Effects of short term metformin administration on androgens in normal men

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ABSTRACT

Objectives: To study the effect of metformin on androgens in normal men.

Methods: A total of 12 healthy males volunteered to participate in the study. A blood sample was obtained from each of them and analyzed for the following: Testosterone (total and free), sex hormone binding globulin dehydroepiandrosterone sulphate, 17-hydroxyprogesterone, luteinizing hormone, and follicle stimulating hormone. In addition, each participant was subjected to a glucose tolerance test and his insulin level was measured. Metformin 850 mg twice daily for 2-weeks was given to each subject after which the above tests were repeated. A paired t-test was used to assess the statistical significance of any observed differences before and after metformin.

Results: After metformin administration, there was a significant reduction in serum level of total testosterone (p=0.0001), free testosterone (P=0.002), and 17 hydroxyprogesterone (p=0.0001). There was also a significant increase in serum level of sex hormone binding globulin (p=0.009) and dehydroepiandrosterone sulphate (P=0.0008). Serum levels of luteinizing hormone and follicle stimulating hormone showed no significant changes. Similarly, there were no changes in fasting plasma glucose, fasting serum insulin, weight, or blood pressure.

Conclusion: Metformin administration was associated with a reduction in total testosterone, free testosterone, and 17-hydroxyprogesterone and an increase in sex hormone binding globulin and dehydroepiandrosterone sulphate in normal males. The clinical significance of these findings needs further investigation.

Keywords: Metformin, androgen, changes, normal, men.

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Metformin is an insulin sensitizer,[1,2] which, has gained widespread usage in the treatment of type 2 diabetes mellitus.[3] The primary mechanism of action is the improvement of insulin sensitivity in the liver and muscles and suppression of hepatic glucose production through the inhibition of gluconeogenesis and glycogenolysis.[4,5] Metformin gained more popularity in the treatment of diabetes mellitus particularly in reducing mortality in patients with coronary artery disease after the publication of the United Kingdom Prospective Diabetic Study (UKPDS).[3] In addition, metformin is used to treat the state of hyperinsulinemia due to insulin resistance associated with polycystic ovary syndrome (PCOS).[6] In women with PCOS several studies have shown that metformin reduces serum testosterone and increases sex hormone binding globulin (SHBG).[6,7] In spite of the abundance of data on the effects of metformin on androgens and its usefulness in reducing testosterone and increasing SHBG in

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women with and without PCOS.[8] Information on the effects of metformin on normal males is either scarce or lacking. The purpose of this study is to report the effect of metformin administration for 2 weeks on normal healthy male volunteers.

Methods. Twelve healthy volunteers aged between 19 years and 30 years, who have no clinical evidence of any endocrinopathy and on no medication were studied. After informed consent was obtained from each subject and the project was approved by the ethical committee, a complete physical examination was performed. Their mean body mass index (BMI; Kg/m^2) was 23.6, blood pressure was measured by a mercury sphygmomanometer in the sitting position after rest for 5 minutes and the mean blood pressure was 110/73 mm Hg. After an over night fast for a minimum of 8 hours, a baseline venous blood sample was obtained at 8am for fasting plasma glucose, insulin, total and free testosterone, SHBG, dehydroepiandrosterone sulphate (DHEA-S), 17-hydroxyprogesterone, luteinzing hormone (LH) and follicle stimulating hormone (FSH). A standard oral glucose tolerance test (OGTT) was performed using 75g of anhydrous glucose. Blood specimens were obtained at 0, 30, 60, 90 and 120 minute intervals for the same above mentioned parameters. Each subject was given metformin, 850mg twice daily for 2 weeks. Metformin tablets were provided for the study subjects on a daily basis each morning to make sure that subjects were compliant and to monitor any side effects of metformin by history especially gastrointestinal disturbance.

At the end of the 2 weeks, the glucose tolerance test was repeated as mentioned above, blood specimens were obtained for the same parameters.

Laboratory procedures. Serum from blood samples was separated immediately after they were collected and kept frozen at -20°C until analyzed.

Total testosterone was analyzed by radioimmunoassay (RIA) using a commercial kit (Dia Sorin) with a sensitivity of 0.059 ng/ml (conversion factor to nmol/L=3.48). (Dia Sorin Incorporation 1990 Industrial). Free testosterone and 17-hydroxyprogesterone were analyzed by (RIA) using commercial kits from Diagnostic Products Corporation (DPC) with sensitivities of 0.15 pg/ml (conversion factor to pmol/L =3.48 and 0.2 nmol/l) (Diagnostic Products Corporation, 1999). Sex hormone binding globulin was measured by Immunoradiometric Assay (IRMA) using DPC kits from Diagnostic with a sensitivity of 0.04 nmol/l. (Diagnostic Products Corporation, September 2000) Dehydroepiandrosterone sulphate was analyzed by RIA using commercial kits from aboratories with a sensitivity of 0.046 mmol/l. Samples were analyzed for gonadotropins (LH and FSH) and insulin by microparticle enzyme immunoassay (MEIA) using IMX, ABBOTT Diagnostics Instruments from ABBOTT Laboratory Diagnostic Division with a sensitivity of 0.5 IU/L, 0.2 IU/L and1.0μU/ml.

Statistical analysis. Data was analyzed using Epi info, version 6, software. For total testosterone, DHEA-S, 17-hydroxyprogesterone, LH, and FSH, the mean value, of the 5 readings for each one of them taken from the OGTT had been calculated before and after metfomin. Then, the paired t-test was used to assess the statistical significance of the differences in serum levels of these hormones before and after metformin.

Results. There was no statistically significant change in body weight in our subjects before and after metformin, mean weight was 76.6 kg and 76.5 kg. The blood pressure readings before and after metformin demonstrated no statistically significant difference; the mean blood pressure was 110/73.3 mm Hg and 108/70.4 mm Hg.

The effect of metformin on glucose and insulin levels. All subjects had normal OGTT. The mean glucose concentration at 0 and 120 minutes post-glucose challenge was within the normal range before and after metformin. The mean fasting glucose was 98.7 mg/dl and 96.5 mg/dl before and after metformin. At 120 minutes post-glucose challenge the mean glucose was 87.6 mg/dl and 86.1 mg/dl before and after applying metformin. At base line, insulin concentration was 6.5 mu/ml before and after metformin, with no statistically significant difference. At 120 minutes the insulin level was 11.5 μu/ml before and 11.6μu/ml after metformin, **(Table 1)**.

The effect of metformin on androgens gonadotropins and sex hormone binding globulin. Serum total testosterone levels decreased significantly (p=0.0001) after metformin. The level of free testosterone also decreased significantly

Table 1 - The effect of metformin on glucose and insulin levels.

Variable	Before Metformin		After Metformin*	
	Blood glucose mg/dl ± SD	Serum insulin Level μU/ml ± SD	Blood glucose mg/dl ± SD	Serum insulin level μU/ml ± SD
0 min	98.7 ± 9.1	6.5 ± 1.8	96.5 ± 6.97	6.5 ± 2.3
30 min	135 ± 19.5	52.5 ± 6.7	138.9 ± 26.7	45.2 ± 15.8
60 min	112.3 ± 20.6	35.4 ± 16.5	117.8 ± 31.1	43.7 ± 23.8
90 min	97.3 ± 16.5	18.5 ± 11.0	105.2 ± 13.2	22.8 ± 22.8
120 min	87.6 ± 8.9	11.6 ± 10.7	86.1 ± 13.8	11.7 ± 10.7
*P value non significant, ± SD - standard deviation				

Table 2 - The effect of metformin on androgens, gonadotropins and sex hormone binding globulin.

Variable	Before Metformin Mean ± SD	After Metformin Mean ± SD	P Value
Total testosterone (ng/ml)	4.9 ± 1.0	3.8 ± 0.8	0.0001
Free testosterone (pg/ml)	23.3 ± 4.8	20.3 ± 4.3	0.002
SHBG (nmol/l)	47.5 ± 18.2	52.6 ± 18.8	0.009
DHEA-S (µmol/l)	5.7 ± 1.5	6.9 ± 1.8	0.0008
17 Hydroxy progesterone (nmol/l)	9.5 ± 2.9	6.7 ± 2.8	0.0001
LH (IU/L)	5.7 ± 2.5	5.6 ± 2.9	0.6
FSH (IU/L)	3.6 ± 1.6	3.9 ± 2.2	0.5

P-values are obtained by the pair t-test, ± standard deviation,
SHBG - sex hormone binding globulin,
DHEA-S - dehydroepiandrosterone sulphate,
LH - luteinzing hormone, FSH - follicle stimulating hormone

(p=0.002) after metformin. On the other hand, there was a significant (p=0.009) elevation in serum levels of SHBG and DHEA-S. 17-hydroxprogesterone levels decreased after metformin (0.0001). Luteinizing hormone and FSH levels showed no significant changes in response to metformin administration **(Table 2)**.

Discussion. Our data presents the effects of metformin on androgens in normal males. Studies on PCOS carried out by De Leo et al,[7] Nestler and Jakubawicz[9-10] and Velazquez et al[6,11] where normal women were used as controls, showed that metformin intake was associated with a reduction in serum total and free testosterone, this is similar to our findings in a normal male group.

The observed decrease in free testosterone in our study can be explained by the elevation in the serum level of SHBG.[6] However, the observed reduction in total testosterone is more difficult to explain, due to total testosterone is bound with high affinity to SHBG and levels the change with alteration in SHBG level.[13] Similar to other studies[6,7,9,10,12,14] our findings showed elevation in SHBG and DHEA-S.

Several studies have provided evidence that regulation of serum SHBG by insulin is a physiological phenomenon,[13] and that insulin is the most important factor that affects the plasma concentration of SHBG, in other words a state of hyperinsulinemia causes a reduction of serum level of SHBG.[15-17] Since metformin is an insulin sensitizer, the observed elevation in SHBG in our study is an expected finding. Our results did not show any reduction in blood glucose nor in insulin level after metformin usage, a finding consistent with some previous studies, which showed no change or slight insignificant change in insulin level.[4,5,18-21] We could not demonstrate any change in insulin sensitivity using glucose to insulin gastrointestinal ratio,[22] homeostasis model assessment (HOMA methods),[23] quantitative insulin sensitivity check index QUICKI,[24] corrected insulin response,[25] and acute insulin secretary response to glucose[25] to calculate insulin sensitivity. Taking into account that fasting gastrointestinal ratio[22] and HOMA[23] model are a good measure of insulin sensitivity in obese PCOS women[22,23] and are not recommended to be good measures of insulin sensitivity in non-obese PCOS women.[22] The remote possibility that there was a change in insulin sensitivity in our group could not be excluded, since we did not use clamp technique to measure insulin sensitivity, therefore, we can not exclude that improvement in insulin sensitivity was achieved completely.

The plasma level of DHEA-S in our study increased significantly while 17-hydroxyprogesterone concentration decreased after metformin. The increase in the concentration of DHEA-S cannot be explained by an increase in the plasma concentration of SHBG,[6] as DHEA-S does not bind to plasma SHBG. An increase in plasma level of DHEA-S reflects adrenal production, which is under the influence of insulin.[26] Our results are in agreement with what Nestler et al[27] demonstrated when metformin was administrated to a control group of non-obese normotensive men, where there was increase in DHEA-S levels by 80% in none obese normotensive men. His explanation was that the rise in serum DHEA-S might be due to decrease in insulin level or improvement in insulin sensitivity or direct action of metformin on adrenal androgens. Similar to the findings from the studies in women with PCOS[9,10] we found a reduction in 17-hydroxyprogesterone in normal men after metformin therapy. This reduction in 17-hydroxyprogesterone can be explained by the fact that insulin acts to modulate 17-hydroxylase and[17,20] lyase activity of adrenal steroids.[28,29]

Our findings showed that metformin given for a 2 week period has no direct effect on gonadotropin levels in normal men. However, the effect of prolonged intake of metformin on gonadotropins can not be excluded.

In conclusion, our findings demonstrate a reduction in serum total, free testosterone and 17-hydroxyprogesterone in normal men and an increase in SHBG and DHEA-S in response to metformin taken for a period of 2 weeks. Detailed interpretation and analysis of our findings, can not be applied here, due to the methodology of our study, which was designed simply to assess the hormones before and after metformin administration for 2 weeks, so the long-term consequences and the clinical significance of these findings should be evaluated in future

studies especially among diabetics who are likely to be maintained on metformin therapy for a long duration.

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