Evaluation of testosterone level in infertile Sudanese male with type 2 diabetic and its relation with lipid profile

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Abstract
Background: Diabetes mellitus (DM) is increasing in men of reproductive age, and the number of diabetic men attending fertility clinics is largely increased. Testosterone is the male sex hormone, and plays a vital role in glucose homeostasis, lipid metabolism, and semen parameters.

Objectives: The aim of this study to assess serum total Testosterone levels among in infertile Sudanese males with type 2 diabetes mellitus and its relation to lipid profile and comparison with apparently healthy control.

Materials and Methods: A case-control study was conducted on 50 non-diabetic control group and 50 infertile diabetic men test group with age range (30-70 years). The analyzed variables age, BMI, serum levels of total Testosterone, HbA1c, lipid profile, and semen parameters were measured.

Results: Serum Total Testosterone levels were significantly lower in infertile male with type diabetes when compared with control group (7.45 ± 1.07 versus 22.21 ± 3.1, p<0.000). Where as serum HbA1c (%), triglyceride, and LDL were significantly increased (10.73±1.72 versus 4.96±0.72, 102.4±12.5 versus 81.11±6.43, 96.68±8.43 versus 84.39±6.65, p < 0.01 respectively). In addition the study observed no significant difference in cholesterol and HDL levels (p > 0.05 ) in the test group when compared with their control. In comparing the results of patients with primary infertility with that with secondary infertility in the test group there is no significant difference in all measured variables (p > 0.05 ). In addition serum testosterone level is significantly inversely correlated with age (r = -0.31, p = 0.02), HbA1c (%) (r = -0.30, p = 0.02), total cholesterol (r = -0.28, p = 0.03), and LDL (r = -0.26, P = 0.04), and insignificantly correlated with BMI, triglyceride, and HDL (p > 0.05). The testosterone level in the diabetic infertile test group illustrated significant inverse correlation with number of sperm with abnormal morphology (r = -0.34, p= 0.01), and insignificantly correlated with sperm count, motility and semen volume (P > 0.05) as in table 3.

Conclusion: It concluded that, type 2 diabetes mellitus significantly altered serum total testosterone level in infertile male, which is inversely correlated with dyslipidemia and semen quality.

Keywords: Glysemic control, type 2 diabetes, serum total testosterone, Lipid profile, HbA1c, semen quality.

Introduction
Diabetes mellitus is an endocrine disorders, characterized by hyperglycemia, polyuria, polydipsia and polyphagia, and affects many organs and is a major causes of renal failure, lower limb amputation for non-traumatic cause, and blindness in adults and with its rate appears to be increasing through the world becoming the...
most frequent causes of death.\textsuperscript{[1,2]} Sex derive and
desire are regulated by sex hormones which are
affected by psychological factors and metabolic
disorders such as diabetes mellitus. Testosterone
is the male sex hormone, is essential for many
vital processes in the body.\textsuperscript{[1,2,3]} Lack of
testosterone affects the male fertility and cause a
decrease in libido, impotency, decrease in muscle
mass, fatigue, decrease in bone density and
osteoporosis. An inverse relationship exists
between testosterone levels and insulin
concentrations in healthy men. Biochemical
evidence indicates that testosterone is involved in
promoting glucose utilization by stimulating
glucose uptake, glycolysis,\textsuperscript{[2,3,4]} and lipolysis.
There has been an alarming increase, of epidemic
proportions, in both obesity and diabetes in the
general population with increased cardiovascular
risk associated with type 2 diabetes mellitus
(T2DM).\textsuperscript{[3,5]}

Many researchers have demonstrated that the level
of testosterone in type 2 diabetes mellitus is lower
than normal subject. This observation is also true
for the free testosterone level.\textsuperscript{[3,4]} Some other
studies did not observe such relation \textsuperscript{[5,6]}. There is
few and inconsistencies studies concerning the
effect of diabetes mellitus on semen parameters,
and rarely focused on this disorder in the infertile
patients. Modern researches illustrated that
diabetes mellitus causes subtle molecular
abnormalities that are essential for sperm function
and quality. DM changes conventional sperm
parameters. In addition, DM results in histologic
damage of the epididymis, with a negative effect
on sperm transit. Many mechanisms explain the
sperm damage in diabetic patients. These include
oxidative stress, endocrine diseases, and
neuropathy.\textsuperscript{[7,8]} Because the diabetes is a
common disease and considering the importance
and the role of testosterone it would be worthwhile
to investigate the factors that can cause a decrease in
testosterone level in infertile Sudanese males with
type 2 diabetes mellitus and its relation to serum
lipid profile

\textbf{Materials and Methods}

Current study was a case-control study, was
conducted in diabetic clinic –Khartoum-Sudan. In
this study100 men aged 30-70 years, including 50
diabetic infertile patients and 50 non diabetic.
Weight was measured to the nearest 100 grams
using a digital personal scale. Height was
measured using a tape meter, to the nearest
centimeter, in standing position without shoes.
Body mass index (BMI) was calculated by taking
a person's weight (Kg) dividing by their height
(meter) squared. BMI=Weight/Height (Kg/m)

All the male participants with a BMI between 20
and 40 having other inclusion criteria were
included in the study.

\textbf{Inclusion Criteria}: any infertile male patients
who have Diabetes type 2.

\textbf{Exclusion Criteria}: fertile Sudanese male with
type2 diabetic A known history of hypogonadism,
panhypopituitarism, or chronic debilitating disease
such as renal failure, cirrhosis and dyslipidemia
were excluded from our study or those who were
on testosterone supplementation were excluded
from the study's.

\textbf{Data Collection and Clinical Examination}

On the same fasting blood samples obtained for
the case control study and follow-up studies,
glycosylated hemoglobin (HbA1c), cholesterol
triglycerides, and total and high -density lipoprotein (HDL) and low density lipoprotein
(LDL), were measured using standard
methodologies in the laboratory of the diabetic
clinic-Khartoum -Sudan.

\textbf{Sample Collection}

Blood was taken from each participant by
standard procedures, serum for total testosterone,
total cholesterol, triglyceride, HDL, and blood
container contain EDTA anticoagulant for HbA1c
estimation.

Semen samples were obtained after a
recommended 2–5 days of sexual abstinence. All
samples were subjected to a conventional light
microscopic semen analysis to determine
liquefaction, semen volume, sperm concentration
and motility according to WHO recommendations
Sperm morphology was assessed according to Tygerberg Strict Criteria (Kruger et al., 1988). Semen analysis was performed within 1h of ejaculation, following a period of incubation at 37°C to allow for liquefaction.

**Ethical Considerations**
Informed consent was taken from all participants and ethical approval was obtained from EL-Neelain University Research Committee.

**Estimation of testosterone level**
Serum total testosterone was measured using ELIZA method.

**Estimation of glycosylated hemoglobin (HbA1c)**
HbA1C was determined by ion exchange chromatography (DS5, Drue Company). Based on American Diabetes Association glycemic control was defined when HbA1C < 7 percent was achieved.

**Estimation of lipid profile**
Total cholesterol, triglycerides, HDL were measured by spectrophotometry.

**Quality control**
Samples representing the normal and pathological level of all measured parameters were used for assessment of the quality control. Results ±2SD of the target values of the control sera were accepted.

**2.10 Statistical analysis**
Data was analyzed by computer software, by using SPSS program manual Master sheet (SPSS version 17). The means and standard deviations of all measured parameters were obtained, and the T-test was used for the comparison between the test and control group, and the mean difference is significant at p ≤ 0.05, Correlation(r) between all measured parameters is considered to be statistically significant at P ≤ 0.05.

**Results**
The samples of this study were 100 men between 30 and 70 years including 50 infertile diabetic test group and 50 non-diabetic healthy men control group sex and age matched shown in table 1, 62% of the test group with primary infertility where as secondary infertility comprised 38 % of the test group in figure 1.mean age and BMI did not show any significant difference between diabetic and non-diabetic groups in table 1.

The serum Total Testosterone levels were significantly lower in infertile male with type2 diabetes as compared to control group ( 7.45 ± 1.07 versus 22.21 ± 3.1, p<0.000).Where as serum HbA1c (%), triglyceride, and LDL were significantly increased (10.73±1.72 versus 4.96±0.72, 102.4±12.5 versus 81.11±6.43, 96.68±8.43 versus 84.39±6.65, p <0.01 respectively), and no significant difference in cholesterol and HDL level (146.74±7.69 versus 144.54±6.71, 38.35±4.71versus37.17±5.69, p > 0.05 respectively) as shown in table 1.In comparing the results of patients with primary infertility with that with secondary infertility in the test group there is no significant difference in all measured variables (p > 0.05) in table 2.

In the test group serum testosterone level is significantly inversely correlated with age (r = -0.31, p = 0.02), HbA1c (%) ( R = -0.30, p = 0.02), total cholesterol(r = - 0.28, p = 0.03), and LDL(r = -0.26, P = 0.04) in figures 1,2,3,4, and insignificantly correlated with BMI, triglyceride, and HDL (p > 0.05) in figures 5,6,7.

The testosterone level in the diabetic infertile test group shows significant inverse correlation with number of sperm with abnormal morphology (r= -0.34, p= 0.01), and insignificantly correlated with abnormal sperm count and motility (P > 0.05) as in table 3.
Table 1 Comparison of serum total testosterone, HbA1c (%), total cholesterol, triglyceride, LDL, and HDL in the test group with their control group

| Parameters                  | Case (Mean±SD) | Control (Mean±SD) | P-value |
|-----------------------------|----------------|-------------------|---------|
| HbA1c (%)                   | 10.73±1.72     | 4.96±0.72         | 0.000   |
| Serum Testosterone          | 7.45±1.07      | 25.22±6.70        | 0.000   |
| Serum cholesterol (mg/dl)   | 146.74±7.69    | 144.54±6.71       | 0.768   |
| Serum triglycerides (mg/dl) | 102.4±12.5     | 81.1±6.43         | 0.004   |
| Serum LDL (mg/dl)           | 96.68±8.438    | 84.39±6.65        | 0.006   |
| Serum HDL (mg/dl)           | 38.35±4.71     | 37.17±5.69        | 0.668   |
| BMI (Kg/m)                  | 24.39±3.49     | 25.27±4.35        | 0.543   |

Table 2 Comparison of serum total testosterone, HbA1c (%), total cholesterol, triglyceride, LDL, and HDL between diabetic patients with primary and secondary infertility in the test group

| Parameters                  | Primary (Mean±SD) | Secondary (Mean±SD) | P-value |
|-----------------------------|-------------------|---------------------|---------|
| HbA1c (%)                   | 10.9±2.9          | 10.5±2.5            | 0.617   |
| Serum Testosterone          | 6.8±4.6           | 8.5±5.6             | 0.419   |
| Serum cholesterol (mg/dl)   | 142.3±38.8        | 154.1±35.6          | 0.287   |
| Serum triglycerides (mg/dl) | 85.6±27.6         | 82.0±22.5           | 0.636   |
| Serum LDL (mg/dl)           | 89.6±24.4         | 92.4±30.2           | 0.723   |
| Serum HDL (mg/dl)           | 37.0±10.1         | 40.6±8.9            | 0.208   |

Figure 2 Ascotter plot shows the relationship between serum total Testosterone (ng/ml) and Age (years)
Figure 3 Scatter plot shows the relationship between serum total Testosterone (ng/ml) and BMI (Kg/m).

Figure 4 Scatter plot shows the relationship between serum total Testosterone (ng/ml) and HbA1c (%).
Figure 5: Scatter plot shows the relationship between serum total Testosterone (ng/ml) and serum cholesterol (mg/dl).

Figure 6: Scatter plot shows the relationship between serum total Testosterone (ng/ml) and serum Triglyceride (mg/dl).
Figure 7 Ascotter plot shows the relationship between serum total Testosterone (ng/ml) and serum total LDL cholesterol (mg/dl)

![Graph showing the relationship between serum total Testosterone and serum total LDL cholesterol.](image)

\[ R = -0.26^* \quad P = 0.04 \]

Figure 8 Ascotter plot shows the relationship between serum total Testosterone (ng/ml) and serum total HDL cholesterol (mg/dl)

![Graph showing the relationship between serum total Testosterone and serum total HDL cholesterol.](image)

\[ R = -0.132 \quad P = 0.362 \]

Table 4 Correlation of serum testosterone level with sperm count, morphology, motility and semen volume in the study group

| Parameter      | Statistic | Motility | count | Abnormal morphology | Semen volume |
|----------------|-----------|----------|-------|----------------------|--------------|
| Testosterone   | Person correlation | 0.14     | 0.17  | 0.29                 | 0.19         |
|                | significance          | 1.3      | 1.05  | 0.01                 | 0.6          |

Correlation is significant at \( p \leq 0.05 \)
Correlation is significant at \( p \leq 0.01 \)
Correlation is significant at \( p \leq 0.001 \)
Discussion
Diabetes mellitus is endocrine disease that causes long term damage to many organs including the reproductive system. Testosterone is male hormone that play a vital role in glucose, lipid metabolism and sperm quality. Many studies have illustrated the relationship between increased BMI and serum testosterone level in patients with T2DM. The results of these studies are inconsistence. Some studies observed a significant correlation between BMI and serum total testosterone level. In contrast, other studies demonstrated insignificant correlation between total testosterone and BMI. However, our study recorded insignificant correlation between BMI and serum total testosterone levels. In the current study serum total testosterone is significantly lower in the infertile patients with type 2 diabetes mellitus compared to healthy subjects. This finding is consistence with studies performed by Jihan MM et al (2016), Tajar et al (2010), and Dhindsa et al (2004). They demonstrated significant number of men with type 2 diabetes have low testosterone level. Many cross sectional studies and systemic analyses illustrated that T2DM is associated with low serum Total Testosterone level. Al hayek et al in (2013) found that 36.5% of patients with diabetes had low serum testosterone level, Kapoor et al in (2007), found that 20% (71 men) had low total Testosterone level. Type 2 diabetes mellitus is high risk factor affecting male reproductive function, and affect the testosterone production and metabolism, causing alteration in testosterone level. Testosterone production could be inhibited by the functional alterations in the Leydig cell either directly through the testis or through changes along the hypothalamus pituitary and testicular axis. The major transporter of sex steroids sex hormone binding globulin could interfere in the bioavailability of this male hormone. To date the exact mechanism by which type 2 diabetes inhibits testosterone biosynthesis is obscure. Leydig cell testosterone biosynthesis is primarily regulated by pulsatile secretion of luteinizing hormone (LH). In addition reduced levels of testosterone in infertile patients with type 2DM may be related to diabetic create - oxidative stress that causing a significant decrease in testicular and adrenal androgen synthesis. Furthermore Yen et al, in their study observed that metabolic alterations caused by type 2DM attenuated the chemical responsiveness of testicular Leydig cells and impair the secretions of testosterone.

In the present study testosterone level in the infertile test group with type 2 DM is significantly inversely correlated with age, which disagree with Dhindsa et al (2004) and Agbecha et al (2017). The significant negative correlation recorded between testoster-one and age in our infertile diabetic patients clarify the physiological role of advanced age on testosterone reduction. Observational studies consistently show that men with type 2 diabetes independent of age and obesity have lowered circulating testosterone levels. More over in a prospective study, the metabolic syndrome is observed to predict low serum testosterone level. Important finding In the present study, testosterone level in the infertile test group with type 2 DM is significantly negatively correlated with HbA1c (%). Whom reflect the significant relationship that exist between the good glycemic control (based on HbA1C≤7) and serum testosterone level Uncontrolled hyperglycemia induced low testosterone level. Systemic review concluded that poor glycemic control in type 2 DM is associated with low serum testosterone level. Many authors reported that locally circulating hormones, growth hormone, and cytokines affect Leydig cell steroidogenesis. Hence, increased insulin resistance or hyperglycemia as in the case of our study group, may interfere in decreased testosterone biogenesis due to decreased central stimulation. Serum testosterone levels reflect the integrity of the HPG axis, and low testosterone levels noted in cases of uncontrolled hyperglycemia may reflect a defect.
at one or more functional levels of the hypothalamus-pituitary-testis axis. However it is obscure how the axis of hypothalamus-pituitary-testis forms the relationship between insulin and testosterone level) [19].

In the present study testosterone level in the infertile test group with type 2 DM is significantly inversely correlated with triglyceride, total cholesterol, and LDL. Which is consistent with Nirmali et al (2015) [27] and Samathia et al (2012) [28]. Whom deduced negative association of total testosterone level with cardiac risk factors in patients with type 2 DM. Some evidence from epidemiological study deduce that total testosterone level is correlated inversely with total cholesterol, LDL-cholesterol, and triglyceride, and positively with HDL-cholesterol. In addition trail of testosterone replacement therapy have illustrated improvement of lipid profile in dyslipidemic subjects. But the mechanism through which testosterone interfere in lipid profile is obscure. Furthermore, recently some researchers suggested that low testosterone can be considered as new cardiovascular risk factor in adult [29].

In the present study Sperm concentration and motility is reduced in infertile test group with type 2 DM, and serum testosterone level is significantly inversely correlated with number of sperm with abnormal morphology. In accordance with Delfino et al (2007) [30], whom observed significant changes in sperm morphology in type 2 diabetic patients. Jing et al (2017) [31] conducted systemic review and meta-analysis to assess the effect of DM on semen parameter and concluded that DM had negative effect on the quality of the semen. Growing evidence explore that DM affect the male fertility through its adverse effect on male reproductive function in multiple level. The disruption of the spermatogenic process, and the apoptosis of the germ cell in DM are considered to be linked to local autoimmune damage [32]. Insulin stimulate many of the lydig cell functions that can affect the spermatogenesis outcome [33].

Conclusion
Type 2 diabetes mellitus causes low total testosterone level, that affects the cardiovascular risk factors and semen parameters.

Consent
As per international standard or university Standard, patient’s written consent has been collected and preserved by the authors.

Ethical Approval
As per international standard or university standard, written approval of Ethics committee has been collected and preserved by the authors.

Competing Interests
Authors have declared that no competing interests exist.

Authors’ contribution
This work was carried out in collaboration between all authors. Author DSEA designed the study, performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Authors (DSEA) and AMA managed the analyses of the study and the literature searches. All authors read and approved the final manuscript.

Acknowledgements
The authors gratefully acknowledge the diabetes patients at Reproductive Health Care Center and Jafar ibn Auf Hospital in Khartoum state, Sudan, for their assistance with obtaining the data used in this study.

References
1. Power A. Diabetes Melitus. In: Fauci AS, Braun wald E, Kasper DS, Hauser SL, Lango DL, Jameson JL, et al., editors. Harrisons Internal Medicine 17nd edition. New York: McGrawHill Inc; 2008. p. 2275-7.
2. Kelly D, Jones T. Testosterone a metabolic hormone in health and disease. Journal of Endocrinology, 2013; 217: 25–45.
3. Ding EL, Song Y, Malik VS, Liu S. Sex differences of endogenous sex hormones and risk of type 2 diabetes: a systematic
review and meta-analysis. *JAMA*, 2006; 295(11): 1288-99.

4. Al Hayek AA, Khader YS, Jafal S, Khawaja N, Robert AA, Ajlouni K. Prevalence of low testosterone levels in men with type 2 diabetes mellitus: a cross-sectional study. *J Fam Community Med.*, 2013; 20: 179-86.

5. Kapoor D, Aldred H, Clark S, Channer KS & Jones TH. A clinical and biochemical assessment of hypogonadism in men with type 2 diabetes: correlations with bioavailable testosterone and visceral adiposity. Diabetes Care. 2007; 30: 911-17.

6. Rhoden. E. L, Ribeiro. E. P, Teloken. C, Souto. C. A.V. Diabetes mellitus is associated with subnormal serum levels of free testosterone in men. BJU International., 2005; 96(6): 867–70.

7. Ari Kawe, A.p., A. Daramola, A. Odofin, L, Obika, Alloxan-induced and insulin-resistant diabetes mellitus affect semen parameters and impair spermatogenesis in male rates. Afr. J.Reprod. Health.,10(3), 2006,106-113.

8. Wang, Y.,Z.Zhang,w, G Guo, W.Sun, X.Miao, H.w u.L.Cai, Sulforaphane reduction of testicular apoptotic cell death in diabetic mice is associated with the upregulation of Nrf2 expression and function. Am. J. physiol, Endocrinol. metab.307(1),2014,E 14-23.

9. Aftab SA, Kumar S, Barber TM. The role of obesity and type 2 diabetes mellitus in the development of male obesity-associated secondary hypogonadism. *Clin Endocrinol (Oxf).*., 2013; 78:330-7.

10. Shota Janjgava, et.al. Influence of testosterone replacement therapy on metabolic disorders in male patients with type 2 diabetes mellitus and androgen deficiency , European Journal of Medical Research., 2014, 19: 56.

11. Kitty Kit Ting Cheung, et.al. Testosterone level in men with type 2 diabetes mellitus and related metabolic effects: A review of current evidence, J Diabetes Invest., 2015; 6: 112–123.

12. Jihan Mohamed Mohiedin,Noon Babiker, Omer Mohamed Abda. Effect of type 2 Diabetes Mellitus on Sudanese male fertility. Sch. J. App.med.sci., 20016,4 (6E):2216-2223.

13. Tajer A,Fori G, O Neill TW, Lee Dm, Silman Aj, Finn JD et al, characteristic of secondary, primary and compensated hypogonadism in ageing men,evidence from the European male ageing study. J clin Endocrinol metab.2010,05(4):1810-8

14. Dhindsa S, Prabahkar S, Sethi M, Bandyopadhayay A, Chaudhuri A, Dandonia P. Frequent occurrence of hypogonadotropic hypogonadism in type 2 diabetes. *J Clin Endocrinol Metab.*, 2004; 89: 5462–8.

15. Al Hayek AA, Khader YS, Jafal S, Khawaja N, Robert AA, Ajlouni K. Prevalence of low testosterone levels in men with type 2 diabetes mellitus: a cross-sectional study. *J Fam Community Med.*, 2013; 20: 179-86.

16. Kapoor D, Aldred H, Clark S, Channer KS & Jones TH. A clinical and biochemical assessment of hypogonadism in men with type 2 diabetes: correlations with bioavailable testosterone and visceral adiposity. Diabetes Care., 2007; 30: 911–17.

17. Rato, L., M. Alves, T. Dias,, G. LOPES, J. Cava co, S. Socrorro, p. Oliveira. High energy diets may induce a pre-diabetic state altering testicular glycolytic metabolic profile and male reproductive parameters-Andrology. (3).2013,495-505

18. Brand JS, Wareham NJ, Dowsett M, Folkerd E, van der Schouw YT, Luben RN, et al. Associations of endogenous testosterone and SHBG with glycated
haemoglobin in middle-aged and older men. Clin Endocrinol (Oxf) 2011;74:572-8.
19. Saez JM. Leydig cells: Endocrine, paracrine, and autocrine regulation. Endocr Rev 1994;15:574-626.
20. Zhao J, Zhai L, Liu Z, Wu S, Xu L. Leptin level and oxidative stress contribute to obesity-induced low testosterone in murine testicular tissue. Oxid Med Cell Longev 2014; 2014: 190945
21. Maggio M, Basaria S. Welcoming low testosterone as a cardiovascular risk factor. International Journal of Impotence Research. 2009; 21: 261–64.
22. Agbecha A, Usoro CA. Serum testosterone and insulin resistance in type 2 male diabetics attending University of Calabar teaching hospital, Nigeria. J Med Soc 2017;31:178-84.
23. Dhindsa S, Miller MG, McWhirter CL, Mager DE, Ghanim H, Chaudhuri A, et al. Testosterone concentrations in diabetic and nondiabetic obese men. Diabetes Care 2010;33:1186-92.
24. Andersson B, Márin P, Lissner L, Vermeulen A, Björntorp P. Testosterone concentrations in women and men with NIDDM. Diabetes Care 1994;17:405-11.
25. Laaksonen DE, Niskanen L, Punnonen K, Nyyssönen K, Tuomainen TP, Valkonen VP, et al. The metabolic syndrome and smoking in relation to hypogonadism in middle-aged men: A prospective cohort study. J Clin Endocrinol Metab 2005;90:712-9.
26. Neagla F, Almilgs,Eman A,Eissa, Emain R.,Amer, Mohamed El-Assal. diabetes mellitus link with hypogonadism in male patients with type 2 diabetes mellitus age 40-50 years.Benha medical journal .2015,32:29-35.
27. Nirmali mattack, Runi Deve, Tridie kutum, Dibyarata Patgiri. The evaluation of serum level of testosterone in type2 diabetic men and its relation with lipid profile .journal of clinical and diagnostic research .2015,vol-9(1);BC04-BC07
28. Samathia P, Venkateswarla M, Slva prabodh V. Lipid profile levels in type 2 diabetes mellitus from the tribal population of Adlabad in Andhra Pradesh , India, journal of clinical and diagnostic research . 2012;6(4):590.
29. Maggio M, Basarla S. Welcoming low testosterone as a cardiovascular risk factor.International journal of impotence Research .2009;21:261-64
30. Delfino M .Imbrogo N. Elia J, Capogreco E,M azzilli F. Prevalence of diabetes mellitus in male partners of infertile couple, Minerva Urol Nefrol.2007;59:131-135
31. Jing-zhen Zhu,Xing-You Dong,Lia-Lia Liang, Zi-Qian Zhang,Xiao Tan Hu.Long Kun Li.Effects of diabetes mellitus on semen quality in adult men .systemic review and meta- analysis .Int J clin Exp Med 2017.10(8):11290-11303
32. La Vignera S, Condovelli R, Vicaan E, D Agata Rand Calogero AE.Diabetes mellitus and sperm parameters. J androl 2012, 33:145-153
33. Perrorr .Sapori MH,Chatliain PC, Rogemand N and Saez JM. Modulation of lydig cell functions by culture with sertoli cells or with sertoli cell-conditioned medium ,effect of insulin ,sematomedin C and FSH. Mol cell Endocrinol 1987,50:193-201.