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# Current topics in testosterone replacement of hypogonadal men

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All forms of hypogonadism – primary, secondary and late-onset – require testosterone substitution. The indication is given when the patient presents with symptoms of androgen deficiency and the serum testosterone levels are below normal. Several testosterone preparations and modes of application are available of which those producing physiologic serum levels should be preferred e.g. preferentially transdermal gels and long-acting intramuscular testosterone undecanoate. Testosterone substitution must be monitored at regular intervals, best at 3, 6 and 12 months after initiation and then annually. Parameters for surveillance include well-being, libido and sexual activity, measurement of serum testosterone levels, haemoglobin and haematocrit, PSA and digital rectal examination, and, biannually, bone mineral density. Testosterone has positive effects on comorbidities such as obesity, metabolic syndrome, diabetes type II, cardiovascular diseases and osteoporosis.


## Introduction

The testes have the dual function of producing hormones and sperm. Hypogonadism, defined as impaired function of the testes is expressed either as testosterone deficiency or as infertility or as a combination of both. Its causes are so manifold that the reader is referred to relevant textbooks for pathophysiological details [1,2].

However, all forms of hypogonadism – primary, secondary and late-onset – are characterized by symptoms of testosterone deficiency. Hypogonadism with testosterone deficiency is the universally

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accepted indication for testosterone treatment. As practically all tissues and all organs have androgen receptors and/or may also show non-genomic response to testosterone, testosterone deficiency results in a broad spectrum of symptoms. Some of these symptoms such as fatigue, loss of libido, erectile dysfunction, depression and osteoporosis can also be caused by diseases other than hypogonadism and would not respond to testosterone treatment unless serum levels are below normal. Therefore, all current guidelines agree that the combination of symptoms of testosterone deficiency and subnormal testosterone levels are prerequisites for diagnosis and treatment ([3–5], for review [6]).

Disregarding these clear precepts, testosterone preparations appear to be increasingly prescribed and consumed for symptoms without documented testosterone deficiency. This seems to be more prevalent in the USA than in Europe as US sales of testosterone preparations have quadrupled from 2000 to 2011 although the number of low testosterone levels revealed in laboratory testing has remained constant [7]. In comparison, in the UK testosterone prescriptions have doubled within a decade [8], almost in pace with low testosterone levels detected in laboratory testing [7]. Prescribing testosterone at random without documented low testosterone levels and symptoms of testosterone deficiency is not in accordance with current good medical practice and borders on testosterone abuse, culminating in doping with androgens to enhance physical performance in sports and bodybuilding. Currently discussed unwanted side-effects of testosterone may, at least in part, be caused by improper prescribing (see below).

## Normal testosterone levels: thresholds for substitution

As stated above, good clinical practice requires that testosterone is only to be administered if symptoms of testosterone deficiency are encountered and levels are below normal. But what is the lower limit of normal?

First, due to the diurnal rhythm of testosterone in serum [9] blood samples of testosterone should always be taken in the morning hours from men with a regular day–night rhythm in order to obtain results comparable to reference values.

While there is not one fixed lower limit of normal, symptom-specific threshold levels do exist [10,11]. Loss of libido and vigour may already occur below 15 nmol/L, but certainly is present below 12 nmol/L when obesity also begins; below 10 nmol/L depressive moods, sleep disturbance, lack of concentration and diabetes type II are found and erectile dysfunction and hot flushes are reported below 8 nmol/L. These symptom-specific threshold levels may explain why the lower limit of normal and the limits for starting testosterone substitution vary in different countries [12]. The perception of the physician rather than the patient's complaints may govern prescribing behaviour.

As loss of libido is the most frequent and an early symptom demanding treatment [13,14], mostly occurring below 12 nmol/L, this is a distinct signal for beginning testosterone therapy. Levels for younger men also serve as reference values for older men, and several recent large epidemiological studies from the USA [15], Australia [16] and Europe [17,18] show that 12 nmol/L is the lowest value for testosterone in the serum of healthy men.

While total testosterone remains the most important diagnostic parameter (assuming that it is determined with methods under strict quality control), free testosterone can be used as an additional parameter in borderline cases. However, many labs offer the “free androgen index” (FAI) as a pseudo-parameter for which total testosterone is simply divided by SHBG. This index correlates with the actual real free testosterone in women, but not in men and should therefore not be used. Free testosterone can easily be calculated from total testosterone and SHBG by using a generally available formula (http://issam.ch/freetesto.htm). Only free testosterone measured by this method can be helpful in male diagnosis [19,20]. Free testosterone values below 225 pmol/L (65 pg/ml) require testosterone treatment.

Mass spectrometric methods (LC-MS/MS) are the gold standard for measuring testosterone and provide the option of simultaneously determining a great spectrum of steroid hormones [21]. These methods continue to be reserved for large centres and have persevered only in connection with clinical studies relevant to licensing procedures. In addition, comparison of various methods for determining testosterone has shown that well-controlled immunoassays correlate very well with results from mass spectrometry, at least for diagnostic purposes [15,17,22]. The reliability of immunoassays becomes

critical when applied to women and prepubertal boys, but is adequate for routine diagnostics of hypogonadism and for monitoring substitution therapy. Nevertheless, the Journal of Clinical Endocrinology and Metabolism has decided that from the beginning of 2015 it will require use of mass spectrometry for testosterone measurements in clinical studies [23]. However, on February 12, 2014 the president of the US Endocrine Society, Teresa K. Woodruff, suspended this requirement "pending further scientific review".

## Testosterone preparations and modes of application

Testosterone was synthesized in 1935 and has now been in clinical use for almost 8 decades [24]. While today intramuscular, subdermal, transdermal, oral and buccal testosterone preparations are available (Table 1), until the 1990s only preparations resulting in unphysiologic serum levels existed. Aware of these shortcomings, WHO, NIH and FDA jointly formulated general principles of testosterone therapy as "Guidelines for the use of androgens in men" [25].

Only preparations of natural testosterone should be used for substitution therapy in hypogonadism since the full spectrum of testosterone action in the body can only be achieved if testosterone is aromatized to estradiol ***and*** 5alpha-reduced to dihydrotestosterone (DHT). As conversion to these metabolites occurs at physiologically well-balanced rates from natural testosterone as secreted by the testes, and not with synthetic androgens, testosterone remains the first choice for substitution purposes. The need for substituted testosterone to act not only directly but also after conversion to its metabolites was recently reconfirmed in an elegant study on body composition and sexual function following testosterone substitution with or without an aromatase inhibitor [26]. Similarly, administration of DHT instead of testosterone leads to loss of bone mass as no estradiol is formed [27].

The concept of using natural testosterone has outlived the phase of trying modified androgens e.g. anabolic steroids in the 1960s and 1970s and has not yet been challenged by the slow clinical development of Selective Androgen Receptor Modulators (SARMs). By definition, SARMs appear to be unsuited for the treatment of hypogonadism as they only target selected testosterone functions [28]. For example, the clinically most advanced SARM GTx-024 (Enobosarm®) has been shown to prevent sarcopenia, but as it lacks all other androgenic effects (including those on bones) it may find an indication for cachectic cancer patients [29], but not for hypogonadism where the full spectrum of testosterone action is required.

A second recommendation from WHO/NIH/FDA [25] demands that testosterone substitution should result in circulating serum levels as close to physiology as possible. Accordingly testosterone treatment of hypogonadal males should avoid supraphysiologically high testosterone serum levels as well as subnormally low levels. Until then mostly intramuscular testosterone enanthate or cypionate had been

**Table 1**
Testosterone preparations for substitution of hypogonadism.

| Route of application | Preparation | Trade name |
|---|---|---|
| Intramuscular | **Testosterone enanthate** | |
| | 250 mg/2–3 weeks | Testoviron® Depot 250<br>Testosterone Depot® |
| | **Testosterone undecanoate** | |
| | 1000 mg, first after 6, then every 12 weeks | Nebido® |
| | 750 mg, every 10 weeks | Aveed® |
| Transdermal | **Testosterone** | |
| | Two systems every 48 h | Testopatch® |
| | 125 mg in 5 g gel daily | Testotop® |
| | 100 mg in 5 g gel daily | Tostran® |
| | 2 × 50 mg in 5 g gel daily | Testogel®, Androgel®, Testim®, Androtop® |
| | 60 mg in 2% solution daily | Axiron® |
| Oral | **Testosterone undecanoate** | |
| | 3-4 capsules à 40 mg daily | Andriol® Testocaps |
| Buccal | **Testosterone** | |
| | 2 tablets daily | Striant® |

used, resulting in a rollercoaster pharmacokinetic pattern, and patients disliked the ups and downs of well-being, vigour, sexual activity and emotional stability [30]. Currently mostly transdermal gels and intramuscular testosterone undecanoate tend to be used [7,8] and these preparations, if properly administered, result in physiologic serum testosterone levels.

*Intramuscular testosterone application*

Since free testosterone, when injected, is degraded with a half-life of only 10 min, esterification of the molecule leads to more suitable forms of injectable preparations.

**Testosterone enanthate** has been the most common preparation for testosterone substitution since its introduction in the early 1950s. This substance has a terminal half-life of 4.5 days; maximum concentrations are reached after 10 h following a single injection of 250 mg [30]. Multiple-dose pharmacokinetics reveal an optimal injection interval of 2–3 weeks at a dose of 200–250 mg. **Testosterone cypionate** and **testosterone cyclohexanecarboxylate** resemble the pharmacokinetic properties of testosterone enanthate [30]. They do not provide an advantage over the enanthate ester.

**Testosterone propionate** has a terminal half-life of only 19 h. After a single injection of 50 mg, maximum concentration is reached after 14 h. It is obvious that this substance requires frequent injections. Multiple-dose pharmacokinetics reveal optimal intervals of 2–3 days, but fluctuations below normal-range values persist [30]. Therefore the substance is not suitable for long-term treatment of hypogonadism.

Intramuscular preparations widely used in the past (Sustanon® and Omnadren®) contain a mixture of testosterone esters assumed to act synergistically due to different kinetic profiles. However, they may produce even higher initial peaks and perhaps shorter duration of action. These ester mixtures do not appear to provide an advantage over single-ester preparations.

While already in use as an oral preparation (see below), an **injectable form of testosterone undecanoate** in teaseed oil with prolonged duration of action was described in China. Samples brought by the author to Germany in 1993 confirmed the long half-life (21 days) in volunteering hypogonadal men who showed serum levels in the normal range. The vehicle was changed from Chinese teaseed oil to castor oil and, thus Europeanized, the preparation was developed for clinical use. In castor oil, an even longer half-life of about 34 days was observed [30]. Peak values remain within the normal range in order to achieve a steady state at the beginning of substitution, the second 1000 mg injection is given 6 weeks after the first, further injections follow 10–14 weeks later. Individual intervals are determined according to serum testosterone levels which are measured immediately before the next injection [31]. These determinations are then repeated at yearly intervals. Values that are too high lead to extension of injection intervals, those that are too low to a shortening in injection intervals. Slow intragluteal injections are recommended [32,33].

Although the injectable preparations are generally considered very safe, pulmonary oil microembolism (POME), clinically characterized by brief respiratory symptoms including cough, urge to cough and dyspnoea immediately after the injection have been observed after injection of testosterone enanthate in castor oil at a rate of 1.5% of injections [34]. Similarly, POME may occur after intramuscular injection of testosterone undecanoate, although at a much lower rate: the manufacturer has reported 1.5 POME cases per 10,000 injections in the clinical database and 0.7 cases per 10,000 injections in the 9-year postmarketing surveillance. Few of these patients needed to be treated with epinephrine [35]. Nevertheless, FDA has required a special warning of POME when recently licensing intramuscular testosterone undecanoate (Aveed®) in the USA. This preparation contains only 750 mg testosterone undecanoate and therefore requires slightly shorter injection intervals of about 10 weeks [36].

*Subdermal testosterone implants*

**Testosterone pellet implants** were among the first modalities applied for testosterone replacement therapy, reaching back to the late 1930s. Modern pellets are produced by high-temperature moulding and are available in two sizes, containing 100 or 200 mg of crystalline steroid, with a length of 6 or 12 mm and a common diameter of 4.5 mm [30]. Implanted by a tunnelling technique

using a trocar, they remain under the skin of the lower abdominal wall and are totally biodegradable. If 3–6 implants are inserted, slowly declining serum testosterone levels in the normal range are achieved for 4–6 months. There is, however, an initial burst release, so that supraphysiological levels of about 50 nmol/L result. The overall terminal half-life was calculated at 71 days [37]. A review of 973 implantations in 221 men showed adverse local effects such as extrusion, bleeding and inflammations/infections in 11% [38]. In a cross-over study of pellets versus injectable testosterone undecanoate most patients preferred injections, mainly because of the simpler delivery [39].

*Oral testosterone*

When pure testosterone is applied orally, it is readily absorbed by the intestine, but very effectively eliminated by the first-pass effect of the liver. In order to overcome this, more than 1 g of testosterone would have to be administered in one dose. If, however, **testosterone undecanoate** is administered **orally**, due to the long aliphatic side chain the molecule is absorbed via the lymph and reaches circulation and target organs before the liver. Capsules of 40 mg are commercially available, and three to four such capsules have to be taken over one day for full substitution of hypogonadism [40]. Absorption is improved if the capsules are taken with meals [41]. Pharmacokinetic analysis shows high intra- and inter-individual variability in serum concentrations [30] and profiles are difficult to predict with precision. This preparation is best suited as a supplement to reduced but still present endogenous testosterone production since it does not fully suppress pituitary gonadotropin secretion and Leydig cell function. Long-term use is safe, as has been demonstrated in a 10-year observation study [42]. The same impact of an accompanying meal, especially containing fat, can be observed with a new self-emulsifying testosterone undecanoate preparation given in 300 mg capsules [43]. With one daily dose this preparation shows more stable testosterone kinetics.

For completeness it should be mentioned that synthetic 17$\alpha$-alkylated **methyltestosterone** and **fluoxymesterone** have become obsolete as they may develop liver toxicity on long-term use as required for substitution of hypogonadism, while oral testosterone undecanoate does not show this unwanted side-effect.

*Buccal testosterone*

Incorporating testosterone into polyethylene matrices with limited water-solubility represents an attempt to develop new forms for buccal application. The mucoadhesive tablets containing 30 mg of testosterone (Striant®) adhere to the gingiva above the incisors for many hours and slowly release testosterone into the circulation. Twice daily application results in even serums levels [44,45]. In 3–15% patients experience irritation, inflammation or gingivitis, but those who become accustomed to the tablets tolerate them well [45].

*Transdermal testosterone*

Transdermal testosterone preparations mimic physiological diurnal variations and their kinetic profile is closest to ideal substitution. They may be used as first choice and are especially well suited for patients with fluctuating symptoms caused by other preparations. In addition, upon removal, testosterone is immediately eliminated and they are therefore specifically suited for substitution in advanced age to treat quickly when side effects should occur [3].

Scrotal patches consisting of a thin film containing 15 mg native testosterone were the first transdermal preparation on the market. They were applied daily in the evening and lead to sufficient serum testosterone levels for 22–24 h. Adequate long-term substitution was achieved without serious side-effects in patients treated up to 10 years with these patches [46]. Later developments superseded this initially useful preparation.

Non-scrotal transdermal systems also result in physiological serum levels with an appropriate number of systems, which have to be applied in the evening [30]. As resorption of testosterone depends on the use of enhancers, in some cases considerable skin reactions limit their use. Although the patches mentioned above are hardly used today, a new testosterone patch was developed recently

causing little skin irritation and which must be changed only every other day; however, 2 systems with either 1.8 or 2.4 mg resorbed per day must be used [47].

A further transdermal application is the use of testosterone gels applied to large skin areas in order to allow sufficient amounts of the hormone to be resorbed. Physiological levels result when these gels, applied in the morning to the upper arm, shoulders and abdomen are left to dry for five to ten minutes. Thereafter the danger of interperson transfer is negligible, especially if the skin is washed after evaporation of the alcohol. Long-term use over several years showed good results [48–50].

If a preparation with a higher testosterone concentration i.e. 125 mg in 5 g gel is used, less gel needs to be applied. If this gel is applied to the scrotum only one fifth of the amount required when used on other skin areas is necessary for substitution due to higher scrotal skin permeability [51]. However, this preparation has only been licensed for non-scrotal skin application.

When using gels or solutions care must be taken to avoid unintended transfer by skin contact with children or women. Several such cases of androgenization of women, girls and boys have been reported [52,53]. The lowest risk is anticipated with those applications in which the residual testosterone gel is washed off shortly after application, as skin penetration follows within a few minutes of gel application [51]. The FDA considered the risk of interpersonal testosterone transfer so serious that in 2011 the manufacturers of testosterone gels were required to include a black box warning in the package slip.

## Sexual function

Loss of libido and erectile dysfunction are early signs of hypogonadism and return to normal under testosterone substitution [54]. In fact, they are the first symptoms to respond to substitution, i.e. as early as 2–4 weeks [55]. For objective evaluation to psychosexual effects weekly questionnaires on sexual thoughts and function, sexual intent and desire, satisfaction with sexuality, frequency of erection and number of morning erections and ejaculations may be used (e.g. [56,57]). While such questionnaires e.g. ADAM are inappropriate for diagnosing hypogonadism, they are well suited for monitoring substitution therapy.

The older the hypogonadal patients become, the more frequent arteriosclerosis may accompany testosterone deficiency and testosterone supplementation alone may not be sufficient to treat erectile dysfunction. In these cases combination of testosterone with PDE5 inhibitors has provided beneficial effects [58–60].

As antihypertensive medications are also prescribed with advancing age it should be remembered that these medications may lead to or aggravate erectile dysfunction. Betablockers may reduce testicular and penile perfusion, with the exception of nebivolol. Similarly diuretics, especially thiazides, may cause erectile dysfunction by a yet unknown mechanism [61].

## Bone density and fracture rates

Testosterone replacement therapy closes the epiphyses in hypogonadal adolescents who have not gone through puberty, and will increase low bone mineralization in hypogonadal adults, preventing and reversing osteoporosis [62,63]. The effects on bones take longest to become quantifiable [55]. As the testosterone effects on bones are mainly transmitted through estradiol [64,65] it is important that natural testosterone that can be converted to estradiol be used for substitution. In addition to conversion to oestrogens, the androgen receptor (AR) polymorphism is important for effects on bones. It has been shown that the length of CAG repeats in exon 1 of the AR correlates negatively with bone density in normal and hypogonadal men [66], as does the response to testosterone treatment [67].

The effects of testosterone substitution in men with Klinefelter syndrome are controversial. While mostly positive effects on bone density in Klinefelter patients are described (e.g. [68]), bone mineral density was found lower in adult Klinefelter patients than in aged-matched controls despite testosterone substitution [69]. It may well be that factors other than lack of testosterone cause osteoporosis in Klinefelter patients. Rather than due to testosterone deficiency, this may be a consequence of an additional *SHOX* gene, an X-chromosomal and Y-chromosomal gene that may escape X-chromosomal inactivation and may thus have a general negative impact on bone mineralization [70].

Osteologists have been reluctant to recognize hypogonadism as a cause of osteoporosis. Recently, however, guidelines for diagnosis and treatment of osteoporosis in men have introduced testosterone determination as part of the basic diagnosis of osteoporosis and recommend testosterone substitution when values are below 7 nmol/L [71]. However, at such low levels osteoporosis is already quite advanced and it seems advisable to initiate therapy at a borderline level of 12 nmol.

Although testosterone levels and AR polymorphism correlate with bone mass and fracture rates it has not been proven to date that testosterone substitution prevents fractures in hypogonadal men, especially in patients with late-onset hypogonadism.

## Obesity, metabolic syndrome, diabetes type 2 and cardiovascular diseases

Patients with hypogonadotropic hypogonadism show a fivefold higher incidence of the metabolic syndrome than eugonadal male controls [72]. Its incidence is even higher in men with Klinefelter syndrome [73]. Testosterone substitution leads to weight loss in hypogonadal men. Normalization of testosterone by substitution of testosterone undecanoate given i.m. over 5 years led to a mean weight loss of 15.3 kg in 255 hypogonadal patients, as well as to reduction in fasting glucose and $HbA1_c$ levels [74,75].

Several studies have demonstrated that low testosterone is a predictor for the metabolic syndrome (e.g. [76]) and progress has been made in the use of testosterone as complementary therapy for treatment of the metabolic syndrome and type II diabetes. Thus a placebo-controlled trial showed that men with metabolic syndrome and type II diabetes treated by diet and exercise experienced additional positive effects on $HbA1_c$ concentrations when testosterone was given simultaneously [77]. Survival rate of type II diabetic patients with testosterone levels below 10.4 nmol/L was improved when they received testosterone substitution in addition to conventional treatment compared to patients without testosterone substitution [78]. At the same time, substitution with testosterone had a significant positive influence on existing erectile dysfunction often occurring in type II diabetic patients [79].

Today vascular-induced erectile dysfunction is considered a risk marker for coronary heart disease and infarction [80]. In turn testosterone substitution may have a preventive effect on progression of arteriosclerosis and may even improve coronary blood flow and cardiac function [81]. While testosterone (and other anabolic androgenic steroids) consumed at extremely high doses often seen as in body builders may lead to coronary sclerosis (overview in [82]), at physiological levels it may have a cardioprotective effect [81]. An increase of circulatory endothelial progenitor cells under testosterone therapy may play a role [83].

In a metaanalysis of four studies Toma et al. [84] concluded that testosterone appears to be a promising therapy to improve functional capacity in patients with heart failure. However, other studies have challenged the positive assessment of testosterone in this condition. A trial of testosterone in hypogonadal elderly men with limited mobility was prematurely terminated because of a slightly higher incidence of cardiovascular adverse events in the verum group (106 vs. 105 patients) [85]. However, these patients were seriously disabled, the cardiovascular events were very heterogeneous and the patients were started on rather high testosterone doses causing increased estradiol levels and thereby predisposing for thromboembolism. In a metaanalysis of 27 trials covering 2994 mainly older men Xu et al. [86] found a marginally increased risk for myocardial events in men receiving testosterone compared to those in the placebo groups (OR 1.54, 95% CI 1.09–2.18). This effect was not noticed in those trials sponsored by a pharma company. The authors concluded that this slightly higher risk must be weighed against the benefits of testosterone therapy.

A recent study by Vigen et al. [87] claiming a higher incidence of myocardial infarctions in patients on testosterone than those without testosterone appears to be flawed by statistical calculations, improper patient allocation and follow-up and by including 100 women in a cohort of supposedly 1100 men! Finally, the paper by Finkle et al. [88] also found a higher risk for myocardial infarction in men who had been prescribed testosterone, especially those with a history of heart disease. This paper was based exclusively on health care data and the patients had never been seen by the authors. Basal testosterone, clinical details and indications for therapy were not known for either cohort nor was it known whether the medication has been consumed at all and for how long. The reference group consisted of men who had been prescribed phosphodiesterase-5-inhibitors. As the study and the control group were different, the comparison appears not warranted.

Despite these flaws the papers cause irritation among physicians prescribing and patients receiving testosterone and need to be carefully interpreted. They demonstrate the need for large-scale prospective trials, as long requested by experts and now finally by organizations such as the US Endocrine Society (February 11, 2014).

## Prostate cancer

The 2008 guidelines on late-onset hypogonadism (LOH) [3] state "*There is no conclusive evidence that testosterone therapy increases the risk of prostate cancer or BPH. There is also no evidence that testosterone treatment will convert subclinical prostate cancer to clinically detectable prostate cancer. However, there is unequivocal evidence that testosterone can stimulate growth and aggravate symptoms in men with locally advanced and metastatic prostate cancer*".

In the meantime no data have been produced opposing this statement. On the contrary, prostate carcinoma seems to occur in testosterone-treated men as often as in untreated men. In a 4-year observation period of men aged over 40 years with initial testosterone levels below 8.7 nmol/L, the incidence of prostate cancer was 1.6% in 398 testosterone-treated cases, whereas the 644 untreated elderly hypogonadal men showed an incidence of 2.0% [89]. If androgens should be involved in the pathogenesis of prostate carcinoma, then it is important to note that intraprostatic testosterone and DHT concentrations are independent of circulating testosterone and DHT levels and circulating levels are therefore unlikely to be directly involved in prostate disease [90,91].

As androgen deprivation therapy in prostate cancer patients can lead to all symptoms of hypogonadism including metabolic syndrome, coronary heart disease, osteoporosis and bone fractures, not to mention decreased quality of life [92], can such long-term testosterone therapy depriviation be justified in view of the side effects? According to current opinion, testosterone substitution may be justified if, after at least one year of successful prostate cancer treatment, no signs of relapse are present, the Gleason score before surgery was below 8 and PSA below 10 ng/ml [5,93]. However, both extreme caution and constant monitoring are mandatory as large-scale definitive studies are lacking.

## Timing of testosterone treatment

As a general rule hormone substitution is initiated when levels fall below the lower limit of normal. In the case of all forms of hypogonadism this means when testosterone levels fall below 12 nmol/L. However, there are situations where this rule may require consideration and modification.

### *Klinefelter syndrome*

One of these situations arises in patients with Klinefelter syndrome (KS). Early observations indicated that KS boys were better adjusted and had improved cognitive and learning abilities when they were substituted from early puberty onwards [94]. However, only recently has this question been taken up again in clinical trials (e.g. [95]). It is also difficult for the endocrinologist to convince psychiatrists and social workers of the usefulness of this hormone in adjusting the potentially criminal adolescent to his environment as long as testosterone is considered an agent causing aggression and overlooking its real function in socialisation. Proper trials are required.

As there may already be a testosterone deficiency in KS boys before puberty, some investigations even propagate testosterone substitution during infancy. In a retrospective study comparing 34 47,XXY-boys who had received short courses of testosterone treatment at ages 3 and 6 years with 67 non-treated KS patients, a significant advantage in cognitive, language, intellectual and neuromotor functions was observed in treated boys [96]. These encouraging findings open a new dimension for testosterone substitution in KS patients.

KS patients were usually considered infertile as 90% are azoospermic and the remaining men have only very few sperm in their semen. Today, thanks to modern techniques of sperm retrieval and assisted reproduction in a substantial proportion of men with KS sperm can be found in isolated testicular tubules, especially when microscopic biopsy techniques are used. By direct intracytoplasmic injection (ICSI) of the isolated sperm into oocytes pregnancies may result [97]. If the patient

undergoing TESE has already received testosterone substitution, this may have suppressed his residual spermatogenesis; therefore cessation of testosterone treatment before TESE attempts - depending on the half-life of the testosterone preparations used - is advisable. Some groups have reported positive outcomes following treatment with hCG, anti-oestrogens and/or aromatase inhibitors before TESE [98]. However, these trials were not controlled and it remains questionable whether such pre-treatment really has an effect, as controlled, double-blind trials of these drugs in other types of male infertility have shown no positive effects. Currently TESE and cryopreservation of biopsy material is being considered in pubertal boys before any testosterone treatment [99].

*Secondary hypogonadism*

Similarly, in patients with secondary hypogonadism due to e.g. post-hypophysectomy, pituitary tumours or isolated hypogonadotropic hypogonadism, the question arises whether testosterone substitution before induction of spermatogenesis by gonadotropins or pulsatile GnRH may impair the chances for successful development of spermatogenesis. So far, no clear evidence has been produced in favour of this hypothesis, but well-designed comparative trials are lacking. Nevertheless, once spermatogenesis has been successfully induced and then the patient starts or resumes testosterone substitution, the process of inducing spermatogenesis for a second time at a later stage is faster than the first round of stimulatory therapy [100].

## Monitoring of testosterone substitution

Some recent publications indicate that testosterone is frequently prescribed without proper patient follow-up [7,8,87,88]. However, although careful monitoring at regular intervals is emphasized in most of the current guidelines [6], they are not consistent concerning follow-up intervals and the parameters to be evaluated. It is advisable to see the patient following initiation of treatment after 3, 6 and 12 months and then annually.

On these occasions the patient should be interviewed for mental and physical activity, libido and sexual activity. Questionnaires may be useful, especially at the beginning of treatment. Body weight, waist circumference and fat distribution, beard growth and hair pattern as well as possible development of breast tissue and size and surface of the prostate (by digital rectal examination) should be monitored.

Taking into consideration the pharmacokinetics of the respective testosterone preparation [30], serum testosterone should be measured at the scheduled visits, but always before the next dose is applied and the levels should then be in the lower normal range. If found too high or too low, the doses or dosing intervals need to be adjusted.

The prostate of the hypogonadal patient will grow to its normal size under testosterone substitution. PSA will also increase, but should not exceed the normal range. If it does, a urological consultation is required.

Haemoglobin, erythrocytes and haematocrit belong to routine surveillance. Too high testosterone levels or action may lead to polycythaemia [101]. The older the patient, the higher his BMI and the more active his androgen receptor is, the more likely polycythaemia may develop [32]. In such events testosterone substitution must be reduced or temporarily suspended to prevent embolic or thrombotic events. Phlebotomy may be indicated on an emergency basis.

Bone density should be measured prior to commencement of testosterone substitution and then at regular intervals.

Any eventual comorbidities (see above sections) require careful diagnosis and additional treatment.

## Conclusion

As described above, testosterone substitution is a powerful tool for the treatment of hypogonadism, and different testosterone preparations are available to achieve effective substitution. In cases of congenital or acquired irreversible forms of hypogonadism such as isolated hypogonadotropic hypogonadism, post-hypophysectomy, Klinefelter syndrome and anorchia there are no alternatives to testosterone for long-term substitution. However, hypogonadism may not only favour coinciding diseases such

as obesity, metabolic syndrome, diabetes type II, and cardiovascular disease, but these comorbidities may in turn also cause aquired hypogonadism [102] and their prevention may protect against hypogonadism.

Accordingly an analysis of 9 studies in obese men showed a clear connection between weight reduction and increased total testosterone; weight loss of between 10 and 20% led to an increase in testosterone of 3–6 nmol/L [103]. The European Male Aging Study (EMAS) confirmed a significant connection between altered weight and total and free testosterone in 2395 men followed for 4.4 years; here weight loss led to increased testosterone while weight gain correlated with lowered testosterone [104]. Furthermore Yeap et al. [105] demonstrated that lifestyle of 3453 men over 65, followed for a median of 5.7 years, clearly influenced testosterone values, while lifestyle, generally considered as healthier and correspondingly scored, was accompanied by higher testosterone values. These results were confirmed in a further Australian study [106]. This means that preventive measures are as important as treatment of hypogonadism and that patients with congenital or acquired irreversible forms of hypogonadism should be encouraged to adopt a favourable life-style in addition to carefully monitored testosterone substitution.

**Practice points**

- Testosterone is the most important male hormone and not a life-style drug. Its prescription requires the proper diagnosis of hypogonadism as well as regular monitoring of therapeutic effectiveness.
- Substitution therapy should use natural testosterone and aim at physiologic serum levels in order to avoid adverse side effects.
- Testosterone does not cause prostate carcinoma, but may support its growth. Therefore a prostate carcinoma must be excluded before substitution is initiated.
- Overdosing should be avoided as it may cause polycythaemia possibly leading to thromboembolism, especially in obese and ageing hypogonadal patients.
- If erectile dysfunction in hypogonadal patients does not respond to testosterone substitution, combination with phosphodiesterase-5-inhibitors may be considered.

**Research agenda**

- The significance of the physiologic diurnal rhythm of serum testosterone needs to be investigated. Is it an epiphenomenon or does it have implications for the quality of life of eugonadal as well as testosterone-substituted hypogonadal men?
- Optimisation of testosterone treatment in men with Klinefelter syndrome: When to start (before, during, after puberty)? Which preparation? Which doses? Long-term follow-up.
- Determining whether testosterone substitution not only increases bone mineral density, but also decreases fracture rates in hypogonadal men.
- Large-scale controlled clinical trials are mandatory to determine the beneficial and adverse effects of testosterone on cardiovascular function and on the prostate.
- Creating Leydig cells from stem cells for transplantation to hypogonadal men in order to outdate exogenous pharmacological testosterone treatment.

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