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Comparison of Serum Free Testosterone, Luteinizing Hormone and Follicle Stimulating Hormone Levels in Diabetics and Non-Diabetics Men- a Case-Control Study

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ABSTRACT

Background: Diabetic men have been claimed to have lower serum free testosterone (FT) concentrations than non-diabetic men. The aim of this study was to investigate if serum FT, luteinizing hormone (LH), and follicle stimulating hormone (FSH) concentrations are different in patients with type 2 diabetes mellitus compared with healthy men and to identify factors associated with low serum testosterone concentrations in men with type 2 diabetes.

Methods: Serum FT, LH and FSH concentrations in 65 men with type 2 diabetes between 50 and 55 years compared with 65 non-diabetic men in Valye-Asr Hospital in Birjand, Iran during October 2009 to August 2010. In addition, the relationships between serum FT concentrations with LH and FSH concentrations as well as other factors including age, fast blood sugar and glycemic control (HbA_{1C}) were investigated.

Results: Serum FT concentrations were significantly lower in type 2 diabetic patients (2.53±1.68 pg/ml) than in healthy men (4.29±2.43 pg/ml) ($P < 0.001$). There were not any significant differences in LH and FSH concentrations between the two groups. There was a significant negative relationship between serum FT and HbA_{1C}, but this relation for LH and FSH was positive and non-significant ($P < 0.001$).

Conclusion: Serum FT concentration is lower in type 2 diabetic patients compared with non-diabetic men suggesting further research to better understand the underlying biologic mechanisms.

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Introduction

Diabetic men have significantly lower concentrations of free and total testosterone than non-diabetic men [1,2]. Hypogonadism has also been reported in men with type 2 diabetes [3, 4]. Furthermore, results from the Massachusetts Male Aging Study and others suggest that low concentrations of endogenous androgens such as testosterone had been related to the development of insulin resistance, which is an important cause for inducing type 2 diabetes and other abnormalities including hyperglycemia, hypertension, dyslipidemia, or carotid atherosclerosis [5-8].

However, a recent study showed that men with higher estradiol levels had an increased risk of later diabetes independent of obesity, while men with lower total testosterone and sex hormone–binding globulin (SHBG) had an increased risk of diabetes that appeared to be dependent on obesity [9].

In the present study, we evaluated whether free testosterone (FT) concentrations are lower in patients with type 2 diabetes in comparison to healthy men and to find the relationships between follicle stimulating hormone (FSH) and luteinizing hormone (LH) in diabetic and non-diabetic men. Moreover, we investigated the correlations among fasting blood sugar (FBS), LH, FSH, glycemic control (HbA_{1C}) and FT in men with type 2 diabetes.

Methods

This case-control study was conducted at the Diabetic Clinic of Valy-Asr hospital in Birjand City (Iran) during October 2009 to August 2010. Sixty-five male type 2 diabetes mellitus patients from 50 to 55 years were selected from patients who had attended to the diabetic special clinic. Inclusion criteria for diabetic patients were the level of 10-12 hours fasting blood sugar (FBS) > 126 mg/dl or random FBS > 200 mg/dl along with diabetic symptoms and/or two hours glucose tolerance test more than 200 mg/dl. Patients with a known history of hy-

pogonadism, panhypopituitarism, coronary heart disease (CHD) or chronic debilitating disease, such as HIV, cirrhosis, renal failure, and/or patients who had used medicines that influence hormones such as corticosteroids, androgens etc were excluded from the study. Control group were sixty-five subjects without history of diabetes mellitus whose FBS were less than 126 mg/dl on two occasions and were matched for age and as well as exclusion criteria of diabetic patients. Informed consent was obtained from all participants, and the study was approved by the BUMS Research Ethics Committee.

Ten ml of venous blood of all subjects was collected and centrifuged. The serums of samples were separated and frozen till performing the experiments. Quantitative determination of serum FT was measured by competitive enzyme immunoassay test using kit (IBL, code no. DB52181, Italy). The intra-assay coefficients of variance (CV) were 17.0, 4.9 and 4.7%, for FT concentrations of 1.17, 15.96 and 62.46 pg/ml, respectively. Serum LH and FSH concentrations were measured by a microplate immunoenzymometric assay using kits from (Costa mesa, code no. 625-300, CA, 92627, USA) and (Costa mesa, code no. 425-300, CA, 92627, USA) respectively. The intra-assay CVs were 5.4, 4.2 and 4.8% for LH of 2.8, 15.2 and 48.5 mlU/ml, respectively. They were 4.3, 4.3 and 2.9% for FSH of 5.9, 16.0 and 41.3 mlU/ml, respectively. HbA_{1C} was measured by Research Center Lab of BUMS using latex immunoturbidimetric assay (LIA). This method utilizes the interaction of antigen and antibody to directly determine the HbA_{1C} in whole blood. The intra-assay CVs were 1.43 and 1.72%, for HbA_{1C} of 5.48 and 10.28 U/L, respectively.

Statistical analysis

The data are presented as means ± standard deviation (SD). Independent T and Chi-Square tests were used to compare relative frequency of abnormal FT concentrations in diabetic and non-diabetic patients. The relationships of age, FBS, FSH, LH, HbA_{1C} and FT in the diabetic patients were evaluated by Pearson correlation test. *P*-value less than 0.05 were considered statistically significant. All statistical analysis was performed using statistical package of SPSS 11.

Results

Mean of age in diabetic patients was 52.95±1.69 years and in non-diabetic 53.22±1.61 years (P=0.701). FBS in diabetic subjects (198.95±63.80 mg/dl) was significantly higher compared to that of non-diabetic subjects (90.94±14.97mg/dl) (P=0.033). Mean FT concentrations in diabetic men were significantly lower in comparison to non-diabetic men (P<0.001) but there was no significant difference between two groups in the means of LH and FSH (Table 1). In diabetic patients mean FT concentrations were significantly higher in patients with HbA_{1c} < 8.0% compared to patients with HbA_{1c} >8.0% (3.29±1.83 *vs.* 2.28±1.57) (P=0.042) and there was also a significant relationship between the level of HbA_{1c} and FBS concentration (r=0.44, P<0.001) (Table 2).

In 53 (81.5%) diabetic men and 32 (49.2%) non-diabetic men, FT concentrations were under the normal rate (<3.84 pg/ml) (P=0.001). The results also showed that there was a significant negative relationship between serum FT and HbA_{1C}, but this relation for LH and FSH was positive and non-significant. The relationship between age, FBS, LH, FSH, HbA_{1C} and FT in diabetic patients has shown in Table 3.

Table 1: Comparison of the mean of luteinizing hormone (LH), follicle stimulating hormone (FSH), and free testosterone (FT) concentrations in the serum of diabetic and non-diabetic subjects

	Diabetic		Non-diabetic		
Variables	**Mean**	**SD**	**Mean**	**SD**	***P* value**
LH (pg/ml)	5.10	4.15	4.97	2.78	0.840
FSH (pg/ml)	5.27	4.74	5.02	1.92	0.690
FT (pg/ml)	2.53	1.68	4.29	2.43	0.001

Table 2: Comparison of the means of fasting blood sugar (FBS) and free testosterone (FT) with respect to HbA_{1C} level ≤8 (N=16) and HbA_{1C} level >8 in diabetic patients

	HbA_{1C} (≤8)		HbA_{1C} (>8)		
Variables	**Mean**	**SD**	**Mean**	**SD**	***P* value**
FBS (mg/dl)	154.38	35.36	213.51	64.49	0.700
FT (pg/ml)	3.29	1.83	2.28	1.57	0.042

Table 3: Correlation between age, fasting blood sugar (FBS), luteinizing hormone (LH), follicle stimulating hormone (FSH), free testosterone (FT) and HbA_{1C} in diabetic patients

	Age	**FBS**	**LH**	**FSH**	**HbA_{1C}**
Age	1.000				
FBS	0.670	1.000			
LH	0.115	0.041	1.000		
FSH	0.068	0.010	0.623	1.000	
HbA_{1C}	0.120	0.444	0.143	0.140	1.000

Discussion

The present patient-based study shows that serum FT concentrations were significantly *(P*<0.001*)* lower in male patients with type 2 diabetes in comparison to healthy men. However, there were no significant differences between LH and FSH in diabetic men compared to healthy men.

A previous study among 38 young (aged 18-35 years) type 1 diabetic and 24 type 2 diabetic patients showed that type 2 diabetic men had significantly lower serum total and FT concentrations and inappropriately low LH and FSH concentrations with high prevalence of hypogonadotrophic hypogonadism in comparison with type 1 diabetic subjects [10]. Their group had previously shown that type 2 diabetic patients with a mean age of 55 years had a frequent occurrence of hypogonadotrophic hypogonadism, as reflected in low plasma concentrations of testosterone and inappropriately low LH and FSH. In the present study, we did not determine the relationship between FT and LH but, Ando et al. 1984 and Fukui et al. 2007 had demonstrated a negative correlation between FT and LH [11, 12]. Other studies carried out in middle-aged populations, had also confirmed the occurrence of hypogonadotrophic hypogonadism due to low plasma concentrations of testosterone, LH and FSH [13, 14]. We tested serum FT concentration instead of total testosterone concentration in healthy men and patients with type 2 diabetic. FT concentration had a stronger correlation with the presence of diabetes mellitus than total testosterone concentration because FT concentration is a more sensitive and active marker for insulin resistance or atherosclerosis [15]. In a large population-based cohort, Schipf et al. showed that there was an association between total testosterone and incidence of type 2 diabetes mellitus. Their prospective findings suggest the importance of measuring total testosterone in men as the predominant male sex hormone [16].

Low levels of sex hormone–binding globulin (SHBG) and total testosterone in men may be involved in the pathogenesis of type 2 diabetes in humans. In a 9-month lifestyle intervention research on 225 subjects the levels of SHBG were posi-

tively correlated with insulin sensitivity, independently of sex, age and total body fat [17]. They concluded that possible mechanisms by which high circulating SHBG prevents the development of type 2 diabetes involve regulation of fasting glycemia but not alteration of insulin secretory function [17].

In another study on 1709 men that evaluated for 7-17 years, SHBG was an independent predictor of incident type 2 diabetes mellitus even after adjusting for FT or total T [18]. Bedsides, FT was not significantly associated with type 2 diabetes mellitus. SHBG may contribute to the risk of type 2 diabetes through nonandrogenic mechanisms, which should be investigated as they may provide novel targets for diabetes prevention [19]. Although we did not evaluate the level of SHBG in subjects, but the serum FT concentration was significantly lower in type 2 diabetic patients compared to that of healthy men and it can be used as a marker of men patients with type 2 diabetes.

There was a significant negative correlation between testis weight and fasting blood sugar, moreover in male diabetic rats, with a variety of hypogonadisms such as atrophy of testis and low sperm count, FT, LH and SHBG were normally preserved [20].

Conclusion

Serum free testosterone is lower in type 2 diabetic patients compared with non-diabetic men and there is also a negative correlation between FT and HbA_{1C} in men with type 2 diabetes. This finding suggests further research to better understand the underlying biologic mechanisms.

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Conflict of interest statement

There is no conflict of interest in this article.

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References

1. Andersson B, Marin P, Lissner L, Vermeulen A, Bjorntorp P. Testosterone concentrations in women and men with NIDDM. *Diabetes Care*. 1994;17:405-411.

2. Barrett-Connor E. Lower endogenous androgen levels and dyslipidaemia in men with NIDDM. *Ann Intern Med*. 1992;117:807-811.

3. Fushimi H, Horie H, Inoue T, Kameyama K, Kanano K, Ishihara S. Low testosterone levels in diabetic men and animals: a possible role in testicular importance. *Diabet Res Clin Pract*. 1989;6:297-301.

4. Goldstein I, Inigo Sanez de Tejada. Erectile dysfunction and diabetes. In: Kahn CR, Weir GC, editors. *Joslin's Diabetes Mellitus*, 13th ed. Philadelphia: Lea & Febiger; 1994. pp. 852-866.

5. Haffner SM, Karhapaa P, Mykkanen L, Laakso M. Insulin resistance, body fat distribution and sex hormones in men. *Diabetes*. 1994;43:12-19.

6. Simon D, Charles MA, Nahoul K, Orssaud G, Kremski J, Hully V, et al. Association between plasma total testosterone and cardiovascular risk factors in healthy adult men: The Telecom Study. *J Clin Endocrinol Metab*. 1997;82:682-685.

7. Stellato RK, Feldman HA, Hamdy O, Horton ES, McKinlay JB. Testosterone, sex hormone-binding globulin, and the development of type 2 diabetes in middle aged men. *Diabetes Care*. 2000;23:490-494.

8. Fukui M, Kitagawa Y, Nakamura N, Kadono M, Mogami S, Hirata C, et al. Association between serum testosterone concentration and carotid atherosclerosis in men with type 2 diabetes. *Diabetes Care*. 2003;26:1869-1873.

9. Vikan T, Schirmer H, Njølstad I, Svartberg J. Low testosterone and sex hormone-binding globulin levels and high estradiol levels are independent predictors of type 2 diabetes in men. *Eur J Endocrinol*. 2010;162:747-754.

10. Chandel A, Dhindsa S, Topiwala S, Chaudhuri A, Dandona P. Testosterone concentrations in young patients with diabetes. *Diabetes Care*. 2008;31:2013-2017.

11. Ando S, Rubens R, Rottiers R. Androgen plasma levels in male diabetics. *J Endocrinol Invest*. 1984;7:21-24.

12. Fukui M, Soh J, Tanaka M, Kitagawa Y, Hasegawa G, Yoshikawa T, et al. Low serum testosterone concentration in middle-aged men with type 2 diabetes. *Endocr J*. 2007;54(6):871-877.

13. Kapoor D, Aldred H, Clark S, Channer KS, Jones TH. Clinical and biochemical assessment of hypogonadism in men with type 2 diabetes: correlations with bio-available testosterone and visceral adiposity. *Diabetes Care*. 2007;30:911-917.

14. Mulligan T, Frick MF, Zuraw QC, Stemhagen A, McWhirter C. Prevalence of hypogonadism in males aged at least 45 years: the HIM study. *Int J Clin Pract*. 2006;60:762-769.

15. Rhoden EL, Ribeiro EP, Teloken C, Souto CAV. Diabetes mellitus is associated with subnormal serum levels of free testosterone in men. *B J U Int*. 2005;96:867-870.

16. Schipf S, Haring R, Friedrich N, Nauck M, Lau K, Alte D, et al. Low total testosterone is associated with increased risk of type 2 diabetes mellitus in men: results from Study of Health in Pomerania (SHIP). *Aging Male*. 2011;14(3):168-175.

17. Peter A, Kantartzis K, Machann J. Relationships of circulating sex hormone–binding globulin with metabolic traits in humans. *Diabetes*. 2010;59:3167-3173.

18. Lakshman K, Bhasin S, Araujo A. Sex hormone–binding globulin as an independent predictor of incident type 2 diabetes mellitus in men. *J Gerontol A Biol Sci Med Sci*. 2010;65A:503-509.

19. Dhindsa S, Prabhakar S, Sethi M, Bandyopadhyay A, Chaudhuri A, Dandona P. Frequent occurrence of hypogonadotrophic hypogonadism in type 2 diabetes. *J Clin Endocrinol Metab*. 2004;89:5462-5468.

20. Komaki K, Ohno Y, Aoki N. Gonadal hormones and gonadal function in type 2 diabetes model OLETF (Otsuka Long Evans Tokushima Fatty) rats. *Endocr J*. 2005;52:345-351.