Archives of Stem Cell and Therapy

Campbell et al., Arch Stem Cell Ther 2021; 2(1):8-11.

Commentary

Updated protocols for optimizing sperm recovery after steroid use

Kevin J Campbell, John F. Sullivan, Larry I. Lipshultz*

Scott Department of Urology, Baylor College of Medicine, Houston, TX, USA

*Author for correspondence: Email: larryl@bcm.edu

Received date: December 18, 2020
Accepted date: March 08, 2021

Abstract

The prevalence of hypogonadism is an increasing problem that affects increasingly more men of reproductive age. With the mainstay of hypogonadal treatment involving testosterone therapy (TTh), the fertility potential of many of these men must be investigated and considered accordingly. There exist multiple treatments for the recovery of anabolic steroid-induced hypogonadism, including gonadotropin replacement therapy to induce both spermatogenesis as well as intratesticular testosterone production. The use of follicular stimulating hormone (FSH) has been reported to decrease the time to recovery of spermatogenesis in anabolic steroid-induced azoospermia. A new formulation of testosterone in the form of tri-daily applied intranasal testicular gel mimics gonadotropin releasing hormone (GnRH) pulsatile release of luteinizing hormone (LH) to promote production of testosterone and decreases feedback inhibition of spermatogenesis. Additionally, immature testicular tissue transplant may aid in the fertility preservation for patients anticipating significant suppression of spermatogenesis.

Keywords: Spermatogenesis, Anabolic, Testosterone, Hypogonadism, Follicle stimulating hormone, Enclomiphene

Introduction

Hypogonadism is a clinical condition characterized by low serum testosterone levels in addition to global symptoms which include malaise, fatigue, decreased libido, erectile dysfunction, and mood changes. Hypogonadism has also been linked to osteoporosis, cardiovascular disease, and early onset diabetes [1].

As has been previously discussed, testosterone therapy (TTh) may be initiated in the male for a myriad of reasons, including but not limited to, primary and secondary hypogonadism, testosterone deficiency in the ageing male, and to optimize body musculature [2]. The use of TTh for hypogonadism continues to grow and more younger men are now receiving treatment; as many as 12.4% of all testosterone prescriptions are written for men <39 years of age [3]. Anabolic steroid-induced infertility is a commonly seen diagnosis in the male of reproductive age who presents to a men's health clinic. An estimated 3 million men are on TTh; however, exogenous testosterone use can disrupt the hypothalamus-pituitary-gonadal (HPG) axis and markedly impair spermatogenesis [4].

With use of exogenous TTh, the HPG axis becomes quiescent and endogenous testosterone production ceases. In addition, in most men there is cessation of sperm production within the testes due to decreased gonadotropin secretion [5]. In men who have previously taken TTh and now desire fertility, various pharmacological agents are employed to stimulate endogenous testosterone production and restore spermatogenesis [2]. It has previously been reported that the use of human chorionic gonadotropin (hCG) in addition to selective estrogen receptor modulators (SERMs) such as clomiphene citrate have been shown to result in return of sperm to the ejaculate in testosterone-induced azoospermia [6] (Figure 1).

The spontaneous recovery of spermatogenesis after cessation of TTh is possible but may take months to years and the associated hypogonadal symptoms can be devastating. HCG is a naturally occurring protein that mimics LH and may be used as a therapy to support the return of spermatogenesis quickly with minimal side effects [6]. Studies have shown that testosterone-induced infertile patients can recover sperm in the ejaculate in when treated with HCG combined with clomiphene citrate, tamoxifen, anastrozole, or FSH [2,6].

Citation: Campbell KJ, Sullivan JF, Lipshultz LI. Updated protocols for optimizing sperm recovery after steroid use. Arch Stem Cell Ther 2021; 2(1):8-11.

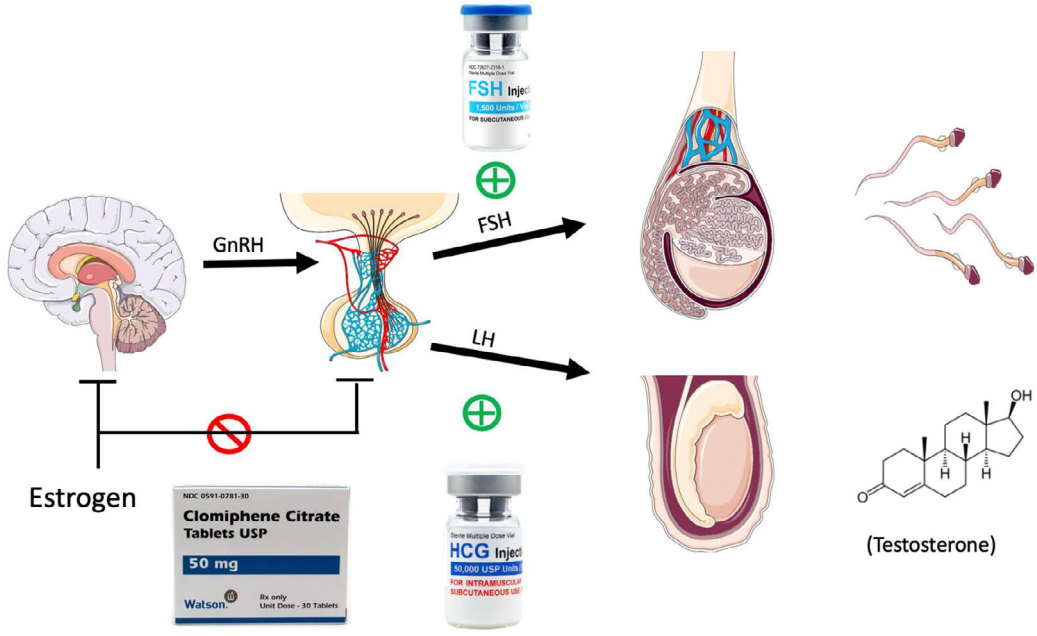

Figure 1: Hypothalamic-pituitary-gonadal axis. HCG mimics LH to induce Leydig cells testosterone production in the testicles. FSH acts on Sertoli cells to promote spermatogenesis. SERMs such as clomiphene citrate block the negative feedback on the hypothalamus and anterior pituitary to increase LH and FSH gonadotropic secretion.

Follicle Stimulating Hormone

Testosterone is synthesized and secreted by the testicular interstitial Leydig cells. The process of spermatogenesis takes 72-74 days and is dependent on high concentrations of intratesticular testosterone of approximately 40-fold higher that the serum testosterone levels [7]. Follicular stimulating hormone (FSH) greatly enhances spermatogenesis by stimulating the functions of Sertoli cells and increasing mitoses of spermatogonia. Once mitosis has been initiated in spermatogonia, testosterone alone can maintain spermatogenesis.

In lieu of upstream stimulation, exogenous FSH has been used to potentiate the recovery of Sertoli cell function recovery and sperm production [6,8]. Studies have suggested the increased effectiveness of adding FSH to HCG as a substitute for clomiphene [6,9].

A recent review of anabolic-induced azoospermic patients undergoing spermatogenesis recovery with FSH were noted to have a faster return of sperm to the ejaculate than those on clomiphene [9]. The results were 6.4 and 14.8 months, respectively. A review of patients who had previously failed recovery of spermatogenesis on clomiphene and underwent a second attempt on FSH revealed a 73% pregnancy rate. These results reiterate that FSH in combination with hCG may be considered as an alternative to combination hCG and clomiphene in the treatment of testosterone-induced azoospermia. Thus, FSH and hCG dual therapy may result in more rapid recovery of sperm to the ejaculate. Additionally, patients who have failed dual therapy with hCG and clomiphene should be considered for subsequent FSH treatment.

Enclomiphene

Clomiphene citrate has been used as an agent to increase sex hormones LH and FSH and intratesticular testosterone in hypogonadal men. In the anabolic-steroid induced hypogonadal male, gonadotropins are routinely low due to negative feedback from elevated levels of exogenous androgens. Enclomiphene citrate is a transisomer of clomiphene citrate that has been proposed as treatment in men with secondary hypogonadism. The single isomer of enclomiphene demonstrates pure estrogen antagonism and has improved activity compared to a mixture of isomers found in clomiphene in both antagonism and agonism activities.

In a phase 2 clinical trial, Wiehle et al. evaluated 73 men with secondary hypogonadism taking oral enclomiphene citrate and 1% topical testosterone gel [10]. Serum gonadotropins, testosterone, and semen analyses were analyzed over 3 months. The investigators found that men in the enclomiphene citrate arms had or continued to have normal sperm parameters while men treated in a topical testosterone arm showed declining sperm counts. Enclomiphene 25mg raised baseline testosterone (n 32 average 209.8 ng/dL to 405.8 ng/dL) compared to placebo (n=26, average 213.7 ng/dL to 198.5 ng/dL) and was comparable to topical 1% testosterone (n=30, average 210 ng/dL to 462.6 ng/dL). FSH (mIU/mL) was increased compared to topical testosterone and placebo at 9.4 to 14.9, 6.0 to 2.4, and 6.1 to 5.4, respectively. LH (mIU/mL) was increased compared to topical testosterone and placebo at 5.3 to 11.7, 3.9 to 1.4, and 3.9 to 3.7, respectively. There were no men with sperm concentration $<15 \times 10^6$/mL on at least 2 semen analyses following completion of enclomiphene dosing. Additionally, the safety profile of both 12.5 mg

and 25 mg daily dose levels of enclomiphene citrate was acceptable.

Intranasal Testosterone

The use of TTh in a dose-dependent and pulsatile-mimicking application cycle is a current area of focus for researchers seeking to combat hypogonadal symptoms in men trying to recover spermatogenesis following the withdrawal of conventional TTh. With the cessation of TTh, these patients are often rendered symptomatically hypogonadal. Recent preliminary results show the potential to offset hypogonadism symptoms that accompany exogenous testosterone cessation through administration of intranasal testosterone gel [8,11].

Natesto is a nasal administered exogenous 4.5% testosterone gel, administered from a non-pressurized, manual pump dispenser with a specialized nasal applicator which administers 125 uL (5.5mg of testosterone). Previous studies have shown that a single nasal dose has a rapid absorption with a Tmax at 60 minutes, a half-life that ranged between 10-100 minutes [12]. Three to four daily doses achieve eugonadal levels of circulating testosterone comparable to normal pulsatile regulated release of testosterone. It has also been suggested that men on Natesto maintain FSH and LH levels as well as total motile sperm counts with normal range [8,11].

In a recent study performed by Ramasamy et al., the authors completed a prospective, open label, single arm trial that evaluated the impact of intranasal testosterone three times daily on hypogonadal patients over a 6-month period [11]. The authors investigated the patients for changes in LH, FSH, sexual function, and quantitative changes in sperm concentration and total motile count. In the majority of these hypogonadal men, testosterone levels had statistically significant improvement to the upper tercile, while FSH and LH were decreased but maintained within normal limits. When evaluated over the 3- and 6-month time frames, intranasal testosterone gel demonstrated a decline in sperm concentration and total motile count. The study completed after 6 months though the sperm parameters' nadir had not been demonstrated. Additionally, the mean International Index of Erectile Function (IIEF) scores showed marginal improvement from 23.1 to 24.3. While patients in this study had not previously been on TTh, the clinical application of this therapy for clinically symptomatic hypogonadal males who have ceased TTh in an effort to recover spermatogenesis remains an area of hopeful investigation.

Additional Therapies

The pathology in anabolic steroid-induced azoospermia begins its reversal with removal of the offending agent and the induction of spermatogenesis is then augmented with supplemental agents. Though removal of the steroids may be all that is needed for recovery of spermatogenesis, there are other factors to consider which may be contributors to the azoospermia.

Varicocele

A varicocele is present in 15% of the male population and may be found in 19-40% of men as a citing factor in men who present to an infertility clinic [13]. Varicoceles are dilations of the veins of the pampiniform plexus thought to be caused by increases in vascular pressure from anatomical variants or abnormalities in the normal anti-reflux venous system. The vascular dilation leads to pooling of blood in close proximity to the testicle. The temperature of the testicle increases and may lead to decreases in spermatogenesis as well as testosterone production [13]. Varicoceles may be the contributing factor in approximately 5% of males with azoospermia [14]. Weedin et al performed a meta-analysis in 2010 to evaluate the effectiveness of varicocele repair in the recovery of spermatogenesis in men with nonobstructive azoospermia [15]. A total of 233 men with clinical varicoceles underwent repair with a mean 13-month follow-up. Postoperative semen analysis revealed motile sperm in 39% of men leading to 14 reported spontaneous pregnancies.

Stem cell transplants

Immature testicular tissue transplant may aid in the fertility preservation for patients anticipating suppression of spermatogenesis. Cancer survivorship has seen the rise of an important co-endpoint in the ability to maintain fertility potential after treatment. There is a rising incidence of all cancers diagnosed in the United States in those younger than 65 years of age with 9% of cancer diagnoses in those younger than 45 and 1% younger than 20 years of age [16]. In these patients, barriers to fertility can be associated with derangements of the HPG axis, surgical deficits, induced sexual dysfunction, as well as spermatogonial insult or depletion from chemotherapeutic and radiation treatment. The survival rate of children with cancer is over 80%, and 30% of survivors will be infertile as adults [17]. The risk for prolonged or permanent infertility following 2 years of therapy cessation is over 25% of patients on alkylating agents and in 20-47% of patients on platinum agents [16]. Antimetabolites, nucleoside analogs, vinka alkaloids, and antitumor/antibiotic described regimens have been shown to carry a temporary or low risk for infertility [16]. With this in mind, the need to preserve fertility prior to oncological treatment is emphasized by the ASCO guidelines [18]. Post-pubertal males may cryopreserve ejaculated sperm. This is a safe and non-invasive form of sperm preservation with success rates approaching 90% [19]. Reports have demonstrated that cryopreserved sperm may maintain viability up to 28 years, giving a long interval for patients to determine their reproductive timing [20].

In the pre-pubertal patient, cryopreservation of ejaculated sperm is often not feasible. In these patients who have not yet have active spermatogenesis, the only current options are experimental. Immature testicular tissue (ITT) obtained *via* surgical testicular sperm extraction (TESE) has been investigated for cryopreservation and future use. Isolation and auto-transplant or *in vitro* maturation of germ cells for subsequent IVF/ICSI. In 1999 Radford and colleagues biopsied and cryopreserved testicular cell suspensions in 11 patients prior to cancer treatments for non-Hodgkin's lymphoma [21]. In 5 of the patients, testicular cells were later reintroduced into the testes after cancer therapy completion, though their fertility status is unknown [21].

Xenotransplantation of cryopreserved ITT in nude mice has demonstrated survival in a limited proportion of spermatogonia and mitotic ability as well as differentiation initiation [22]. There is a concern for the loss of germ cells due to ITT grafting and tissue engineering has been a focus at improving the cellular environment to enhance transplantation outcomes by optimizing early tissue graft revascularization, protecting cells from toxic insults and ischemic injury, and searching for strategies to promote cellular differentiation [23]. Proof of principle has been provided by cryopreserved prepubertal testicular tissue autologously grafted under the back or scrotal skin of castrated pubertal Rhesus macaques which matured to produced functional sperm [24].

Conclusion

There exist multiple strategies in the andrologist's armamentarium to aid in the recovery or maintenance of spermatogenesis following impairment from anabolic steroids, anatomical variants, or gonadotoxic onco-therapeutics. Sperm cryopreservation is the best pre-treatment for future fertility, as in the case of patients anticipating suppression of spermatogenesis. However, many patients present post-therapy with a desire to restore spermatogenesis after the initial insult has occurred. The recovery of spermatogenesis in these patients is linked with the interplay between hormones within the HPG. HCG, SERMs, and FSH offer direct and indirect testicular signaling to induce spermatogenesis and the recovery or maintenance of fertility. In these cases, cessation of testosterone often renders the patient hypogonadal. Novel therapies as with FSH, enclomiphene citrate, and intranasal testosterone offer exciting strategies to combat hypogonadal symptoms while enhancing spermatogenic potential.

References

1. Mulhall JP, Trost LW, Brannigan RE, Kurtz EG, Redmon JB, Chiles KA, et al. Evaluation and management of testosterone deficiency: AUA guideline. The Journal of Urology. 2018 Aug;200(2):423-32.
2. Tatem AJ, Beilan J, Kovac JR, Lipshultz LI. Management of Anabolic Steroid-Induced Infertility: Novel Strategies for Fertility Maintenance and Recovery. The World Journal of Men's Health. 2019 Mar 26;38(2):141-50.
3. Layton JB, Li D, Meier CR, Sharpless JL, Stürmer T, Jick SS, et al. Testosterone lab testing and initiation in the United Kingdom and the United States, 2000 to 2011. The Journal of Clinical Endocrinology and Metabolism. 2014 Mar;99(3):835.
4. McBride JA, Coward RM. Recovery of spermatogenesis following testosterone replacement therapy or anabolic-androgenic steroid use. Asian Journal of Andrology. 2016 May;18(3):373.
5. de Souza GL, Hallak J. Anabolic steroids and male infertility: a comprehensive review. BJU International. 2011 Dec;108(11):1860-5.
6. Wenker EP, Dupree JM, Langille GM, Kovac J, Ramasamy R, Lamb D, et al. The use of HCG-based combination therapy for recovery of spermatogenesis after testosterone use. The Journal of Sexual Medicine. 2015 Jun;12(6):1334-7.
7. Coviello AD, Bremner WJ, Matsumoto AM, Herbst KL, Amory JK, Anawalt BD, et al. Intratesticular testosterone concentrations comparable with serum levels are not sufficient to maintain normal sperm production in men receiving a hormonal contraceptive regimen. Journal of Andrology. 2004 Nov 12;25(6):931-8.
8. Masterson T, Best J, Bitran J, Molina M, Ibrahim E, Kaiser U, et al. PD58-05 The Effect of Natesto on Spermatogenesis, Reproductive Hormones, and Hypogonadal Symptoms. A phase IV study. The Journal of Urology. 2020 Apr;203(Supplement 4):e1201-2.
9. Campbell K, Sullivan J, Valero-Carrion R, Kraus J, Stocks B, Lawrence A, et al. Dual therapy with rFSH and HCG results in a significantly shorter time to return of spermatogenesis than combination HCG and Clomiphene. J Sex Med. 2020.
10. Wiehle RD, Fontenot GK, Wike J, Hsu K, Nydell J, Lipshultz L, et al. Enclomiphene citrate stimulates testosterone production while preventing oligospermia: a randomized phase II clinical trial comparing topical testosterone. Fertility and Sterility. 2014 Sep 1;102(3):720-7.
11. Ramasamy R, Masterson TA, Best JC, Bitran J, Ibrahim E, Molina M, et al. Effect of natesto on reproductive hormones, semen parameters and hypogonadal symptoms: a single center, open label, single arm trial. The Journal of Urology. 2020 Sep;204(3):557-63.
12. Rogol AD, Tkachenko N, Bryson N. Natesto™, a novel testosterone nasal gel, normalizes androgen levels in hypogonadal men. Andrology. 2016 Jan;4(1):46-54.
13. Pastuszak AW, Wang R. Varicocele and testicular function. Asian Journal of Andrology. 2015 Jul;17(4):659.
14. Esteves SC, Glina S. Recovery of spermatogenesis after microsurgical subinguinal varicocele repair in azoospermic men based on testicular histology. International Brazilian Journal of Urology. 2005 Dec;31(6):541-8.
15. Weedin JW, Khera M, Lipshultz LI. Varicocele repair in patients with nonobstructive azoospermia: a meta-analysis. The Journal of Urology. 2010 Jun 1;183(6):2309-15.
16. McBride JA, Lipshultz LI. Male fertility preservation. Current Urology Reports. 2018 Jul;19(7):1-1.
17. Thomson AB, Campbell AJ, Irvine DS, Anderson RA, Kelnar CJ, Wallace WH. Semen quality and spermatozoal DNA integrity in survivors of childhood cancer: a case-control study. The Lancet. 2002 Aug 3;360(9330):361-7.
18. Oktay K, Harvey BE, Partridge AH, Quinn GP, Reinecke J, Taylor HS, et al. Fertility preservation in patients with cancer: ASCO clinical practice guideline update. J Clin Oncol. 2018 Jul 1;36(19):1994-2001.
19. Brook PF, Radford JA, Shalet SM, Joyce AD, Gosden RG. Isolation of germ cells from human testicular tissue for low temperature storage and autotransplantation. Fertility and Sterility. 2001 Feb 1;75(2):269-74.
20. Feldschuh J, Brassel J, Durso N, Levine A. Successful sperm storage for 28 years. Fertility and Sterility. 2005 Oct 1;84(4):1017-e3.
21. Radford JA, Shalet SM, Lieberman BA. Fertility after treatment for cancer: questions remain over ways of preserving ovarian and testicular tissue.
22. Gassei K, Orwig KE. Experimental methods to preserve male fertility and treat male factor infertility. Fertility and Sterility. 2016 Feb 1;105(2):256-66.
23. Del Vento F, Vermeulen M, De Michele F, Giudice MG, Poels J, Des Rieux A, et al. Tissue engineering to improve immature testicular tissue and cell transplantation outcomes: one step closer to fertility restoration for prepubertal boys exposed to gonadotoxic treatments. International Journal of Molecular Sciences. 2018 Jan;19(1):286.
24. Fayomi AP, Peters K, Sukhwani M, Valli-Pulaski H, Shetty G, Meistrich ML, et al. Autologous grafting of cryopreserved prepubertal rhesus testis produces sperm and offspring. Science. 2019 Mar 22;363(6433):1314-9.