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ORIGINAL PAPER

Once-a-day Tadalafil administration improves the spermogram parameters in fertile patients

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Summary ***Objectives: We explored the safety of Tadalafil once-daily treatment for 12 week and its effects on semen quality in a clinical subpopulation of men with psychogenic erectile dysfunction (ED). Patients and Methods: Twenty-seven men, between 19 and 35 years, unaware of their fertility status, suffering from psychogenic ED were enrolled. The examination of the seminal fluid was performed twice before administration of Tadalafil and twice after three months of continuous daily administration of Tadalafil 5 mg. The volume of the seminal fluid, the concentration of sperm, the concentration of sperm with type "a+b" motility, the concentration of sperm with type "a" motility, the concentration of normal sperm were taken into consideration. The results before treatment with Tadalafil (T0) and after 3 months of treatment (T3m) were compared.***
Results: The administration of once-daily Tadalafil 5mg, brings to an average increase of the total number of sperm cells, both total and fast motility (type a) and the percentage of nemasperms, and to an average increase of semen volume of only 0.41 ml. These quantitative and qualitative improvements of the seminal fluid resulted statistically significant as regard motility, nemasperm percentage and seminal fluid volume. No unespected safety findings were observed.
Conclusions: Tadalafil administration improves the quality of sperm cells and seminal fluid: in particular motility, percentage of nemasperms and volume of seminal fluid. We emphasize the safety of the once-daily treatment with tadalafil 5mg and the positive effects on spermatogenesis.

KEY WORDS: PDE5 inhibitor; Tadalafil; Spermiogram.

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INTRODUCTION

Tadalafil is a phosphodiesterase 5 (PDE5) inhibitor and it is specifically used for treatment of erectile dysfunction (ED). Additional beneficial effects have been reported in other fields different from andrology, such as in the treatment of pulmonary hypertension (1), LUTS (2), Raynaud's syndrome (3). On the contrary, no adverse effects on spermatogenesis and on the production of sexual hormones were proven (4). Due to its characteristics of duration of action it may be administered daily rather than on-demand. Good tolerability and very few side effects have been amply demonstrated. Its daily administration has a dual effect: the curative effect on the organic erectile dysfunction with predominant vascular component, and the reduction of the psychological influence that, on the contrary, the on-demand administration can enhance.

Sperm motility is a significant parameter for the assessment of the fertilizing capacity of the subject under examination, just like the concentration and morphology of the sperm cells. The cAMP plays a role for the up-regulation of tyrosine phosphorylation, that determines the sperm capacitation and motility. Recently, it has been tried to influence the phosphorylation of tyrosine to increase sperm motility of normo or asthenic semen samples using pentoxifylline, which is a PDE inhibitor. *Yunes* (5) suggests that the increased motility determined by pentoxifylline is mediated by the tyrosine phosphorylation in the sperm tail. The levels of cAMP are regulated by two metabolic reactions: synthesis from ATP through the adenylyl cyclase and its degradation by its cyclic nucleotide (6).

This study has the aim to test the effects of the daily administration of a PDE inhibitor like tadalafil 5 mg on semen quality.

MATERIALS AND METHODS

The aim of the study was to evaluate the effects of Tadalafil 5 mg/daily administered for 3 consecutive months on semen quality in fertile patients. Twenty-seven young patients, from 19 to 35 years, were enrolled (average 29.6 years) suffering from psychogenic erectile dysfunction. They were not aware of their fertility status. Their consent to perform the semen analysis was obtained. They were fertile according to the evaluation criteria of the WHO 2010 (7), with a sperm concentration exceeding 15.000.000/ml., a progressive motility (a+b type) ≥ 35%, volume > 1.5 ml, and normal morphology > 4%. Inclusion criteria were: general state of good health, none chronic medical therapy in the period of three months before the first semen analysis, absence of morphological or vascular alterations at the Doppler ultrasonography, FSH, LH and testosterone levels in the normal range, age not exceeding 35 years. Exclusion cri-

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teria were: smoking, chronic alcoholism, drug use, history of cryptorchidism, prostatitis and infectious-inflammatory diseases of the genitals, varicocele (also already solved surgically), sexually transmitted diseases, urinary tract infections, leucocyturia, leukocytospermia. The diagnosis of psychogenic erectile dysfunction has been confirmed by the administration of the IIEF questionnaire, anamnesis, physical examination and dynamic penile ultrasound Doppler with10 mcg of PGE1.

Methods of collection and analysis of the semen sample
The spermatogenic cycle is completed within about 72 days (8), so we considered sufficient to assess the effects of chronic administration of Tadalafil 5 mg for 3 months, thus covering the whole maturation cycle of the single sperm. The examination of the seminal fluid was then performed twice before administration of Tadalafil (after two weeks from the first one) and twice after three months of continuous daily administration of 5 mg Tadalafil (always at a distance of two weeks), always by the same operator in the same laboratory (the *Centre of Andrology and Reproductive Pro.Andròs* certified ISO 9001-2008 and certified SIA) in accordance with WHO guidelines 2010. So ultimately each patient was subjected to four tests of the seminal fluid, before and immediately after the administration of three months of Tadalafil 5 mg, once-a-day. The collection of the liquid has been rigorously performed in the Centre with a planned withdrawal for 4 days, via masturbation, after cleaning the genitals with liquid soap at physiological pH. The ejaculated fluid was collected directly into sterile containers for urine culture.

Adverse effects or side effects
For all subjects of the study there were no significant variations of the side or unwanted effects of Tadalafil compared to those described in the literature. In any case, no one was forced to discontinue administration, demonstrating the good tolerability of 5 mg, once-a-day.

Results
During the administration of tadalafil no unespected safety findings were observed.
Of each semen analysis, the volume of the seminal fluid (ml), the concentration of sperm (millions of sperm/ml), the concentration of sperm with type "a+b" motility, the concentration of sperm with type "a" motility, the concentration of normal sperm were taken into consideration. The average results of the two spermiograms before treatment with Tadalafil (average T0) and the average results of the two spermiograms after 3 months of treatment (average T3m) were calculated. The "*T0 average results*" were compared with the "*T3m average resuls*" and the differences were calculated.
The mean values of pre-treatment spermiograms (spermiogram I and II) were compared with the average values of spermiograms at the 10^{th} and 12^{th} week of treatment with Tadalafil 5 mg (spermiogram III and IV).
The results were subjected to analysis of variance using "*Student's test*". The differences are statistically significant (Table 1)
What we observed is even more evident in the diagrams that compare the confidence intervals (CI) and the averages of spermiograms' parameters before and after therapy (Table 2).
As showed in the reported diagrams reported, the Confidence Intervals (CI) tend to overlap, but with the averages shifted upward, as it is shown by the graphs that describe the changes in each subjects (an example is in Table 3)

Discussion
Tadalafil (*Cialis®*) is used in the treatment of erectile dysfunction as a potent, reversible, competitive inhibitor of phosphodiesterase 5 (PDE5), an enzyme that inactivates

Table 1.

	Pre treatment	Post treatment	Difference	
Concentration				
Millions/ml	44	48.7	+4.7	n.s.
a + b motility %	53.94	57.59	+3.6%	p < 0.05
Type a %	32.44	35	+2.6%	n.s.
Normal morphology %	48.91	54.74	+5.8	p < 0.05
Volume ml	1.97	2.38	+0.41	p < 0.001

Table 2.

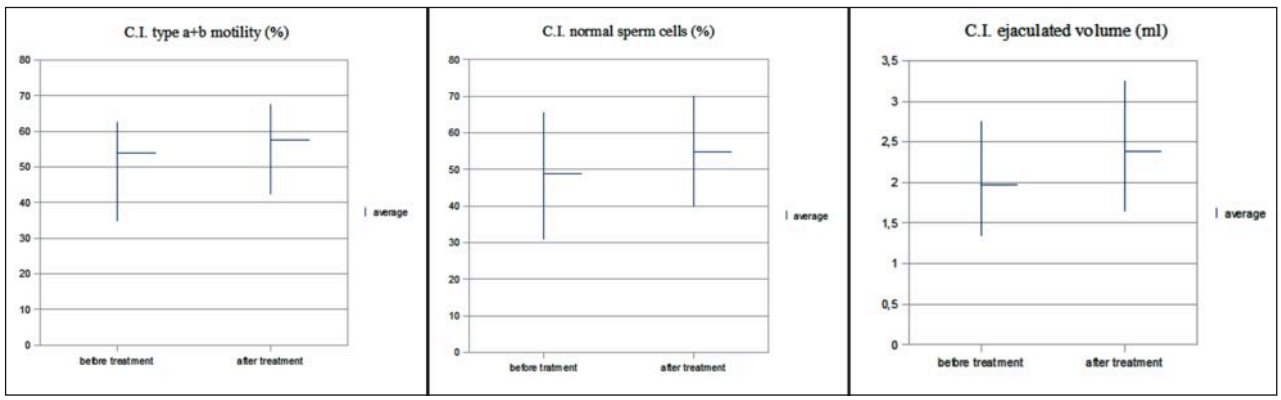
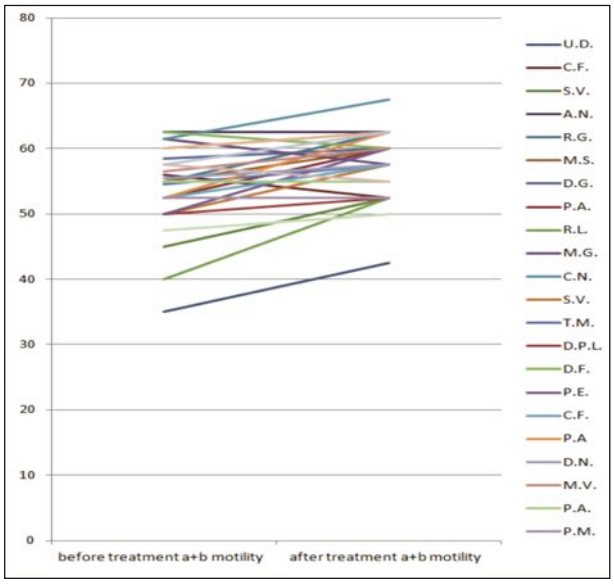


Table 3.



cyclic guanosine monophosphate (cGMP) (9, 10). Inhibition of PDE5 in the corpus cavernosum of the penis increases intracellular cGMP levels, thereby facilitating relaxation of smooth muscle and leading to penile erection (11, 12). Clinical trials with tadalafil administered orally on-demand to patients with erectile dysfunction have demonstrated enhanced erectogenic response in both the clinic setting and the at-home setting (13, 14). The recent interest of many authors to demonstrate the causal link between ED and LUTS has also led to re-evaluate the presence of NOS not only in the endothelium and smooth muscle cells of the corpora cavernosa, but also in the prostate, in the bladder and in the testicular parenchyma.

To show the causal relationship between ED and LUTS with a biological plausibility one of the most studied theories is "*the nitric oxide synthase (NOS)/NO theory*". This hypothesis attempts to explain the link between ED and LUTS by the reduced production of NOS/NO in the pelvis, including the penis, the bladder and the prostate (15). NO is a multifunctional molecule originally described as a vasodilator (16). It is synthesized from its precursor L-arginine via NOS, which exists in three isoforms; two are constitutively expressed in endothelial (eNOS) and neuronal (nNOS) structures, and produce small amounts of NO, whereas inducible NOS (iNOS) is induced by cytokines, infection or other stimuli, and produces large amounts of NO (17). It is widely accepted that NO is important in the relaxation of corpus cavernosum smooth muscle and vasculature. Neurogenic NO is considered the main factor responsible for the immediate relaxation of corpus cavernosum, while endothelial NO is essential for maintaining relaxation (18). Conditions associated with reduced function of nerves and endothelium, e.g. ageing, hypertension, smoking, hypercholesterolaemia and diabetes, can cause circulatory and structural changes in penile tissues, resulting in ED. NO is also present in the human prostate and bladder and putatively modulates smooth muscle tone. Histochemical staining and immunohistochemistry confirmed dense nitrinergic innervation of glandular epithelium, fibromuscular stroma and blood vessels in the normal human prostate (19).

We are interested to the effect of NO on fertility. *Liman et al.* (20) studied the cellular localization of the endothelial (eNOS) and inducible (iNOS) forms of nitric oxide (NO) synthase in the cat testis, using enzyme histochemical and immunohistochemical techniques. Stage-dependent nuclear and cytoplasmic eNOS/iNOS immunoreactivity were found in all germ cells, including spermatogonia, primary spermatocytes (preleptotene, zygotene, and pachytene spermatocytes), and round and elongating spermatids of the seminiferous epithelium. The pachytene spermatocytes exhibited strong positive reactions at all spermatogenic stage. Interestingly, in elongated spermatids eNOS and iNOS immunostainings was observed only in the cytoplasm but not in the nuclei. eNOS and iNOS immunolabeling was observed in the acrosomal vesicle of some round spermatids and in the acrosomal cap of elongating spermatids at stage II. Positive reactions were also observed in the Sertoli and Leydig cells as well as in other tissues including vascular endothelial and smooth muscle cells and peritubular myoid cells. These results suggest that NO may play an important role in chromatin condensation, spermatid shaping, and the final release of sperm from the spermatogenic epithelium. Furthermore, NO may also be involved in spermiogenesis, steroidogenesis, and apoptotic cell death. It is therefore likely that the NO-mediated mechanisms on prostate and testicular parenchyma, increased by Tadalafil, is the basis of the improvement of the quality of the sperm.

Conclusions

The comparison between the parameters of spermiograms, obtained before and after administration of Tadalafil 5 mg, showed an average increase in the number of sperm, both total and fast motility (type a) and the percentage of nemasperms and an average increase of semen volume of 0.41 ml. These quantitative and qualitative improvements of the seminal fluid are statistically significant, as regard total progressive motility (a+b), normal sperm percentage and seminal fluid volume. The administration of Tadalafil has just directed the production of sperm and seminal fluid to the highest values. It is just like the testicles were pushed toward their highest "*functional reserve*". In addition, we emphasize the safety of the once-daily treatment with tadalafil 5mg and the absence of negative effects on spermatogenesis. It would be interesting to test whether the administration of Tadalafil may improve in a statistically significant way the seminal fluid of patients with abnormalities of the baseline spermiogram (at time 0).

References

1. Affuso F, Palmieri EA, et al. Tadalafil improves quality of life and exercise tolerance in idiopathic pulmonary arterial hypertension. Int J Cardiol. 2006; 108:429-31

2. Gonzalez RR, Kaplan SA. Tadalafil for the treatment of lower urinary tract symptoms in men with benign prostatic hyperplasia. Expert Opin Drug Metab Toxicol. 2006; 2:609-17.

3. Baumhaekel M, Scheffler P, et al. Use of tadalafil in a patient with a secondary Raynaud's phenomenon not responding to sildenafil. Microvasc Res. 2005; 69:178-9.

4. Hellstrom WJ, Overstreet JW, et al. Tadalafil has no detrimental effect on human spermatogenesis or reproductive hormones. J Urol. 2003; 170:887-91.

5. Yunes R, Fernández P, et al. Cyclic nucleotide phosphodiesterase inhibition increases tyrosine phosphorylation and hyper motility in normal and pathological human spermatozoa. Biocell. 2005; 29:287-93.

6. Bentley JK, Beavo JA. Regulation and function of cyclic nucleotides. Curr Opin Cell Biol. 1992; 4:233-40.

7. World Health Organization. WHO laboratory manual for the examination and processing of human semen. 5th edition, Cambridge University press, Cambridge, 2010.

8. Meistrich ML, Hess RA. Assesment of spermatogenesis through staging of seminiferous tubules. Methods Mol Biol 2013; 927: 299-307.

9. Francis SH, Corbin JD. Cyclic GMP: synthesis, metabolism, and function. In Advances in Pharmacology, ed. Murad P. New York: Academic Press 1994; 115-70.

10. Padma-Nathan H, McMurray JG, Pullman WE, et al. On-demand IC351 (Cialis®) enhances erectile function in patients with erectile dysfunction. Int J Impot Res 2001; 13:2-9.

11. Thompson WJ. Cyclic nucleotide phosphodiesterases: pharmacology, biochemistry and function. Pharmacol. Ther. 1991; 51:13-3.

12. Lue TF. Erectile dysfunction. N Engl J Med. 2000; 342:1802-13.

13. Brock GB, McMahon CG, Chen KK, et al. Efficacy and safety of tadalafil for the treatment of erectile dysfunction: results of integrated analyses. J Urol. 2002; 168: 1332-6.

14. Carson CC, Rajfer J, Eardley I, et al. The efficacy and safety of tadalafil: an update. Br J Urol Int. 2004; 93: 1276-81.

15. McVary KT, McKenna KE. The relationship between erectile dysfunction and lower urinary tract symptoms: epidemiological, clinical, and basic science evidence. Curr Urol Rep. 2004; 5:251-7.

16. Palmer RM, Ferrige AG, Moncada S. Nitrite oxide release accounts for the biological activity of endotheliumderived relaxing factor. Nature. 1987; 327:524-6.

17. Marletta MA. Nitrite oxide synthase structure and mechanism. J Biol Chem. 1993; 268: 12331-4.

18. Andersson KE. Erectile physiological and pathophysiological pathways involved in erectile dysfunction. J Urol. 2003; 170: S6-14.

19. Bloch W, Klotz T, Loch C, et al. Distribution of nitric oxide synthase implies a regulation of circulation, smooth muscle tone, and secretory function in the human prostate by nitric oxide. Prostate. 1997; 33: 1-8.

20. Liman N, Alan E, Beyaz F, Gürbulak K. Endothelial and inducible nitric oxide synthase (NOS) immunoreactivity and NOS-associated NADPH-diaphorase histochemistry in the domestic cat (Felis catus) testis. Theriogenology. 2013; 80:1017-32.

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