



# The role of androgen therapy in prostate cancer: from testosterone replacement therapy to bipolar androgen therapy

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• Testosterone replacement therapy (TRT) is the primary treatment for male testosterone deficiency. This therapy raises concerns over the risk of prostate cancer (PC), because testosterone has historically been considered the fuel for PC. We discuss the re-evaluation of the relationship between androgen and PC, and highlight the safety of TRT in the treatment of symptomatic men with testosterone deficiency who have low-risk disease after treatment for localized PC with surgery or radiation. Furthermore, we review the clinical application and potential mechanisms of bipolar androgen therapy (BAT) in the treatment of castration-resistant PC, emphasizing that much remains to be done before BAT can be broadly applied.

## Introduction

Prostate cancer (PC) is a malignant condition with the highest incidence and second highest mortality rate among cancers affecting males in Europe and the United States [1]. The incidence of PC has stabilized in recent years, and the mortality rate is declining in most countries around the world [2,3]. Two decades of steep (4% per year on average) declines for PC have been attributed to earlier diagnosis through prostate specific antigen (PSA) testing, as well as advances in treatments. By 2017, the PC death rate had dropped by 52% from its peak in 1993 [1]. But now that patients are living longer, with low levels of testosterone after PC treatment, the incidence of hypogonadism in men is increasing, and there are significant ethical and medical problems associated with treating this condition with testosterone replacement therapy (TRT).

Historically, it was thought that testosterone deficiency (TD) was mostly due to congenital hypogonadism, or gonadal failure caused by pituitary tumors or gonadal injuries [4]. However, it has been confirmed that many other clinical diseases, including pathological obesity, serious illness and psychological stress, can also cause significant TD [5,6]. Male TD can cause a series of worrisome health problems [7], and TRT is currently the first choice for treatment. TRT has been shown to improve or even reverse these symptoms [8,9].

Persistent environmental stimuli cause chronic prostatic inflammation. Inflammatory cytokines such as interferon- $\gamma$  (IFN- $\gamma$ ), tumor necrosis factor- $\alpha$  (TNF- $\alpha$ ) and transforming growth factor- $\beta$  (TGF- $\beta$ ) infiltrate the prostate, resulting in the injury and repair of prostate tissue. This leads to proliferative inflammatory atrophy followed by somatic genomic alterations in a small subset of cells, including glutathione S-transferase P1 (GSTP1) methylation [10], overexpression of MYC and the loss of NK3 homeobox 1 (NKX3.1) [11], thereby promoting prostate intraepithelial neoplasia. Androgen receptor (AR) mutations [12] and TMPRSS2-ERG fusions [13] will drive PC progression.

Androgen deprivation therapy (ADT) is currently the first-line treatment for advanced PC. However, numerous studies have shown that patients with ADT inevitably develop castration-resistant PC (CRPC) [14,15]. A variety of chemotherapy drugs have been approved for the treatment of CRPC. Docetaxel and carbamazepine prevent mitosis by inhibiting tubulin depolymerization, resulting in cell death [16]. Morgans *et al.* found that ADT combined with docetaxel significantly improved median overall survival compared with ADT alone, but side effects of chemotherapy, such as hematological toxicity and neutropenia, often occurred [17]. The US Food and Drug Administration (FDA) has approved the use of abiraterone acetate in patients with metastatic CRPC (mCRPC) before and after chemotherapy [18]. In addition, molecular targeting therapies and immunotherapies, such as olaparib and bevacizumab, have developed rapidly as a new direction of

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CRPC therapy, but at present, the treatment is expensive and can only be applied to a limited patient population, so further studies are needed.

14 CRPC cell lines express 30–90 times higher levels of AR than do normal prostate cells [19], so it seems paradoxical to administer androgen (male sex hormones, including testosterone) to CRPC patients. However, exposure to supraphysiologic levels of testosterone (SPT), followed by rapid lowering of testosterone levels, can provide a therapeutic opportunity. Bipolar androgen therapy (BAT) is a novel strategy for the treatment of metastatic PC in which CRPC cells are rapidly exposed to supraphysiologic androgen levels, and then castration therapy is used to reduce androgen levels in the body. Multiple rapid hyperandrogen–hypoandrogen cycles can restore tumor sensitivity to anti-androgen therapy and inhibit cancer cell growth [20]. Androgen, as an endogenous substance, seems to be safer than other complex and expensive treatments that carry the risk of severe adverse reactions and intolerance.

### The history of androgen therapy: discovery, development and disparity

The relationship between androgen therapy and PC is like a tug-of-war. In 1966, Charles Brenton Huggins won the Nobel Prize in Physiology or Medicine for his groundbreaking research into the significance of androgen therapy in patients with metastatic PC. This discovery led to the belief that the malignant growth of PC depends partly on androgen, specifically testosterone, which promotes the growth and invasiveness of PC cells, and the ‘androgen hypothesis’ was generated. The theoretical basis for this originated in a small study published in 1941, in which, through the use of blood biomarkers, researchers claimed that ADT inhibited tumor growth in patients with metastatic PC. Conversely, the use of exogenous testosterone promoted the progression of tumors [21]. However, it is noteworthy that the assertions in the latter part of the conclusions remain narrow in focus, dealing only with acid phosphatase detection in individuals. Studies on the androgen hypothesis only provided evidence regarding the role of testosterone in the progression of PC, whereas the role of testosterone in the occurrence of PC remained unclear.

In the 1980s, scholars described the relationship between testosterone and PC as ‘fuel on fire’ and ‘energy for tumors’ [22], and historically, PC has been viewed as an absolute contraindication for TRT. But interestingly, based on the androgen hypothesis, a large number of findings have been unexpected: for example, there is no association among endogenous testosterone levels, PSA, prostate volume and PC [23–26], and an analysis of a population study found that PC was not related to the entire range of naturally produced testosterone levels [24]. What’s more, several lines of evidence suggested that lower serum total testosterone and lower free testosterone levels were associated with more aggressive PC and poorer prognosis [27–30]. Overall, the results seem to suggest that the relationship between testosterone and PC is not a simple, linear one, and there is a certain threshold in the middle that is associated with the occurrence and development of cancer.

### A paradoxical viewpoint: the androgen saturation model

Surveys such as that conducted by Traish *et al.* have shown that the maximum level of testosterone binding to AR (saturation) occurs

at a relatively low androgen concentration [31]. AR in human prostate tissue is saturated when it approaches 4 nmol/l (120 ng/dl) *in vitro*, which is equivalent to about 8–8.3 nmol/l (240–250 ng/dl) *in vivo* [32]. Traish’s work on testosterone is complemented by Morgentaler’s study of androgen saturation. It states that PC is sensitive to low testosterone levels, and that excessive testosterone cannot enter the nucleus to stimulate cell growth after ARs have become fully occupied, therefore there is a limited range of testosterone levels that have an effect on PC [33].

The androgen saturation model explains the contradictory observation that prostate tissue is sensitive to changes in testosterone levels at low concentrations, but becomes insensitive to changes in testosterone levels at high concentrations [33,34]. Consistent with the saturation model, two studies investigated the effect of SPT on the prostate of young healthy men after 15 weeks [35] and 5 months [36]. Despite significant elevations in serum total and free testosterone, healthy young men did not demonstrate increased serum or semen PSA levels, or increased prostate volume. In addition, the non-linear ‘U model’ proposed by Salonia *et al.* suggests that men with the highest and the lowest testosterone levels have an increased risk of PC [37]. Therefore, the relationship between testosterone and PC has not been clearly defined, and the research raises questions over whether TRT, the current treatment of choice for men with TD, can be safely applied, particularly for men with a history of PC.

### The association between TRT and prostate cancer: risk or benefit?

*TRT does not increase the risk of prostate cancer*

Although TRT can ameliorate the symptoms of TD, there have been concerns over whether TRT can increase the risk of PC. However, past research strongly indicates that the risk is minimal. A British study found that patients who received TRT had no increased overall risk of PC compared with those who did not receive TRT [38]. Similarly, Zhang *et al.* found that TRT reduced the risk of PC with an average follow-up of 6.9 years [39], and in 2017, Loeb *et al.* found no correlation between TRT and the overall risk of PC [40].

Furthermore, two studies investigated the relationship between TRT and the rate of positive prostate biopsies. The first showed that out of 999 men clinically diagnosed with hypogonadism, 750 (75%) started TRT, and a total of 55 underwent biopsies for suspected PC. The percentage of positive prostate biopsies (37.5%) among men who received TRT was similar to the percentage among men who did not receive the therapy [41]. The second study revealed that the incidence of positive prostate biopsies was lowest in men with TD who were receiving TRT [42]. All of the studies support the view that TRT does not increase the risk of PC, but instead exhibits protective effects. The reduced aggressiveness of PC in patients undergoing TRT is a new discovery, which emphasizes the importance of monitoring testosterone levels.

Saad *et al.* were more concerned about the long-term effects of TRT. They published a paper in 2020 revealing that TRT can reduce the risk of PC, improve erectile dysfunction and reduce the occurrence of adverse cardiovascular events, with an observation period of up to 12 years [43]. The study offers probably the most comprehensive empirical analysis of the long-term safety of TRT. Nevertheless, different views exist in the literature indicating that TRT

might increase the risk of PC [44,45]; however, this evidence is not sufficient, and the authors also explain in the discussion that the findings could be the result of selection bias or uncertain biological effects.

According to the European Association of Urology (EAU) guidelines for the diagnosis and treatment of male hypogonadism, the contraindications for TRT are: (i) advanced or metastatic PC; (ii) male breast cancer; (iii) men with reproductive needs; (iv) hematocrit > 0.54; and (v) severe chronic heart failure [46]. According to the latest research, TRT seems to be safe and effective for the treatment of TD patients, with the exclusion of contraindications (Table 1).

*TRT does not promote recurrence or progression in early-stage prostate cancer*

A large and growing body of evidence suggests that TRT benefits men with TD and does not increase the risk of PC (Table 1). However, it is not conclusive about whether the application of TRT is safe and effective in patients with PC. In fact, in 2007, about 70% of healthcare workers were concerned that TRT could promote the recurrence and progression of PC [47]. A 2019 survey showed that British urological oncologists generally do not use testosterone in their patient management [48], and people are generally cautious about the possible benefits of TRT. However, this uncertainty has gradually decayed as fresh research has provided a deeper understanding of the androgen saturation model. This shift has spurred clinicians to explore the impact of TRT on patients with early-stage PC.

Early examples of research into TRT include a 2004 study by Kaufman *et al.*, which followed seven PC patients who underwent various TRT treatments for 2–13 years after radical prostatectomy (RP). The authors found no evidence of biochemical or clinical recurrence in any patients [49], and this finding was consistent with two later studies in 2005 [50] and 2009 [51].

Similarly, in 2009, Morales *et al.* evaluated the efficacy of TRT in PC with TD after external beam radiotherapy (EBRT). Only one patient showed a transient elevation of PSA levels, which did not exceed 1.5 ng/mL [52]. A retrospective study analyzed data on PC patients who underwent EBRT or brachytherapy (BT) and found

that the patients displayed elevated serum testosterone levels, improved symptoms of TD and no signs of PC recurrence or progression over a follow-up period of nearly 30 months [53]. In 2014, a study of 20 patients who received BT for PC and received long-acting testosterone injections reached the same conclusion that TRT does not accelerate the recurrence and progression of PC [54].

Over the past decade, the implementation of active surveillance (AS) in PC has gained increasing prominence. Researchers attempted to evaluate the impact of TRT on PC patients using AS. In 2016, a survey conducted by Kacker *et al.* revealed that three men (3/28, 10.7%) who received TRT and nine men (9/43, 9.38%) who did not showed an escalation of Gleason score [55]. Hashimoto *et al.* retrospectively reviewed 12 low-risk PC patients who received TRT and AS, and found that none of them demonstrated progression, as evaluated by biopsies [56]. As expected, similar progression was found in groups who did and did not receive TRT, and escalation of Gleason score was not associated with TRT.

*High levels of androgens due to TRT might be protective against PC recurrence*

Interestingly, more recent studies have shown that the overall biochemical recurrence (BCR) rate in patients receiving TRT is lower than that reported in the literature after initial treatment. In other words, TRT promotes survival for PC patients [57–59]. For instance, Kaplan *et al.* argued that TRT did not increase overall patient mortality and the probability of using salvage ADT [57]. Pastuszak *et al.* asserted that in PC patients who underwent RP and were treated with TRT, the rate of BCR was significantly lower than in the control group during the 36-month follow-up [58]. Notably, all of the people with BCR in the treatment group were high-risk patients (Gleason score ≥ 8 or surgical margin positive or lymph node positive) [58]. This result supports the speculation that the relapses seen in the high-risk PC patients were caused by a biological process of the disease itself, and were not related to TRT. Likewise, Ahlering *et al.* reported that TRT not only significantly reduces the risk of BCR of PC, but also significantly delays the recurrence time by up to 1.5 years [59].

**TABLE 1**  
**Studies investigating the safety of TRT in non-PC men with TD**

Authors	No. of patients	Median/mean follow-up (years)	Outcomes	Study type	Refs
Santella <i>et al.</i> (2019)	12,779	4.5	Patients who received TRT had no increased overall risk of PC compared with those who did not receive TRT	Retrospective research	[38]
Zhang <i>et al.</i> (2020)	776	6.9	TRT maintained normal serum testosterone levels and reduced the risk of PC	Retrospective research	[39]
Loeb <i>et al.</i> (2017)	1662	3	No association was found between TRT and overall prostate cancer risk, and patients who received TRT had a lower risk of aggressive PC	Retrospective research	[40]
Yassin <i>et al.</i> (2017)	553	6	The incidence of positive prostate biopsies was lowest in hypogonadal men receiving TRT, with significantly lower severity of PC in terms of staging and grading in the same group	Prospective cohort studies	[42]
Debruyne <i>et al.</i> (2017)	999	3	The proportion of positive biopsies was nearly identical in men on TRT (37.5%) compared to those not on TRT (37.0%) over the course of the study	Prospective cohort studies	[41]
Saad <i>et al.</i> (2020)	805	12	Long-term TRT for up to 12 years reduced the risk of PC	Prospective cohort studies	[43]

TABLE 2

# Studies investigating the safety of TRT in PC men with TD

Authors	No. of patients	Median follow-up (months)	Treatment regimen	Outcomes	Refs
Kaufman <i>et al.</i> (2004)	7	–	RP	No biochemical or clinical evidence of PC recurrence	[49]
Agarwal <i>et al.</i> (2005)	10	19	RP	No patient had detectable (greater than 0.1 ng/mL) PSA	[50]
Khera <i>et al.</i> (2009)	57	13	RP	No patient had a biochemical PSA recurrence	[51]
Pastuszak <i>et al.</i> (2013)	103	27.5	RP	Four and eight cases of cancer recurrence were observed in treatment and reference groups	[53]
Ahlering <i>et al.</i> (2020)	152	42	RP	7.2% and 12.6% experienced BCR in the TRT versus control groups; patients on TRT were approximately 54% less likely to show recurrence, and time to recurrence was delayed by an average of 1.5 years	[59]
Morales <i>et al.</i> (2009)	5	14.5	EBRT	One patient had a transitory increase in PSA level but none had levels of > 1.5 ng/mL	[52]
Balbontin <i>et al.</i> (2014)	20	31	BT	No cases of rising serum PSA, PC progression or recurrence	[54]
Pastuszak <i>et al.</i> (2013)	13	29.7	EBRT/BT	Without evidence of PC recurrence or progression	[58]
Kacker <i>et al.</i> (2016)	28	38.9	AS	Biopsy progression in men on AS seems to be unaffected by TRT over 3 years	[55]
Hashimoto <i>et al.</i> (2016)	12	42.5	AS	No patients demonstrated biopsy progression after TRT	[56]

The evidence presented in Table 2 suggests that receiving TRT to raise androgen levels might have a protective effect on PC recurrence, which provides a new strategy for the treatment of PC patients. These relationships might reflect a possible biological mechanism in which testosterone is important for the differentiation and function of normal prostate epithelial cells. Serum testosterone levels decrease with age, whereas the risk of PC increases [60]. Speculatively, high or normal levels of testosterone might keep prostate cells and early PC cells in a state of differentiation. Conversely, the gradual decrease of testosterone levels caused by ageing might lead to a less differentiated cancer phenotype. Previous studies have shown that men with PC and hypogonadism have a higher risk of aggressive disease [61]. These observations support biologically credible findings of a decreased risk of poorly differentiated PC in men who receive TRT.

## Bipolar androgen therapy: chance and challenge

### Supraphysiologic androgen therapy shows hope for treatment of CRPC

Data have emerged on the use of SPT as a protective agent against PC progression and a therapeutic agent for men with CRPC or with BCR. Feltquate *et al.* administered rapid androgen cycling therapy to 36 patients with advanced PC in 2006, and found that the clinical progress of PC and PSA decreased continuously [62]. A case report by Mathew documented the use of TRT in a patient with CRPC in 2008 and recorded a decrease in PSA that lasted for about one year [63]. Two Phase I clinical studies reported the results of CRPC treatment with testosterone gel in 2009. In the first phase of study, researchers evaluated 15 patients with early CRPC and enabled their testosterone level to reach around 300 ng/dl from castration level by TRT. In this study, one patient was observed to have symptomatic progression, and three (20%) patients demonstrated a decrease in PSA [64]. In the second phase of study, Morris and co-workers evaluated the treatment outcome of 12 patients with CRPC, and found that 30% of the patients showed a decrease in PSA [65]. The average level of serum testosterone level did not reach a supraphysiologic level.

Studies *in vivo* and *in vitro* have confirmed that the growth of AR-positive human CRPC cells can be inhibited by SPT [66–68]. As early as 2005, it was reported that exogenous androgens inhibited the growth of androgen-dependent LNCaP prostate tumor cells both *in vitro* and *in vivo* [66]. In 2014, Song *et al.* noted that low androgen was crucial for the initial growth of PC cells and that increased androgen concentrations promoted PC-cell proliferation. However, a high concentration of androgen led to a dose-dependent inhibition on PC-cell proliferation [67]. Subsequently, they confirmed their findings by testing the incidence and growth of PC xenografts in nude mice in 2017 [68].

Collectively, these studies indicate that the effect of androgens on PC-cell proliferation is biphasic. The creation of the BAT concept was partly influenced by TRT, and here we distinguish between BAT and TRT (Table 3).

### Potential mechanisms of BAT in inhibiting the growth of prostate cancer

The mechanism of BAT treatment in mCRPC is not well understood. One view is that BAT might inhibit DNA re-licensing in cells with high levels of AR and high doses of androgen. AR acts as the transcriptional initiation factor of DNA in PC cells, and activated AR binds to the protein origin of replication sites (ORS), which first binds to the highly conserved origin recognition complex (ORC), consisting of the subunits ORC1–6, in the early stage of G1 [69]. This is followed by the binding of cell division cycle 6 (CDC6) to ORC, which is required to load minichromosome maintenance complex component 2 (MCM2)–7 and chromatin licensing and DNA replication factor 1 (CDT1) onto the DNA, completing the formation of the pre-replication complexes required for the G1-dependent DNA license and DNA replication [70].

DNA replication relies on the DNA licensing process to ensure that the genome is replicated only once per cell cycle. The licensing factors are degraded in M phase or early G1 phase, and prevent replication by activating cell cycle-dependent kinase (CDK)-induced inactivation, nuclear output and proteolysis of ORC1, CDT1 and CDC6 in S-phase, so as to re-license and restart DNA replica-



TABLE 3  
A simple comparison between TRT and BAT

	TRT	BAT
Indications	Male hypogonadism	Advanced PC (CRPC/low metastatic burden recurrent disease)
Contraindications	Advanced or metastatic prostate cancer; male breast cancer; men with reproductive needs; hematocrit > 0.54; and severe chronic heart failure	Uncertain
Application method	Testosterone undecanoate taken orally at an initial dose of 120–160 mg per day for 2–3 weeks, followed by a maintenance dose of 40–120 mg per day; or use 5 mg testosterone gel or 1% skin patch or 0.25 g testosterone injection intramuscularly	Alternating 3-month cycles of BAT (400 mg intramuscularly on days 1, 29 or 57), followed by 3 months of ADT alone
Outcomes	The symptoms of testosterone deficiency (erectile dysfunction, decreased bone mineral density, cognitive impairment, insulin resistance) were significantly improved, and there was no significant increase in the risk of PC	PSA decreased continuously; assessable imaging regression
Side effects	Known results of hormone therapy: hot flashes, fatigue and female breast development. Side effects also include insufficient sperm production, fluid retention, deep vein thrombosis and pulmonary embolism	Adverse reactions were mostly mild (≤ grade 2), such as nausea, hot flashes, fatigue, hair loss, edema, neutropenia and unspecified cardiovascular events, such as myocardial infarction and pulmonary embolism

tion in the next cell cycle [71]. However, under the condition of SPT, ligand-dependent stabilization of AR during mitosis might inhibit the degradation of AR in M phase. This causes a small portion of AR to bind to ORS, preventing re-licensing for DNA during G1 and leading to S-phase arrest [72,73].

The development of DNA double-strand breaks (DSBs) that lead to genomic rearrangements is a frequent hallmark of cancers. Rapid shifts between castration testosterone levels and SPT produce androgenic signals that promote AR and co-recruit topoisomerase IIβ (TOP2β) to positions of TMPRSS2–ERG genomic breakpoints (TEGBs). This triggers recombinant TOP2β-mediated DSBs, which leads to gene rearrangement and the induction of apoptosis [74]. Gene rearrangement could be another mechanism by which BAT inhibits CRPC proliferation [75,76].

Furthermore, AR and AR splice variants are drivers of CRPC progression, and inhibition of their expression could result in decreased cell growth. Lysine-specific demethylase 1 (LSD1) has been shown by Metzger *et al.* to interact with AR by recruiting the AR binding site (ARBS) region, which has a highly conserved segment among species [77]. Balk *et al.* had confirmed that LSD1 acts as a repressor through demethylation of H3K4me1 and H3K4me2, thereby reducing expression of the AR gene in CRPC cells [78]. The mechanism of BAT-induced inhibition of PC cell growth and death could be multimodal, involving cell cycle arrest, apoptosis, disruption of DNA licensing and double-stranded DNA damage (Fig. 1).

The therapeutic effect of BAT in clinical trials

The observation that BAT inhibits PC growth has been incorporated into treatment regimens for advanced PC in recently published studies (Table 4). In the first pilot study of BAT conducted by Schweizer in 2015, 16 asymptomatic mCRPC patients completed at least 3 months of BAT. Initially, they received testosterone cypionate 400 mg intramuscularly on day 1 and etoposide 100 mg on days 1–14 of each cycle. Cycles were repeated every 28 days. This study showed that 50% (7/14) of patients experienced PSA decline, with 28.6% (4/14) showing a PSA decline

greater than 50%. Of the 10 patients who had evaluable soft tissue metastases, 50% had assessable imaging regression [79].

Another trial evaluated the effect of BAT in two groups of patients with asymptomatic metastatic PC or biochemically recurrent disease, who achieved PSA < 4 ng/dl after 6 months of ADT. Of the 29 hormone-sensitive PC (HSPC) patients, 59% (17/29) had a PSA of < 4 ng/mL 18 months after treatment, and 80% (8/10) of patients had an objectively complete (four patients) and partial response (four patients) [80].

Recently, a single-center and two-phase multi-cohort study evaluated the efficacy of BAT in 30 mCRPC patients with progressed tumors after enzalutamide treatment. The two main purposes of this study were to observe the PSA response rate of BAT in patients with mCRPC and to observe whether the patients regained sensitivity to enzalutamide. No treatment-related deaths were reported during either the BAT or enzalutamide regimens, and 30% of patients achieved a PSA response. Among the patients who progressed after BAT, 52% regained a PSA response to enzalutamide treatment [81]. The authors concluded that BAT is a safe and effective treatment for patients with mCRPC, and half of the patients were re-sensitized to enzalutamide.

Enzalutamide is a new AR antagonist that binds directly to AR and inhibits AR binding, AR nuclear translocation and AR-mediated DNA binding [82]. BAT aims to restore tumor sensitivity to anti-androgen therapy and inhibit cancer cell growth through a rapid hyperandrogenic–hypoandrogenic cycle shift. Both therapies are targeted at patients with high AR expression levels, and the combination of the two therapies could provide unexpected therapeutic benefits.

In addition, poly-adenosine diphosphate-ribose polymerase (PARP) inhibitors can block PARP-mediated DNA damage repair, destroy cell homeostasis and lead to cell death. Olaparib, a new oral PARP inhibitor, is used to treat mCRPC patients with defects in DNA repair genes (such as BRCA2 and ATM) [83]. It is promising that a large clinical trial has now been designed to assess the efficacy of BAT in combination with olaparib (ClinicalTrials.gov identifier NCT03516812).

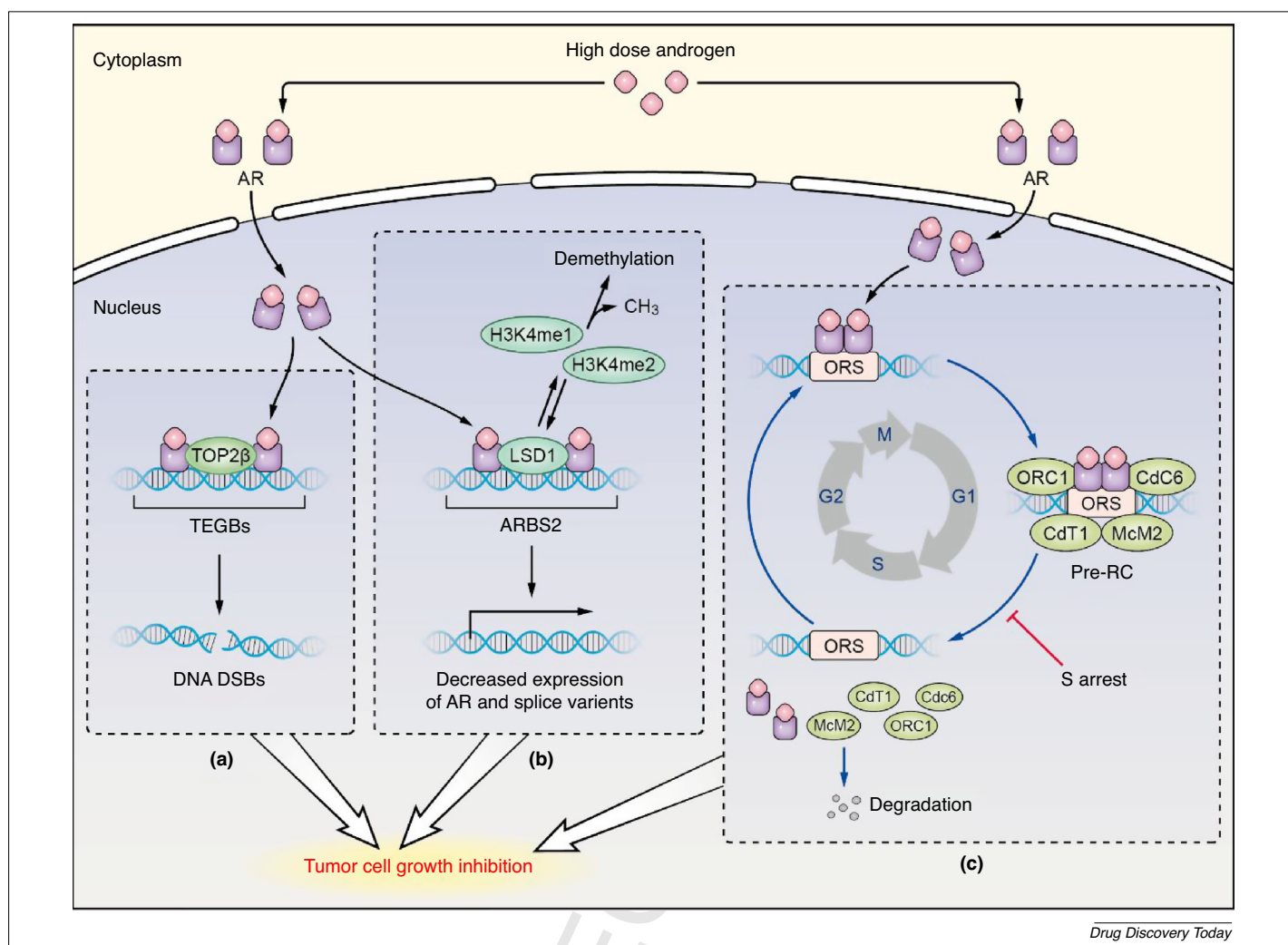


FIGURE 1

Potential mechanism of prostate cancer growth inhibition through high doses of androgen. (a) In the presence of high doses of androgen (pink spheres), androgen signaling leads to recruitment of AR (light purple squares) and TOP2β. TOP2β-mediated DNA DSBs in the regulatory regions of AR target genes result in DNA damage and cell apoptosis. (b) Activation of AR occurs at certain ARBSs and acts as a transcriptional repressor through the recruitment of LSD1 and the demethylation of activating histone marks (H3K4me1 and H3K4me2), resulting in reduced expression of full-length AR and the generation of spliced variants. (c) Under the condition of high doses of androgen, activated AR binds to the protein ORS. Ligand-dependent stabilization of AR during mitosis might inhibit the Q1 degradation of AR in M phase, preventing re-licensing for DNA replication during G1, resulting in S-phase arrest.

### Side effects of BAT and quality of life reports

Feltquate *et al.* reported that the most common side effects of BAT are grade 1 and 2 hepatitis, hot flashes, fatigue and female breast development, all of which are known effects of hormonal therapies [62]. Schweizer and co-workers mentioned in their study that adverse reactions were mostly mild ( $\leq$  grade 2), such as nausea, fatigue, hair loss, edema and neutropenia. Two out of 14 patients Q25 had an asymptomatic pulmonary embolism, but no new pain, skeletal events or urinary tract obstructions were reported during treatment [79].

The results of the BATMAN Phase II study concluded that men treated with 6 months of ADT had an improved quality of life after the first round of BAT. The most common testosterone-related adverse reactions were edema (38%) and hot flashes (52%) [80]. Patients tolerated BAT well without dose-related toxic reactions,

and reported no significant change in their quality of life. They also showed a borderline statistically significant improvement in hand-grip strength [64], and one to five of the patients had improved sexual function [81].

Many of the current reports on BAT pay particular attention to cardiovascular events. Teply *et al.* reported severe (level 3) cardiovascular events during BAT treatment, including myocardial infarction and pulmonary embolism, but metabolic parameters did not show a pattern of increased cardiovascular risk based on changes in inflammatory biomarkers, lipids or glucose metabolism [81]. Several retrospective studies and randomized trials have shown that testosterone increases the risk of cardiovascular diseases [84,85], which has led the FDA to issue warning statements about the potential cardiovascular risks. However, there have been no BAT trials with sufficient capacity to evaluate cardiovascular events.

TABLE 4

Summary of the results of completed trials with BAT

Authors	No. of patients	Patient population	Treatment regimen	Outcomes	Refs
Feltquate <i>et al.</i> (2006)	36	Progressive PC (increasing PSA and clinical metastases)	Testosterone on days 1 to 7, and an estrogen patch on days 8 to 21	No imaging and clinical progress, and PSA decreased continuously	[62]
Q3 Szmulewitz <i>et al.</i> (2009)	15	Early CRPC (with minimal metastatic disease)	Transdermal testosterone at 25, 5.0 or 75 mg/day	One patient experienced symptomatic progression, and 20% (3/15) demonstrated a decrease in PSA (the largest was 43%)	[64]
Morris <i>et al.</i> (2009)	12	CRPC (disease burden or symptoms not designated)	Testosterone via 5 mg transdermal patch or 1% gel for 1 week, 1 month, or until disease progression	30% of the patients showed PSA declines; one patient showed a PSA decline of >50% from baseline	[65]
Schweizer <i>et al.</i> (2015)	16	Asymptomatic CRPC with low to moderate metastatic burden	Testosterone (400 mg intramuscularly day 1 of 28) and etoposide (100 mg oral daily; days 1 to 14 of 28)	50% (7/14) experienced PSA decline, with 28.6% (4/14) showing a PSA response of ≥ 50%. Of the 10 patients who had evaluable soft tissue metastases, 50% had assessable imaging regression	[79]
Schweizer <i>et al.</i> (2016)	29	HSPC (with low metastatic burden or biochemically recurrent disease, who achieved PSA < 4 ng/dl after 6 months of ADT)	Testosterone 400 mg intramuscularly on days 1, 29, and 57	59% (17/29) had a PSA level of < 4 ng/mL at 18 months; 80% (8/10) objective responses were observed (four complete; four partial)	[80]
Teply <i>et al.</i> (2018)	30	mCRPC post progression on enzalutamide	Alternating 3-month cycles of BAT (400 mg intramuscularly on days 1, 29 or 57), followed by 3 months of ADT alone	30% achieved PSA response; 29 patients progressed on BAT, 52% regained PSA response to enzalutamide treatment	[81]

### Concluding remarks and future perspectives

The historical view of androgen as an energy source or driver of malignant growth of PC has proven contradictory. Numerous studies have reported the safe results of TRT application in non-PC and PC populations, indicating that the actual health benefits of alleviating TD symptoms and improving quality of life might outweigh the theoretical but unproven risk of PC progression.

New clinical and experimental data suggest that the high androgen levels induced by TRT could be protective against progression or subsequent tumor recurrence in early-stage PC. In metastatic or locally progressive PC, TRT is more likely to be discontinued owing to worsening clinical symptoms or PSA progression, which could be due to increased levels of AR and AR mutations in PC cells following endocrine therapy, resulting in increased sensitivity of cancer cells to androgens.

It is evident that TRT still needs to be used with caution in patients with progressive PC. But the indications from the available studies are that, after rigorous screening, TRT can be applied to

hypogonadism in patients with AS or low-risk PC after radical treatment.

BAT, which is based on studies of TRT, is an emerging innovative strategy that has changed our understanding of testosterone and its interaction with PC. Existing evidence supports BAT as an appropriate therapy for men with treated PC, particularly those men with low to moderate metastatic burden or castration-resistant cancer. However, not all patients exhibit a positive response to BAT, which is common in all cancer treatments, although it emphasizes that there is still much research to be done before BAT can be widely applied.

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