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Original articles

PERMANENT LOW NORMAL TESTOSTERONE LEVELS AND SEMINOLOGICAL DAMAGES IN MEN FROM 37 TO 45 YEARS OLD

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ABSTRACT

In our practice, we are increasingly seeing men aged from 37 to 45 years' old who have permanent low normal testosterone levels.

Aim: We set out to investigate whether there is an association between permanent low normal testosterone levels and negative change in seminal fluid parameters in young men.

Patients and Methods: For the period from January 2013 to December 2015 at the Andrology office at Hospital "St. Sofia" we examined 73 men aged 37 to 45 years with normal or elevated body mass index, permanent low normal testosterone level, and negative change in seminal fluid parameters. In order to compare the results we obtained, at the very beginning of the study we selected a control group of 20 healthy men of the same age.

Results: We obtained, although within reference ranges, significantly lower values for total testosterone in the 73 men with negative change in seminal fluid parameters, compared with those in the control group without seminal damages ($p<0.001$) We found a high correlation relationship between the level of testosterone and the results of the first ($r = 0.614$, $p<0.001$) and second spermograms ($r = 0.662$, $p<0.001$).

Conclusions:

1. Our study shows that in a number of men at a young age, some decrease in normal testosteronå secretion occurs, with a concomitant negative change in seminal fluid parameters, which is remarkably different from the same parameters in their peers with a high normal testosterone level.

2. We identify permanent low normal testosterone, overweight and obesity as predictors, signaling a possible negative change in seminal fluid parameters.

3. We can say that if obesity plays some role in seminal damages, the mechanism in most cases is most likely related to the sustained permanent low normal testosterone level, as a result of increased adipose tissue.

Keywords: low normal testosterone, BMI, seminal damages,

INTRODUCION

In our outpatient practice, we are increasingly seeing men aged 37 to 45 years with permanent low normal total testosterone (T). De Kretser D. 1979 reviews the endocrinological aspects of male infertility, beginning with the physiological interrelationship of the hypothalamus-pituitary-gonad. According to him, the reduced secretion of luteinizing hormone (LH) and follicle stimulating hormone (FSH) from the pituitary gland leads to impaired testicular function and infertility. However, in such men, gonadotropin deficiency accounts for less than 5% of the causes. An increase in the values of LH and a decrease in those of T are found in about 30% of men with severe testicular damage and are indicative of interstitial cell failure. [1] Pierrepoint C. et al. 1982 measured plasma concentrations of LH, FSH, prolactin, T, and estradiol in men with normal fertility and in infertile men, finding that plasma LH, FSH, prolactin, and T levels were significantly different in fertile than in infertile men with or no sperm in the ejaculate. [2] Low T values are found in approximately 15% of subfertile men. [3] Although androgens are essential for spermatogenesis, it is unclear whether low T levels may have a negative impact on sperm parameters in men belonging to infertile couples. Furthermore, the question of whether an endocrine evaluation should be included in the initial evaluation of the subfertile man remains controversial. In their study, DiGuardo F. et al. 2020 found that most patients in the low T group also had subnormal seminal fluid parameters. [3] Meeker J. et al. 2007 demonstrated a study in which they investigated the influence and predictive ability of reproductive and thyroid hormones on sperm quality among men who were partners in infertile couples, finding that levels of FSH, LH, inhibin B, T and free testosterone were associated with seminal parameters. [4] Jorgensen N. et al. 2016 presented another large retrospective analysis in men with a mean age of 19 years, finding that serum T levels were not associated with total sperm count and concentration, percentage of motile and morphologically normal sperm. [5] Olesen I. et al. 2018 are holding one study comparing

seminal fluid quality with free T and total T level, finding that in men with deteriorating sperm quality between baseline sampling and follow-up, average concentration and total sperm count decreased, with 10% of men reaching azoospermia. [6] Along with lowering serum T levels, a large meta-regression analysis performed by Wu F. et al. 2010 demonstrated a significant decline in sperm count from 1973 to 2010. The reasons are not yet fully understood, but pituitary inhibition, T deficiency, and decreased sperm count, according to them, warrant further investigation. [7] McDonald A. et al. 2010 and McDonald A. et al.2013 in two of their studies on the influence of body mass index (BMI) on the quality and quantity of seminal fluid and the level of sex hormones did not find evidence of an association between increased BMI and sperm parameters, but found that it was negatively associated with male reproductive hormones [8, 9] Salas-Huetos A. et al. are of the opposite opinion, with the results of their study showing that overweight and/or obesity are associated with parameters of low sperm quality and some specific reproductive hormones (eg, inhibin B, T and sex hormone binding globulin). [10] Sermondade N. et al. 2013 confirm the opinion that overweight and obesity are associated with an increased prevalence of oligozoospermia or azoospermia, without being able to answer whether weight normalization improves seminal fluid parameters [11] and Zhong O. et al. 2021 add diabetes to obesity in the negative influence of both sperm parameters and low levels of T. [12] A group of scientists from China found a statistically inverse correlation between some markers associated with obesity (waist/hip ratio and waist/height) and sperm parameters (sperm concentration, ratio of progressive to non progressive motility). The T level was much lower in the obese group than in the normal group. [13]

According to the guidelines defined for clinical practice by the Endocrine Society, the subnormal T level for men should be between 10.4 - 34.7 nmol/L. [14, 15] The recommendations of the International Society of Andrology (ISA), the International Society for the Study of Aging Men (ISSAM), the European Association of Urology (EAU) and the American Society of Andrology (ASA), define a minimum T level of 7.98 nmol/L, and for total testosterone values between 7.98 -10.4 nmol/lL, recommend additional measurement of free testosterone. [16]

AIM

We set out to investigate whether there is an association between permanent low normal testosterone levels and negative change in seminal fluid parameters in young men.

MATERIALS AND METHOD

The study is clinical non-randomized observational study. Between January 2013 and December 2015 in the Andrological office, First Hospital of Obstetrics and Gynecology "St. Sofia" we examined 73 men aged 37 to 45 years with permanent low normal T level, normal or elevated BMI and negative change in seminal fluid parameters. We selected also a control group of 20 clinically healthy men of the same age, who have not used drugs or testosterone preparations. All patients were informed in detail and signed a written consent to participate in the present study.

We tested each man's serum T lewel three times every 45 days for a period of 3 monts [17] We performed the blood collection after a mandatory 30-minute rest period between 8.00 and 9.00 hours in the morning after an overnight fast. Hormonal analysis was performed with a mini Vidas apparatus of Bio-Mérieux company and standard reagents to it according to the radioimmunological analysis method. Normal values for serum T (10.4-29.0 nmol/L), in men were determined by the manufacturer. The mean values of the three T samples in the male control group were 19,04-24,64 nmol/L. We considered these values to be high degree of normality the serum T level, in accordance with the guidelines set for clinical practice by the Endocrine Society. [14, 15] Men with a serum T level from the first sample below 19.0 nmol/L were included in our study after second and third confirmation of the initial result. According to the guidelines set for clinical practice by the Endocrine Society recommendations of ISA, ISSAM, EAU, and ASA and depending on the mean values obtained from the three T samples, we defined two sublevels within the reference range. They were as follows: high degree of normality the serum T level 19.04-24.64 nmol/L and low normal serum T level 8,60-14,28 nmol/L. [14, 15, 16]

According to the World Health Organization criteria for normal and overweight [18] and depending on the T level, all 93 men were divided into 5 groups as follows:

1. Group 1 - 20 men with normal BMI (18,50-24,99) and high normal T level.
(reference group)
2. Group 2 - 18 men with normal BMI (18.50-24.99) and permanent low normal T level.
3. Group 3 - 27 overweight men (BMI 25.00-29.99) and permanent low normal T level.
4. Group 4 - 16 men with grade I obesity (BMI 30.00-34,99) and permanent low normal T level.
5. Group 5 - 12 men with grade II obesity (BMI 35.00-39,99)and permanent low normal T level.

In our study, there were men with normal BMI values, but with long term persistence of medium or low normal T levels, and this necessitated their separation into a separate (second) group, different from the control and other groups.

Spermograms were performed on all men twice, during a period of 25-30 days after 4-5 days of sexual abstinence. The men excreted material in a specially adapted room in the hospital, and the examination was carried out up to two hours after masturbation by a reproductive biologist, according to the World Health Organization (WHO) criteria described in the human reproduction program 2021. [19]

Microbiological examination of seminal fluid and sterile urine was performed twice over a period of 5-7 days. A specialist microbiologist performed the tests for Tri-

chomonas vaginalis, Neisseria gonorrhoeae, Chlamydia trachomatis, Ureaplasma parvum, Ureaplasma urealyticum, Mycoplasma hominis and Mycoplasma genitalium. The latter were performed with the one step rapid test - an immune chromatographic test for in vitro diagnostics by Ameritech for Chlamydia and Mycoplasma system plus by Liofilchem diagnostic for Mycoplasma and Ureaplasma.

We compared the results of all 73 men with seminal damages included in our study with those of 20 healthy men in the control group. For processing the survey data, we used the statistical software IBM SPSS STATISTICS Version 25. We used: Independent Samples T-Test, chi-square test, parametric coefficient of linear correlation – Pearson, non-parametric linear correlation coefficient – Spearman.

RESULTS

On table 1 we present general data on the 93 men we examined.

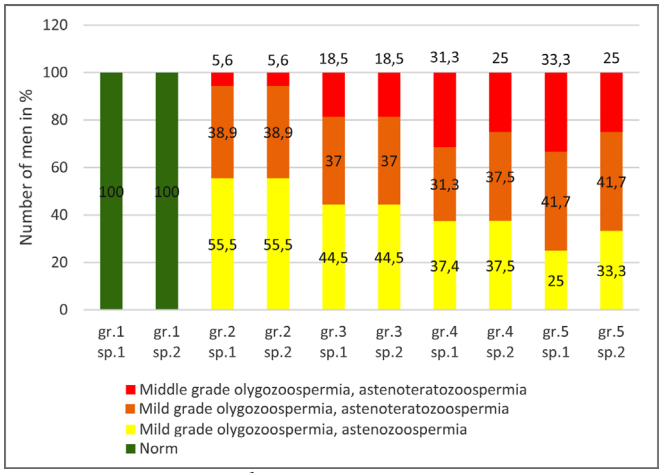
Table 1. General data on the 93 men we examined

Parameter	Average ± SD	Range
Age(years)		
Group 1	40.950 ± 2.743	37 - 45
Group 2	40.611 ± 2.789	37 - 45
Group 3	40.741 ± 2.640	37 - 45
Group 4	41.500 ± 2.338	37 - 45
Group 5	42.750 ± 1.913	39 - 45
BMI (kg/m^2)		
Group 1	21.947 ± 1,434	19.71 - 24.68
Group 2	21.694 ± 1.314	19.44 - 23.80
Group 3	27.250 ± 1.066	25.34 - 28.84
Group 4	32.654 ± 1.213	31.26 - 34.81
Group 5	37.359 ± 1.049	35.91 - 38.94
T (nmol/l)		
Group 1	21.576 ± 0.993	19.04 - 24.64
Group 2	12.199 ± 1.436	9.69 - 14.28
Group 3	11.962 ± 1.590	9.72 – 13.82
Group 4	10.680 ± 1.089	9.01 - 13.05
Group 5	10.236 ± 1.339	8.60 - 13.20

BMI – body mass index, T – total testosterone

Normospermia was found in all men of the first group in the first and second seminal examinations. In the patients of the second to fifth groups, from mild oligozoospermia with asthenozoospermia to moderate oligozoospermia with asthenoteratozoospermia was found in the results of the first and second seminal examinations. (Fig. 1)

Fig. 1. The number of men in the individual groups expressed in percentages with different degrees of the spermograms damages



gr. - group number; sp - spermogram

We did not find significant differences in the sperm parameters (with concentration, total number, percentage of motility and normal sperm morphology) between second and fifth groups, but we found a clear tendency to increase the number of patients with moderate oligozoospermia with asthenoteratozoospermia with the decrease in serum T level, especially in the fourth and fifth groups. groups. We found a high correlation relationship between the level of T and the results of the first ($r = 0.614$, $p<0.001$) and second spermograms ($r = 0.662$, $p<0.001$).

In men of the control group, microbiological examinations of urine and ejaculate showed no growth. Mycoplasma genitalium, Ureaplasma urealyticum and Chlamydia trachomatis was iso-listed in the ejaculate of 9 (12.3%) of the 73 patients studied. Antibiotic treatment with a tetracycline preparation for 10 days was performed in all these men, and control microbiological examinations were negative, but seminal damages remained persistent after therapy.

DISCUSSION

According to some authors, infertility affects 15-20% of couples, and in 20-50% the cause of infertility is the male factor. [20] De Kretser D. 1979 examines the endocrine aspects of male infertility, establishing increased levels of LH and low T levels in about 30% of men with severe testicular damage and defined them as indicators of interstitial cell failure. Routine semen analysis remains the main standard for the evaluation of infertility. [1] In our study, we performed two seminal examinations on each patient 25-30 days apart, all of which were evaluated by a reproductive biologist according to the laboratory manual for the examination and processing of the WHO human sperm 2021. [19] Jorgensen N. et al. 2016 presented a large retrospective analysis finding that serum levels of T were not associated with concentration, total number, percentage of motility and normal sperm

morphology. [5] Other authors demonstrated a varying degree of decrease in the number of sperm, along with a decrease in serum levels of T. [2, 3, 4, 6, 7] According to DiGuardo F., et al. 2020 low T levels are found in approximately 15% of subfertile men. In their study, they found that most patients in the low T group also had subnormal seminal fluid parameters. [3] In our study in the control group men who had a high normal T level, normozoospermia was established. In all the others, we found negative change in seminal fluid parameters of varying severity from a mild degree oligozoospermia with asthenozoospermia to moderate degree oligozoospermia with asthenozoospermia or asthenoteratozoospermia. The analysis of the results showed that the largest number of men (31 men or 42.5%) were with mild degree oligozoospermia with asthenozoospermia and the least number (15 men or 20.6%) with moderate degree oligozoospermia with asthenoteratozoospermia. Using the chi-square test, we found significant differences between the results of seminal examinations of men from the first and the other groups $p<0.001$. Our results demonstrate that in parallel with the decrease in the average of T level from the second to the fifth group, the percentage of men with mild degree oligozoospermia with asthenozoospermia decreases and that of men with moderate degree oligozoospermia with asthenoteratozoospermia increases.We found a high correlation relationship between the level of T and the results of the first ($r = 0.614$, $p<0.001$) and second spermograms ($r = 0.662$, $p<0.001$). DiGuardo F., et al. 2020 recently presented a study in men with low T and severe or very severe oligoasthenozoospermia. [3] Our results show that permanent low normal T level is also a prerequisite for seminal disorders, but they are milder than those presented in men with low T. McDonald A. et al. 2010 and McDonald A. et al. 2013 found no evidence of a relationship between increased BMI and sperm parameters. [8, 9] Other authors believe that overweight and obesity are associated with an increased prevalence of oligozoospermia or azoospermia. [10, 11, 13] In our study we confirm the opinions of recent authors, as in the groups with increased BMI, we observed from mild degree oligozoospermia with asthenozoospermia to moderate degree oligozoospermia with asthenoteratozoospermia. Furthermore, with increasing BMI, the number of men with mild oligozoospermia and asthenospermia decreased, and that of men with moderate oligozoospermia and asthenoteratozoospermia increased.

It is more difficult to explain the permanent low normal T level in men from the second group with normal BMI and negative change in seminal fluid parameters. One possibility is that weight gain is not the only cause, but that there is another one related to lifestyle and unhealthy habits in men, which are also a prerequisite for permanent low normal T level and negative change in seminal fluid parameters. Another possibility is that these men had permanent low normal T and negative change in seminal fluid parameters at the time of the study but prior to the weight gain. Our results unequivocally show that overweight or obesity is an important, but not mandatory prerequisite for the negative change in seminal fluid parameters.

We performed the double microbiological examination of urine and ejaculate in order to exclude an inflammatory process as the cause of the negative change in seminal fluid parameters. Our results show that even after antibiotic treatment, where necessary and negative repeat results, seminological disorders of the same degree of severity continued to persist.

CONCLUSIONS

1. Our study shows that in a number of men at a young age, some decrease in normal testosteronå secretion occurs, with a concomitant negative change in seminal fluid parameters, which is remarkably different from the same parameters in their peers with a high normal testosterone level.

2. We identify permanent low normal testosterone, overweight and obesity as predictors, signaling a possible negative change in seminal fluid parameters.

3. We can say that if obesity plays some role in negative change in seminal fluid parameters, the mechanism in most cases is most likely related to the sustained low normal T level, as a result of increased adipose tissue.

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REFERENCES:

1. De Kretser D. Endocrinology of male infertility. *Br Med Bull.* 1979 May;35(2):187-92. [PubMed]

2. Pierrepoint CG, Jenkins BM, Wilson DW, Gow JG. An examination of blood steroid and gonadotropin concentrations in relation to fertility status and testicular function in men. *Fertil Steril.* 1982 Oct;38(4):465-70. [PubMed]

3. Di Guardo F, Vloeberghs V, Bardhi E, Blockeel C, Verheyen G, Tournaye H, et al. Low Testosterone and Semen Parameters in Male Partners of Infertile Couples Undergoing IVF with a Total Sperm Count Greater than 5 Million. *J Clin Med.* 2020 Nov 26; 9(12):3824. [PubMed]

4. Meeker JD, Godfrey-Bailey L, Hauser R. Relationship between serum hormone levels and semen quality among men from an infertility clinic. *J Androl.* 2007 May-Jun;28(3): 397-406. [PubMed]

5. Jorgensen N, Joensen U, Toppari J, Punab M, Erenpreiss J, Zilaitiene B, et al. Compensated reduction in Leydig cell function is associated with lower semen quality variables: A study of 8182 European

young men. *Hum Reprod.* 2016 May;31(5):947-57. [PubMed]

6 Olesen IA, Joensen UN, Petersen JH, Almstrup K, Rajpert-De Meyts E, Carlsen E, et al. Decrease in semen quality and Leydig cell function in infertile men: a longitudinal study. *Hum Reprod.* 2018 Nov 1; 33(11): 1963-1974. [PubMed]

7. Wu FC, Tajar A, Beynon JM, Pye SR, Silman AJ, Finn JD, et al. Identification of late-onset hypogonadism in middle-aged and elderly men. *N Engl J Med.* 2010 Jul 8; 363(2):123-35. [PubMed]

8. MacDonald AA, Herbison GP, Shovell M, Farquhar CM. The impact of body mass index on semen parameters and reproductive hormones in human males: a systemic review with meta-analysis. *Hum Reprod.* 2010 May-Jun; 16(3): 293-311. [PubMed]

9**.** MacDonald AA, Stewart AW, Farquhar C. Body mass index in relation to semen quality and reproductive hormones in New Zealand men: a cross sectional study in fertility clinics. Hum Reprod. 2013 Dec; 28(12): 3178-87.

10. Salas-Huetos A, Maghsoumi-Norouzabad L, James ER, Carrell DT, Aston KI, Jenkins TG, et al. Male adiposity, sperm parameters and reproductive hormones: An updated systematic review and collaborative meta-analysis. *Obes Rev.* 2021 Jan; 22(1):e13082. [PubMed]

11. Sermondade N, Faure C, Fezeu L, Shayeb AG, Bonde JP, Jensen TK et al. BMI in relation to sperm count: an updated systematic review and collaborative meta-analysis. *Hum Reprod Update.* 2013 May; 19(3):221-231. [PubMed]

12. Zhong O, Ji L, Wang J, Lei X, Huang H. Association of diabetes and obesity with sperm parameters and testosterone levels: a meta-analysis. *Diabetol Metab Syndr.* 2021 Oct 16; 13(1): 109. [PubMed]

13. Ma JX, Wang B, Li HS, Jiang XJ, Yu J, Ding CF, et al. Association between obesity-associated markers and semen quality parameters and serum reproductive hormones in Chinese infertile men. *Reprod Biol Endocrinol.* 2020 Sep 29; 18(1): 95. [PubMed]

14. Bhasin S, Cunningham G, Hayes FJ, Matsumoto AM, Snyder PJ, Swerdloff RS, et al. Testosterone therapy in adult men with androgen deficiency syndromes: an endocrine society clinical practice guideline. *J Clin Endocrinol Metab.* 2006 Jun; 91(6):1995-2010. [PubMed]

15. Harman SM, Metter EJ, Tobin JD, Pearson J, Blackman MR. Longitudinal effects of aging on serum total and free testosterone levels in healthy men. Baltimore Longitudinal Study of Aging. *J Clin Endocrinol Metab.* 2001 Feb; 86(2):724-31. [PubMed]

16. Wang C, Swerdloff RS, Iranmanesh A, Dobs A, Snyder PJ, Cunningham G, et al. Transdermal testosterone gel improves sexual function, mood, muscle strength and body composition parameters in hypogonadal men. Testosterone Gel Study Group. *J Clin Endocrinol Metab.* 2000 Aug; 85(8):2839-53. [PubMed]

17. Kirilov G. [Problematics in the study of testosterone as the main test for assessing androgen deficiency in men. In: Male Reproductive Endocrinology.] [in Bulgarian] Academic Publishing House "Marin Drinov", Sofia. 2013, 159-181.

18. Weir CB, Jan A. BMI Classification Percentile And Cut Off Points. In: StatPearls [Internet]. Treasure Island (FL): StatPearls Publishing. 2022 Jun 27. [PubMed]

19. WHO laboratory manual for the examination and processing of human semen. Sixth Edition. World Health Organization. Jul 27, 2021. [Internet]

20. Minhas S, Bettocchi C, Boeri L, Capogrosso P, Carvalho J, Cilesiz NC, et al. European Association of Urology Guidelines on Male Sexual and Reproductive Health: 2021 Update on Male Infertility. *Eur Urol.* 2021 Nov; 80(5):603-620. [PubMed]

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