amongst the population studied (n =168)
Waist circumference(>90cm)	78	46%
SBP>/= 130 mm hg	94	55%
DBP>/=85 mm hg	50	29.7%
Fasting plasma glucose >/=100mg/dl	150	89%
HDL </= 40 mg/dl	72	42.8%
Tg >/= 150 mg/dl	84	50%

As per table 3 components of MS was studied. 46% had high waist circumference,55% had high SBP, 89% had high fasting sugar, 50% had high triglycerides.

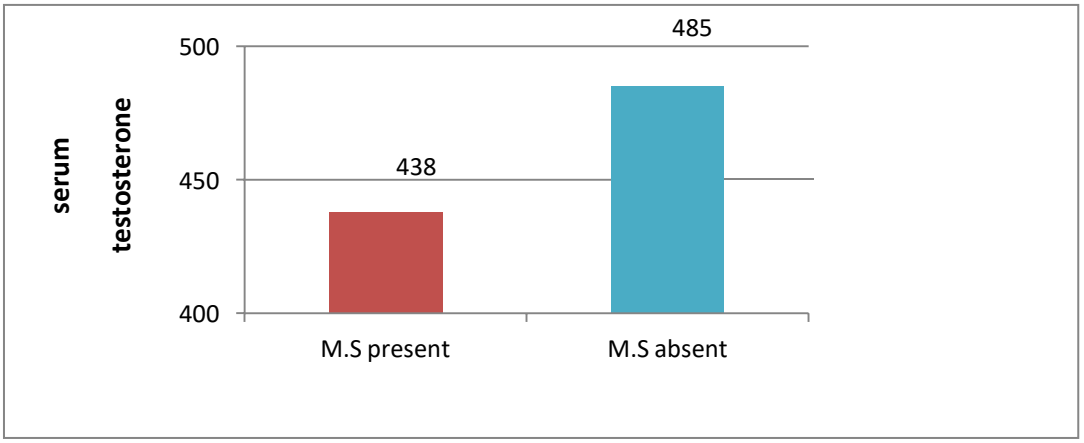
Table 4: Correlation studies of the variables with serum testosterone level

VARIABLE	Pearson correlation coefficient value (r)	p value*
AGE	**-0.49**	**<0.01**
BMI	**-0.161**	**0.037**
WAIST CIRCUMFERENCE	**-0.393**	**<0.01**
TOTAL CHOLESTEROL	0.017	0.825
TRIGLYCERIDE	-0.003	0.96
HDL	0.131	0.09
SYSTOLIC BP	**-0.287**	**<0.01**
DIASTOLIC BP	**-0.299**	**<0.01**
FASTING PLASMA GLUCOSE	-0.09	0.245

p* = significant when <0.05

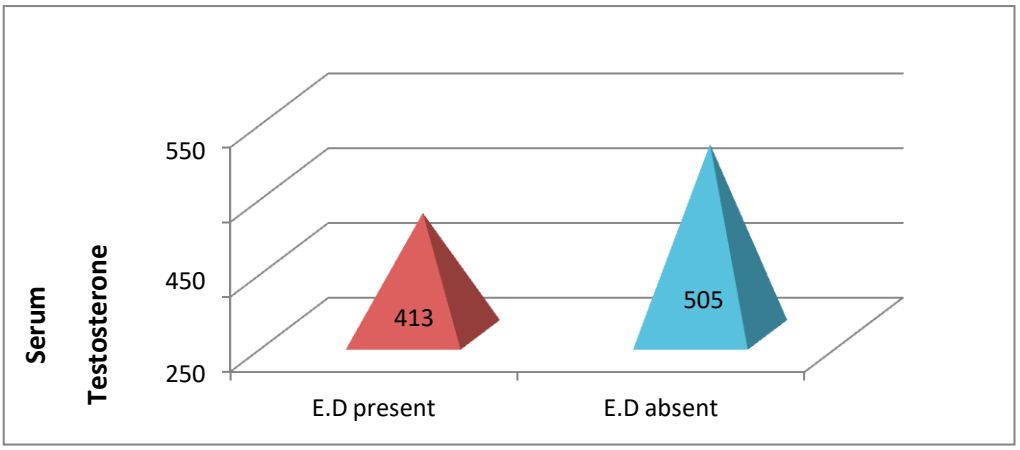
As per table 4 there was a statistically significant negative correlation between serum testosterone levels and age of the patients. On dividing the serum testosterone levels into quartiles, the mean age of the patientsin the lowest quartile was 55.8 years, the mean age of the patients in the second quartile was 53.4 years, the mean age in the 3rd quartile was 50.1 years while the mean age in the 4th quartile was 41.8 years. There was a statistically significant negative correlation between serum testosterone levels and BMI.

Figure 1 Mean serum testosterone levels among those subjects with and without metabolic syndrome



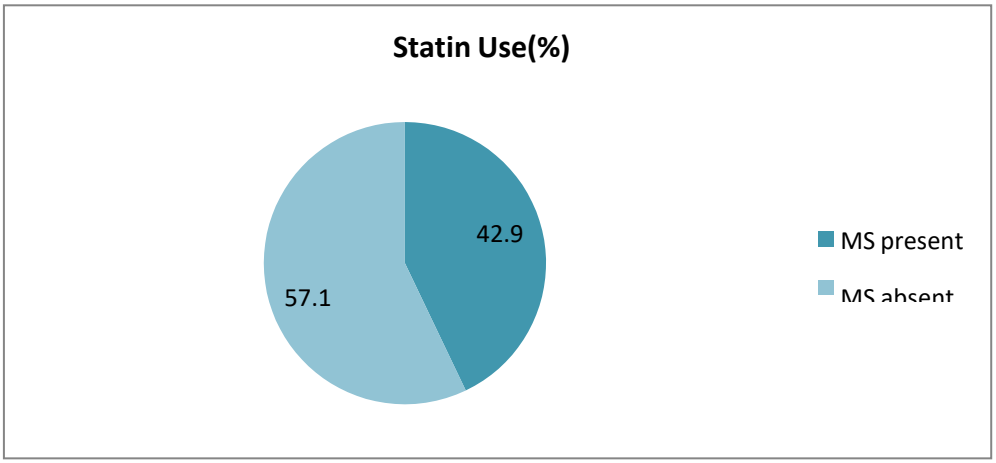
The mean serum testosterone among patients with metabolic syndrome was 438ng/dl whereas the mean serum testosterone among the patients without metabolic syndrome was 485ng/dl. This difference was found to be highly statistically significant. (p=0.001)

Figure 2: Mean serum testosterone levels amongst subjects with andwithout erectile dysfunction



Among the patients with erectile dysfunction mean serum testosterone was 413ng/dlwhereas among patients without erectile dysfunction the mean serum testosteroneas 505ng/dl. This difference was found to be highly statistically significant. **(p=<0.01)** (Figure 18a) Among patients belonging to the lowest quartile of serum testosterone (Q1), 79.5% had ED, similarly in second quartile was 55% in third quartile was 39.02% and in the highest quartile (Q4) only 9.3% of the patients had ED. Thus the presence of ED was maximum among patients with the lowest serum testosterone levels. This difference between the groups was also highly statisticallysignificant. **(p<0.01)**

Figure 3: Percentage of statin users amongst those with and withoutmetabolic syndrome



37.5% of the study population was on statin therapy of which 42.9% of the patients had metabolic syndrome and 57.1% of the users did not have metabolic syndrome.The presence of metabolic syndrome did not differ significantly between the statin users and non users. **(p= 0.52)**

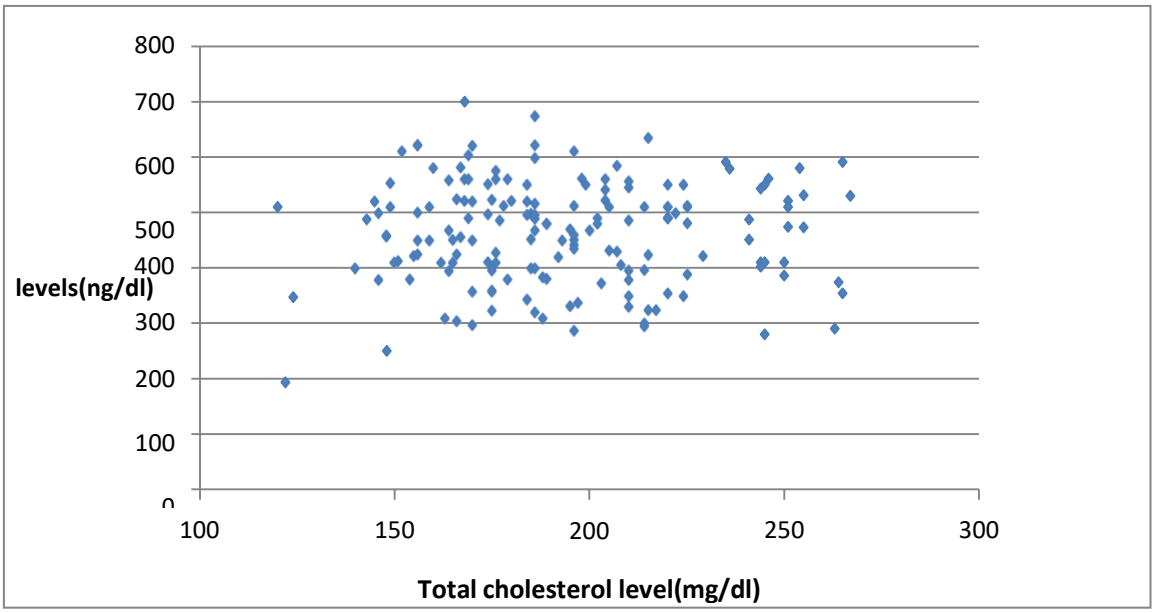


Figure 4: Scatter plot of serum testosterone levels with serum cholesterollevels

There was no significant correlation between serum testosterone and Total cholesterol(p=0.82), LDL(p=0.96).

Discussion

Apart from the fact that metabolic syndrome is associated with cardiovascular disease risk in recent times, it has been demonstrated that hypogonadism is also many times associated with metabolic syndrome and thus a novel term of hypoandrogen-metabolic syndrome has been proposed.[8] AACE recommends screening of all those with obesity or larger waistlines for signs and symptoms of hypogonadism. If the screen is positive a formallaboratory confirmation of serum testosterone is advised.[9] Both low testosterone and metabolic syndrome can be managed with weight loss, with increments in serum testosterone levels noted and thus lifestyle modifications prioritizing weight reduction should be the first step.[10] Weight loss via bariatric

surgery has also been shown to raise serum testosterone levels.[11] Bhattacharya et al. reported that testosterone replacement therapy increased serum testosterone levels, lowered glucose levels, decreased waist circumference, and decreased blood pressure only in those MS patients with hypogonadism but not in those who did not have MS.[12] In our study, the age range was fixed at between thirty to sixty-five years taking into account the fact that with increasing age total testosterone level falls asdemonstrated by Feldman et al in their longitudinal study [13]

In the present study, the IDF criteria was used to define MS as the criteria which has more applicability considering the ethnicity-specific central obesity cut- off values for the South Asian population as was also used by Singh et al in their study.[14]

The present study demonstrated significantly higher serum testosterone levels in those with moderate to vigorous physical activity compared to those with sedentary activity. Similarly, Shiels et al demonstrated that men practicing a higher degree of physical activity had higher concentrations of total testosterone.[15]

In a separate study from India erectile dysfunction was present in 47% of the hypogonadal patients compared to 42% amongst eugonadal subjects but this difference was not found to be statistically significant.[16] S Ghazi et al showed that ED was detected in 94.4% in the hypogonadal group whereas only in 61.0% amongst the eugonadal subjects which was statistically significant. Amongst subjects with diabetes mellitus and erectile dysfunction testosterone levels were 392.4ng/dl versus 524.3 ng/dl amongst those with normal erectile function.[17]

Bansal et al reported that MS was present in 43% of their ED population as compared to 24% amongst those without erectile dysfunction.[18] The postulated mechanisms being uncoupling of eNOS and disturbances in the NO cascade due to reactive oxygen species generated as a result of mitochondrial dysfunction commonly seen in insulin-resistant states such as obesity, MS and diabetes mellitus may contribute to erectile dysfunction.[19]

Conclusions

Lower serum testosterone levels were significantly associated with thepresence of metabolic syndrome in middle-aged male subjects. Parameters like mean age, BMI, physical activity, erectile dysfunction also additionally were significantly associated with serum testosterone levels. Thus this study suggests that lower serum testosterone could be considered as an additional significant parameter in metabolic syndromeand could be used as a risk-enhancing factor in atherosclerotic cardiovascular disease. This study acts as a primer for future studies in which testosterone replacement therapy may be considered in those with metabolic syndrome and lower serum testosterone levels for favorable outcomes.

References

1. Liang J, Peng Q, Yang X, Yang C. The association between serum testosterone levels and metabolic syndrome among women. Diabetology & Metabolic Syndrome. 2021 Dec;13(1):1-9.
2. Reaven GM. Role of insulin resistance in human disease. Diabetes. 1988 Dec 1;37(12):1595-607.
3. IDF Epidemiology Task Force Consensus Group. International Diabetes Federation: The IDF consensus worldwide definition of the metabolic syndrome.
4. Muraleedharan V, Jones TH. Testosterone and the metabolic syndrome. Therapeuticadvances in endocrinology and metabolism. 2010 Dec;1(5):207-23.
5. Jones TH. Testosterone deficiency: a risk factor for cardiovascular disease?. Trends in Endocrinology & Metabolism. 2010 Aug 1;21(8):496-503.
6. Singh R, Artaza JN, Taylor WE, Gonzalez-Cadavid NF, Bhasin S. Androgens stimulate myogenic differentiation and inhibit adipogenesis in C3H 10T1/2 pluripotentcells through an androgen receptor-mediated pathway. Endocrinology. 2003 Nov 1;144(11):5081-8.
7. De Pergola G, Pannacciulli N, Ciccone M, Tartagni M, Rizzon P, Giorgino R. Free testosterone plasma levels are negatively associated with the intima-media thickness of the common carotid artery in overweight and obese glucose-tolerant young adult men. International journal of obesity. 2003 Jul;27(7):803-7.
8. Gould DC, Kirby RS, Amoroso P. Hypoandrogen-metabolic syndrome: a potentially common and underdiagnosed condition in men. International journal of clinical practice. 2007 Feb;61(2):341-4.
9. Garvey WT, Mechanick JI, Brett EM, Garber AJ, Hurley DL, Jastreboff AM, Nadolsky K, Pessah-Pollack R, Plodkowski R. American association of clinical endocrinologists and American college of endocrinology comprehensive clinical practice guidelines formedical care of patients with obesity. Endocrine Practice. 2016Jul 1;22:1-203.

10. Camacho E, Huhtaniemi IT, O'neill TW, Finn JD, Pye SR, Lee DM, Tajar A, Bartfai G, Boonen S, Casanueva FF, Forti G. Age-associated changes in hypothalamic– pituitary–testicular function in middle-aged and older men are modified by weight change and lifestyle factors: longitudinal results from the European Male Ageing Study. European journal of endocrinology. 2013 Mar 1;168(3):445-55.
11. Boonchaya-Anant P, Laichuthai N, Suwannasrisuk P, Houngngam N, Udomsawaengsup S, Snabboon T. Changes in testosterone levels and sex hormone-binding globulin levels in extremely obese men after bariatric surgery. International journal of endocrinology. 2016 May 13;2016.
12. Bhattacharya RK, Khera M, Blick G, Kushner H, Nguyen D, Miner MM. Effect of 12 months of testosterone replacement therapy on metabolic syndrome components in hypogonadal men: data from the Testim Registry in the US (TRiUS). BMC endocrine disorders. 2011 Dec;11(1):1-1.

13. Feldman HA, Longcope C, Derby CA, Johannes CB, Araujo AB, Coviello AD, Bremner WJ, McKinlay JB. Age trends in the level of serum testosterone and other hormones in middle-aged men: longitudinal results from the Massachusetts male aging study. The Journal of Clinical Endocrinology & Metabolism. 2002 Feb 1;87(2):589-98.
14. Haring R, Völzke H, Felix SB, Schipf S, Dörr M, Rosskopf D, Nauck M, Schöfl C, Wallaschofski H. Prediction of metabolic syndrome by low serum testosterone levels in men: results from the study of health in Pomerania. Diabetes. 2009 Sep 1;58(9):2027-31.
15. Mangat C, Goel NK, Walia DK, Agarwal N, Sharma MK, Kaur J, Singh R, Singh G. Metabolic syndrome: a challenging health issue in highly urbanized Union Territory of north India. Diabetology & metabolic syndrome. 2010 Dec;2(1):1-8.
16. Choudhury BK, Choudhury SD, Saikia UK, Sarma D. Gonadal function in young adult males with metabolic syndrome. Diabetes & Metabolic Syndrome: Clinical Research & Reviews. 2013 Jul 1;7(3):129-32.
17. Mazo EB, Gamidov SI, Iremashvili VV, Gasanov RV. Pathogenetic aspects of erectile dysfunction in patients with the metabolic syndrome. Journal of men's health. 2008 Mar;5(1):42-7.
18. Kasapis C, Thompson PD. The effects of physical activity on serum C-reactive protein and inflammatory markers: a systematic review. Journal of the american College of Cardiology. 2005 May 17;45(10):1563-9.
19. Bansal TC, Guay AT, Jacobson J, Woods BO, Nesto RW. Incidence of metabolic syndrome and insulin resistance in a population with organic erectile dysfunction. J

Sex Med. 2005 Jan;2(1):96-103.