The 5ARI Withdrawal Syndrome (5ARI-WS)

The Silenced Androgen Receptor (AR) Theory:
Explaining persistent side effects arising from 5alpha reductase inhibitor (5ARI) use

By “Awor” and “Mew”, Administrators of Propeciahelp.com (August 2010)

1. Abstract

Increasing evidence is starting to accumulate from doctors, scientists, patient groups and online discussion forums, whereby the seemingly unrelated 5ARI substances finasteride, dutasteride, isotretinoin and saw palmetto extract are causing consumers to suffer from long-term, irreversible and serious health damage. The vast array of persistent sexual, mental and physical adverse reactions have a clear denominator in that they all relate to androgen mediated functions.

Individuals affected by this syndrome often develop an acquired form of treatment-resistant tertiary hypogonadism, low Vitamin D3 and low 3α-diol-G values, amongst numerous other hormonal imbalances. The acquired hypogonadism can manifest while on the 5ARI, but typically occurs within days or weeks after discontinuation. Increasing androgen levels by various means usually results in little or no symptomatic improvement; often a deterioration occurs. Such observations have led a number of doctors and the authors of this paper to believe a novel form of chemically induced androgen resistance is involved.

We propose that reducing DHT by 32-94%, depending on the 5ARI inhibitor used, leads to androgen receptor hypersensitivity. The combination of a hypersensitive AR, and the return of baseline DHT levels after 5ARI withdrawal, results in a sharply overexpressed AR signal. This in turn triggers an AR negative autoregulation mechanism, which silences the overexpressed AR signal.

Because this silencing is of persistent nature, we propose that an epigenetic process is involved, possibly in the form of DNA methylation. In this paper we demonstrate that AR gene expression has an important influence on 3α-HSD induction. Beyond physical and sexual AR-mediated dysfunction, we propose a silencing of the AR signal is likely to cause a sharp drop in the 3α-HSD metabolized neurosteroids THDOC, pregnanolone and allopregnanolone. Downregulation of these critical neurosteroids can lead to depression, anxiety and various neurological problems. We believe that this mechanism is a strong contributing factor in the previously reported cases of isotretinoin and also finasteride related suicides.

As the described syndrome seems to apply to 5ARIs in general, which typically manifests after 5ARI withdrawal, we have termed this enigmatic medical problem the "5ARI Withdrawal Syndrome" (5ARI-WS). This paper documents the theory related to 5ARI-WS, which is based on the interaction of four known and scientifically documented molecular mechanisms:

1. AR overexpression (hypersensitivity) as a result of decreased androgen (DHT) availability
2. Silencing of AR signal through negative autoregulation as a result of overexpressed AR signal
3. Lacking induction of the 3α-HSD enzyme as a result of a silenced AR signal
4. Epigenetic changes in homeostasis resulting in permanently altered levels of gene expression

2. Introduction

Increasingly overwhelming evidence is starting to accumulate from doctors, scientists, patient groups and online discussion forums, whereby seemingly unrelated substances such as finasteride, dutasteride, isotretinoin and saw palmetto extract (SPE) based preparations are causing young consumers to suffer from long-term, irreversible and serious health damage. The experienced persistent side effect have a clear denominator in that they all relate to physiological and psychological functions which require androgens to function correctly:

- Loss of libido (1) (2)
- Low energy, fatigue (1)
- Depression (including suicidal depression)* (3) (4) (5) (6)
- Impaired thought processes* (7)
- Memory failure* (7)
- Erectile dysfunction (8) (9) (2)
- Penile atrophy (9)
- Impaired spermatogenesis (10) (11) (12)
- Muscle atrophy/wasting (13) (14) (15)
- Gynecomastia (16)
- Dry skin and dry eyes (17) (18) (19)
- Prostate problems (2)
- Metabolic syndrome (20)
- Osteoporosis (21) (22)
- Anxiety and sleep disorders, muscle twitches/fasciculations (23) (24) (25)

* Indirect action through 3α-HSD as described later in this document

Of substantial note is that most side effects typically surface or reach full extent between 1-14 days after <u>quitting</u> the 5ARI substances. Androgen dependent tissue atrophy (penile, scrotum, muscle, prostate) often takes weeks or months before becoming apparent, while osteoporosis typically takes years to develop. Of equally important note is that attempts to supplement androgens (TRT, DHT) often make symptoms worse.

In addition to the previously listed symptoms, the following blood values have been highly correlated with affected patients:

- Low LH/FSH and Testosterone values
- Low 3α-diol-G values
- Low 25(OH) Vitamin D3 values (26)

From a pharmacological point of view, the common denominator between the involved 5ARI substances is the reduction of DHT at the cellular and circulating level through 5AR inhibition. Indeed, finasteride (27), dutasteride (27) (28) (29) , isotretinoin (30) (31) (32) (33) (34) and saw palmetto extract (35) (36) (37) (38) (39) (40) exert their effect on BPH, hair loss and acne through the potent inhibition of 5-alpha reductase.

As the described syndrome seems to apply to 5ARIs in general, and manifests mainly after 5ARI withdrawal, we will henceforth refer to the enigmatic medical problem described in this paper as the "**5ARI Withdrawal Syndrome**" (5ARI-WS).

To date, the vast majority in the medical and scientific community remain unaware that 5ARIs can cause permanent health damage. Nonetheless, an increasing body of media and medical awareness is starting to recognize aspects of this serious condition, as evidenced below:

- 2006 - Swedish Medical Products Agency begins safety investigation into reports of persistent sexual side effects which continue despite quitting finasteride (Propecia). [Original | Google English translation]
- 2007 - National Swedish TV interviews a 5ARI-WS sufferer, Merck, and research scientists about the damaging effects of Propecia and possibility of permanent sexual side effects. [Watch on Youtube]
- 2007 - Dr. Eugene Shippen, M.D. & author of "The Testosterone Syndrome", speaks out about 5ARI-WS ("The Proscar/Propecia problem - "rare" but dangerous reactions of 5 alpha reductase inhibitors – the "Post Propecia Syndrome") in his itinerary for the 15th Annual World Congress on Anti-Aging Medicine & Regenerative Biomedical Technologies. [Source]
- 2008 - Swedish Medical Products Agency concludes safety investigation, adds persistent erectile dysfunction after discontinuation of finasteride (Propecia) as officially reported side effect & possible outcome from use. [Source, pg. 3, section 4.8]
- 2009 - UK's MHRA updates finasteride 1mg prescription information to include permanent erectile dysfunction as a reported side effect and possible outcome of use. [Source, pg. 7, section 4.8]
- 2009 - Swedish TV broadcasts follow-up news story on persistent sexual side effects from Propecia. Despite long denying it, Merck, Sharpe & Dohme Sweden required to inform customers of this possible outcome from use via labeling updates. [Watch on Youtube | News article – Swedish, Google English translation]
- 2009 - Dr. John Crisler, D.O. speaks out at a medical symposium on the dangers of finasteride, its dangerous effects on the endocrine system and possibility of permanent hypogonadal sexual, mental and physical side effects. [Watch on Youtube]
- 2010 - Dr. Michael Irwig, M.D., F.A.C.E, and Director of Andrology at George Washington University launches clinical study into persistent finasteride sexual side effects from Propecia in young men aged 18-40. [Source | Details & updates]
- 2010 - Dr. Alan Jacobs, M.D. & Neuroendocrinologist confirms existence of irreversible hypogonadism from finasteride use and 5ARI-WS, unlisted mechanisms of action of finasteride/neurosteroid inhibition, and the possibility of acquired androgen resistance as a cause for persistent finasteride side effects. [Source – article 1, article 2, article 3]
- 2010 - The Italian House of Representatives Hon. Alexander Maran (Democratic Party) presents written parliamentary question to Italian Minister of Health, regarding the risks of permanent sexual, mental and physical side effects from finasteride use, questionable safety of Propecia use in young men, and what the Ministry of Health intends to do to protect Italian consumers, and investigate and treat 5ARI-WS in men suffering from the condition. [Source]

Much of the available clinical experience and data, on which this paper is based, relates to men who have taken finasteride. Additionally, from growing anecdotal evidence and member stories on online discussion forums such as propeciahelp.com, as well as other patient groups, it is becoming increasingly clear that other 5ARI drugs and substances such as dutasteride, isotretinoin and saw palmetto extract can cause similar side effects to those of finasteride users, which may persist long-term.

According to user experience of over 1000 men on www.propeciahelp.com, numerous men who quit the drug experience persistent side effects and acquired form of tertiary hypogonadism which remains highly resistant to treatment. In many of these men, supplementation of testosterone or DHT has little, if any, impact on long-term symptomatic relief. As noted by certain doctors (41) (42), there are men who, despite having normal testosterone values after quitting, continue to experience a vast array of hypogonadal-like symptoms, whereby the available androgens often seem to have no effect. As a result, many sufferers and some doctors (41) (42) hypothesize that the mechanism of these persistent hypogonadal symptoms and loss of androgenic action are due to an acquired and novel form of 5ARI-induced androgen resistance.

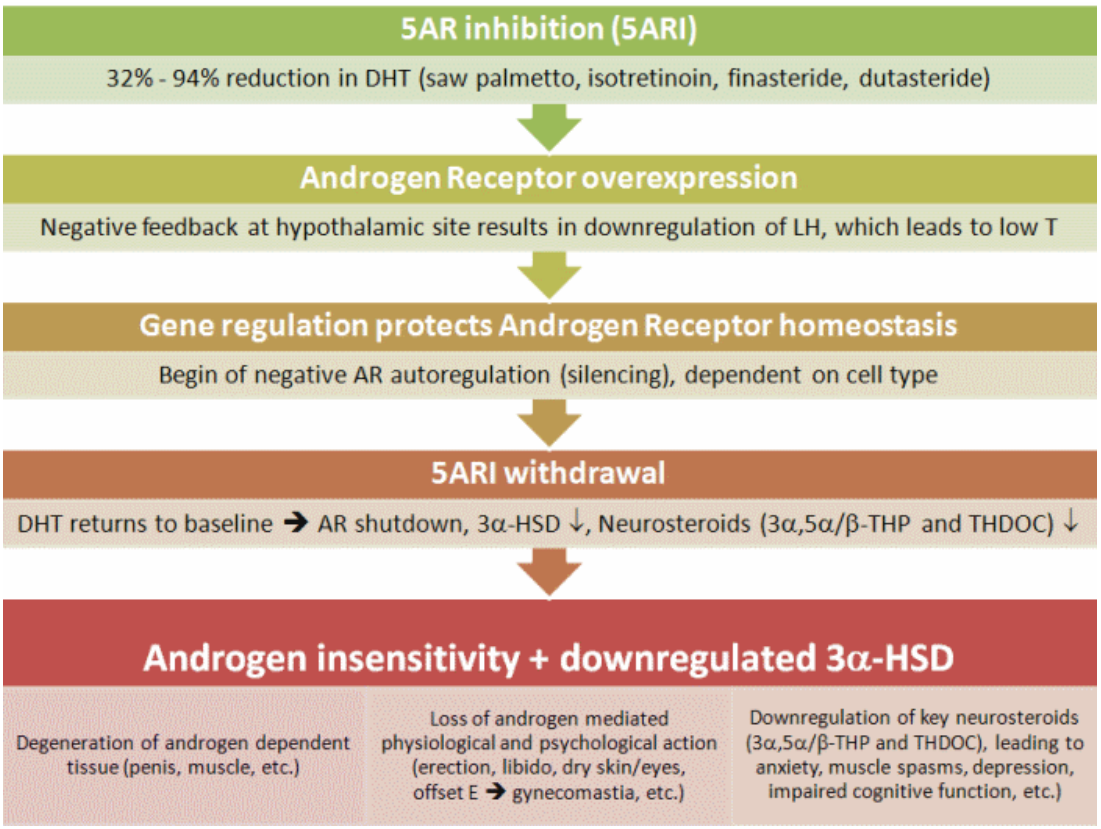
We, the authors of this paper, have spent many months researching scientific publications, compiling case studies, and reviewing adverse reaction reports related to these 5ARI substances. An extensive meta-analysis of existing scientific research material was then used to correlate the timeline and profile of the adverse reaction scope with known molecular mechanisms, in an effort to explain the possible causes of this problem in further detail.

Through this process, we believe to have arrived at a groundbreaking new finding; that is, that all 5ARI substances can cause a common set of persistent side effects resulting from their common mechanism of action, namely the reduction of DHT. As part of our work, we propose a possible molecular mechanism, which may, for the first time, plausibly explain Accutane (isotretinoin) related suicides (43).

Unfortunately, as of June 2010, nothing has been published concerning persistent 5ARI induced side effects or 5ARI-WS from a scientific point of view. Until this occurs, the following theory is meant to be a source of direction and inspiration for future scientific investigation.

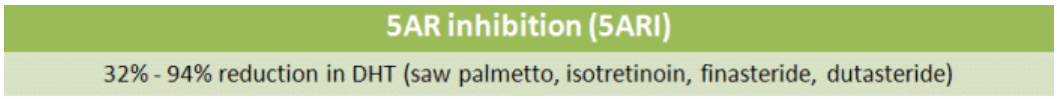
3. The 5ARI-WS Theory

The following illustration provides an overview of the 5ARI-WS theory:



In the following, each of the above illustrated phases will be explained in further detail.

Step 1



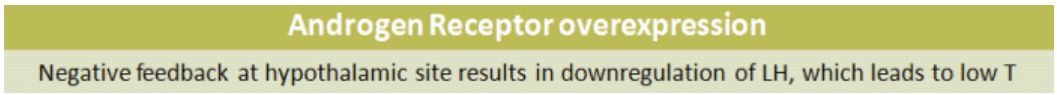
DHT is synthesized from Testosterone by the 5alpha reductase enzyme*. Two isoforms are known to exist: Type 1 and 2 (44). Recently, an additional type 3 isoform has been discovered (45). The main mode of action and common denominator of all 5ARI's is that they reduce DHT levels at the circulating, and above all cellular level in different parts of the body, depending on which isoform is inhibited (46).

* Note: Recent research challenges this pathway and suggests that 5a-reduction rather catalyzes 4-dione into 5a-androstane-3,17-dione, which then undergoes a 17keto-reduction step to DHT through 17b-HSD's (47).

As anti-androgenic and androgen deprivation therapy agents, 5ARIs have a broad range of mechanisms and resultant sexual (48) (49) (50) (51), physiological/neurological (52) (53) (54) and hormonal (55) (56) (57) effects. Depending on the agent (saw palmetto, isotretinoin, finasteride, dutasteride), a significant (32% - 94%) reduction in DHT and neurosteroids (THDOC, Pregnanolone, Allopregnanolone) will result from 5ARI use.

While on the drug, and given the above consequences of inhibiting 5AR and related hormonal effects, side effects can sometimes arise in a subset of men which lead them to stop taking the 5AR-inhibiting substance. While the full extent of 5ARI-WS symptoms typically appears between 1 to 14 days following 5ARI withdrawal, it is also possible that the involved mechanisms can start occurring while still on the 5ARI substance. Such observations have been corroborated by medical professionals who have noted acquired hypogonadism can occur either while on the medication or shortly after discontinuation (41) (42).

Step 2



Affected men often present with low LH, FSH and T values after discontinuing the 5ARI medication. Although few men have baseline hormonal values available before finasteride use, it must be assumed that most individuals had no pre-existing medical conditions and were not hypogonadal before having taken a 5ARI. This is supported by the low statistical probability (2.5%) of young men in their 20s being affected by hypogonadism (58).

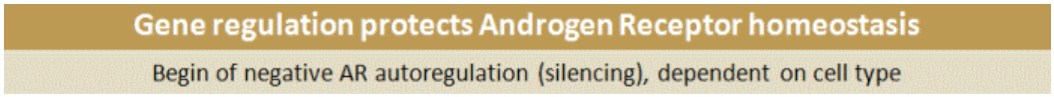
The presentation of low LH, FSH and T values has been diagnosed in such patients as tertiary hypothalamic hypogonadism, implying a hypothalamic failure (59). This is remarkable because normally, low T would be expected to lead to a positive GnRH response as a result of negative feedback on the hypothalamus, which in turn would stimulate LH and T production (60). With most of those affected, this is not the case.

Given the fact that androgens negatively regulate GnRH production (61), we postulate the acquired hypogonadal state of these men is due to the body falsely sensing high androgen levels at the hypothalamus. Further extrapolating from this line of thought, we propose that some form of androgen “amplification” is causing the body to sense an excess of androgens, despite low T values being present. What could be the mechanism of such an “amplification”, which causes the body to sense an excess of androgens despite the presence of low androgen levels?

AR hypersensitivity may provide the answer. Research has shown that reducing activation of the androgen receptor (either by reducing DHT through 5AR inhibition or by AR antagonism through an anti-androgen such as bicalutamide) can lead to androgen receptor hypersensitivity (62) (63) (64) (65) (66). A state of AR hypersensitivity would cause an amplification of the AR signal (67), mainly through HSP90 acetylation (68). We thus postulate that such amplification of the AR signal also occurs at the hypothalamic site, causing excessive negative regulation of GnRH production.

We thus propose that a hypersensitive AR would cause the hypothalamus to falsely sense an excess of androgens, causing the pituitary to reduce LH output and thus lower overall testosterone production to hypogonadal levels – resulting in the acquired hypogonadal values many men affected by 5ARI-WS present with clinically.

Step 3



How does AR hypersensitivity contribute to silencing the AR signal, leading to what we believe to be an acquired form of androgen insensitivity?

We propose that when DHT returns after 5ARI withdrawal and binds to the hypersensitive AR, the resulting overexpressed signal will lead to a strong negative AR autoregulatory response. In the following, we will attempt to outline the complex molecular mechanisms involved in such a response.

Overview of androgen receptor function and regulation

The androgen receptor (AR) mediates the physiologic effects of androgens (T, DHT) by binding to genomic androgen response elements (AREs), which influence transcription of AR target genes (69). In the process of transcription, the target gene DNA sequence is copied into an intermediary molecule called RNA. Transcription is controlled by other DNA sequences called transcription factors, which show a cell where genes are located, and control how often they are copied (70).

The RNA copy made from a gene is then fed through a structure called a ribosome, which translates the sequence of nucleotides in the RNA into protein (70). In some cases the protein must undergo further transformation in the form of posttranslational modification in order to become a functional gene product (typically a protein). The functional gene product (gene expression) is ultimately what exerts androgenic response in the body (69). If this gene product is not expressed at normal levels, androgen dependent functions will fail.

After 5ARI withdrawal and restoration of DHT levels to baseline, a hypersensitive AR, probably through amplification at the transcriptional level (68), will result in an overexpressed gene product if not offset by other regulation mechanisms. Several studies have documented that the androgen receptor has sophisticated mechanisms to autoregulate its own gene expression levels through negative and positive autoregulatory feedback loops (71) (72) (73) (74) (75). A (genetic network) feedback loop means that the receptor is regulated by its own gene product (76). These autoregulation mechanisms normally assure that AR gene expression remains within precisely determined ranges (73). If this were not the case, cancer, runaway muscle growth or blood clotting could occur, which would eventually lead to death.

Those affected by 5ARI-WS share a number of common hypogonadal-like symptoms; however, not all symptoms are experienced to the same degree by all affected individuals. Such variability is likely explained through the unique genetic profile of each affected individual. To date, over 200 AR target genes (androgen responsive genes – ARGs) have been identified (69), giving credence to the fact there are innumerous androgen-mediated processes within the body which can be affected to an individual degree by gene regulation mechanisms. Alternative RNA splicing can further increase the number of proteins translated from target genes (77) (78).

As described previously, transcription factors are complex molecular machines that control the expression of target genes. At any given time, depending on the context and cellular stimuli, a transcription factor will affect only a subset of its target genes (79). Proteins that modulate the activity of transcription factors, often called modulators, play a critical role in creating tissue- and

context-specific gene expression responses to the signals cells receive. Currently, nearly 300 modulators of AR gene expression are known (79). These modulators work in concert to amplify or silence gene expression (67).

It is assumed that AR hypersensitivity is the result of a set of modulators that enhance AR transcription (enhancers, promoters, activators) (67). If the resulting level of expression becomes too high, another set of autoregulation mechanisms will take effect to offset this (71) (80) (81) (75) (79) (82), for example:

- modulators and chromatin remodeling at the transcriptional level
- DNA methylation at the transcriptional level (83) (84)
- RNA splicing at the posttranscriptional level (78)
- RNA sequestration at the translational level
- Posttranslational modifications such as phosphorylation, acetylation, sumoylation, etc.

Autoregulatory mechanisms can thus operate at the transcriptional, post-transcriptional, translational and posttranslational level (82), and the result of such (negative) autoregulation can be a partial or complete silencing of the AR signal (72). As stated above, transcription factors only affect a subset of target genes. Given that modulators only affect a subset of transcription factors, it is clear that gene regulation of the androgen receptor is a very sophisticated and fine grained mechanism, allowing for an individual regulation of the expression of target genes.

The effect of these modulators or the balance thereof can be temporarily modified by epigenetic regulation processes such as phosphorylation, or more permanently modified by processes such as DNA methylation. Such modifications are considered to be responsible for more or less permanent changes in gene expression levels. It has been hypothesized that epigenetic changes in gene expression are the basis for persistent side effects from drugs. This includes permanent side effects from 5ARI class substances such as isotretinoin (85), and presumably other 5ARIs such as finasteride, dutasteride and even saw palmetto extract (SPE).

Scientific research has shown that methylation of the androgen receptor promoter CpG island is associated with loss of androgen receptor expression in prostate and human leukemia cancer cells. The demethylating agent 5-aza-2’ deoxycytidine induced a re-expression of AR RNA (86) (87). This gives further credence to the concept that DNA methylation may be involved in the persistent silencing of the AR signal.

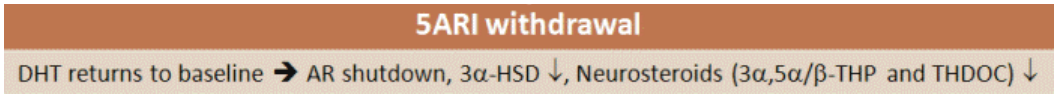
Of interesting note, a small number of affected men have reported experiencing a short period of recovery after administrating ciprofloxacin or other antibiotics. Ciprofloxacin is known to significantly alter protein levels in both directions through changes in gene expression rates over a broad range of target genes (88). This could indicate that a change in proteins which regulate AR expression levels allowed for a short lasting “recovery” phase, giving further support to the hypothesis that some form of misguided AR negative autoregulation mechanisms lies at the root of this problem.

Theory related to AR gene regulation

In summary, theory related to gene regulation in the context of 5ARI-WS is as follows:

- According to research, it seems plausible that AR hypersensitivity, due to androgen depletion, occurs through amplification at the transcriptional level.
- Given the sophistication of AR autoregulation mechanisms, it seems plausible that an errant, overexpressed AR at the transcriptional level can lead to downstream negative autoregulation, i.e. at the posttranscriptional, translational or posttranslational level. Such gene regulation mechanisms can partially or totally silence the AR signal in response to an overexpressed AR signal.
- Given the cell specific expression of 5AR isoforms, and the fine grained nature of AR autoregulation mechanisms which vary depending on cellular context and genotype of the affected person, it seems plausible that not every AR-mediated body function is affected to the same degree and that not all suffering men have the same symptomatic profile.
- Given the role of epigenetic processes, such as chromatin remodeling and DNA methylation in the persistent alteration of homeostasis (82) (89) (85), it seems plausible that the combination of these elements could work in concert to persistently downregulate AR gene expression.

Step 4



Consumers experiencing side effects from 5AR inhibitors will typically choose to withdraw from using the medication in order to resolve their side effects, as per the manufacturer's claims that side effects experienced will resolve upon discontinuation of the medication (90).

Within 7 days of quitting the 5ARI medication, as presumably functional 5AR enzyme returns, baseline DHT levels are restored (91). Following the suppression of DHT by over 70% while on the medication (i.e. finasteride) (91), the return of baseline DHT levels represents a rapid increase of DHT concentrations at the cellular level by over 333% (from 30% to 100%, which equates to 1 / 0.3 = 3.33). Depending on where the body has determined the reference point for "normal" androgen levels to be (i.e. 30% mark or 100% mark), and assuming the body has become "accustomed" to a significantly reduced level of DHT (and thus gene expression) while on the medication, we postulate that a rapid restoration of baseline (100%) DHT levels after a period of androgen deprivation may be perceived as excessive by the body.

Assuming that this flood of DHT now binds to an already overexpressed and hypersensitive AR, we propose that the resultant strongly overexpressed AR signal, combined with the body's perception of a much lower "normal" level of gene expression, leads to a substantial negative and persistent autoregulatory response. We propose that this response effectively silences the AR signal and that in effect, all dependent downstream processes (e.g. induction of 3α-HSD enzyme) become downregulated. We propose that methylation is involved in the context of this negative autoregulatory response, leading to persistent silencing of the AR signal.

4. Evaluation of the Hypothesis

Evidence of AR silencing: Downregulation of 3α-HSD and low 3α-diol-G values

Most tested 5ARI-WS patients tested via serum assays display low levels of 3α-androstanediol glucuronide (3α-adiol-G). What is this telling us? In order to answer this question, we need to precisely understand what regulates the production of 3α-diol-G.

5α -DHT is reduced by the enzyme 3α-HSD to the inactive androgen 3α-diol-G. (92). 3α-diol-G is an inactive androgen because it does not bind to the androgen receptor (93). By deactivating androgens, 3α-HSDs determine the amount and the type of androgen available for the androgen receptor and hence affect transcription of genes under androgen control (92).

3α-diol-G is considered to be a marker of androgen metabolism (94) (95). Studies have proposed that 3α-diol-G is a marker of 5α-reductase activity (96) as well as a marker of androgen insensitivity (97) (98). However, little has been published on how androgens (DHT, T) induce 3α-HSD to transform these hormonal precursors into 3α-diol-G.

Two theoretical possibilities exist:

- Direct: Androgens induce 3α-HSD expression by directly influencing its modulators
- Indirect: Androgens modulate AR gene expression, which in turn induces 3α-HSD

Unfortunately, very little has been published on the regulation of 3α-HSD. Research related to the gene regulation of 3α-HSD in bacteria (a short chain version of 3α-HSD), may give us some clues, although this is purely hypothetical. In the hsdA encoding gene of 3α-HSD in the Comammonas testeroni, hsdA is regulated by several activators and repressors which interact with steroid substrates (99). Repressor repA positively regulates hsdA gene expression through de-repression by forming a binding complex with testosterone (99).

This does not answer the question, however, as to which mechanism (direct, indirect) is the most important in determining 3α-HSD gene expression in humans, as 3α-HSD in humans belongs to the AKR superfamily (100). Our approach to this question was to study documented CAIS (Complete Androgen Insensitivity Syndrome) cases with an androgen receptor defect, effectively mimicking a human AR “knock-out” model.

Should 3α-HSD be mainly induced by testosterone or DHT, then 3α-diol-G levels would be expected to fall within normal ranges with CAIS subjects. If, however, 3α-HSD is induced by AR gene expression, and since AR gene expression would be missing in such CAIS patients, we would expect to find substantially lower levels of 3α-diol-G in these patients.

A study of two CAIS patients with a post-receptor defect, but normal wild type (WT) AR gene and normal mRNA size provided suitable data. Testing of these two patients revealed the following (101):

- No detectable androgen binding to AR
- Normal to high testosterone values
- High LH/FSH
- Normal 5AR activity
- LOW 3α-diol-G (only 25.9 – 38.3% of normal male reference range level)

These results suggest that direct induction of 3α-HSD through androgens, or perhaps other hormones, can only account for roughly 25 – 40% of total 3α-HSD induction (perhaps through de-repression by T/DHT as with short chain version of 3α-HSD). Considering that the patient whose result was 25.9% (50/193 g/24hr) had testosterone levels much closer to the male reference level of 5.8 ng/dl (101) than the other, which was higher, a figure of roughly 25% is probably more realistic than is 38.3%.

This result suggests that 3α-HSD is mainly (~75%) induced by AR gene expression. Hence, low 3α-diol-G values would infer low or lacking AR gene expression.

Verification of 3α-diol-G as a marker of AR gene expression

To verify this hypothesis, we should be able to apply 3α-diol-G as a marker of AR gene expression to prove or disprove the involvement of some form of silencing of the AR signal.

With WT men, 3α-diol-G and testosterone levels are known to be significantly correlated (r = 0.37; p less than 0.01) (102). In other words, an increase/decrease in testosterone should lead to a proportional and significant increase/decrease in 3α-diol-G.

This is also evidenced by the following graph from a study, which compares mean T, DHT, 3α-diol-G and ADT-G values from before and after castration (103):

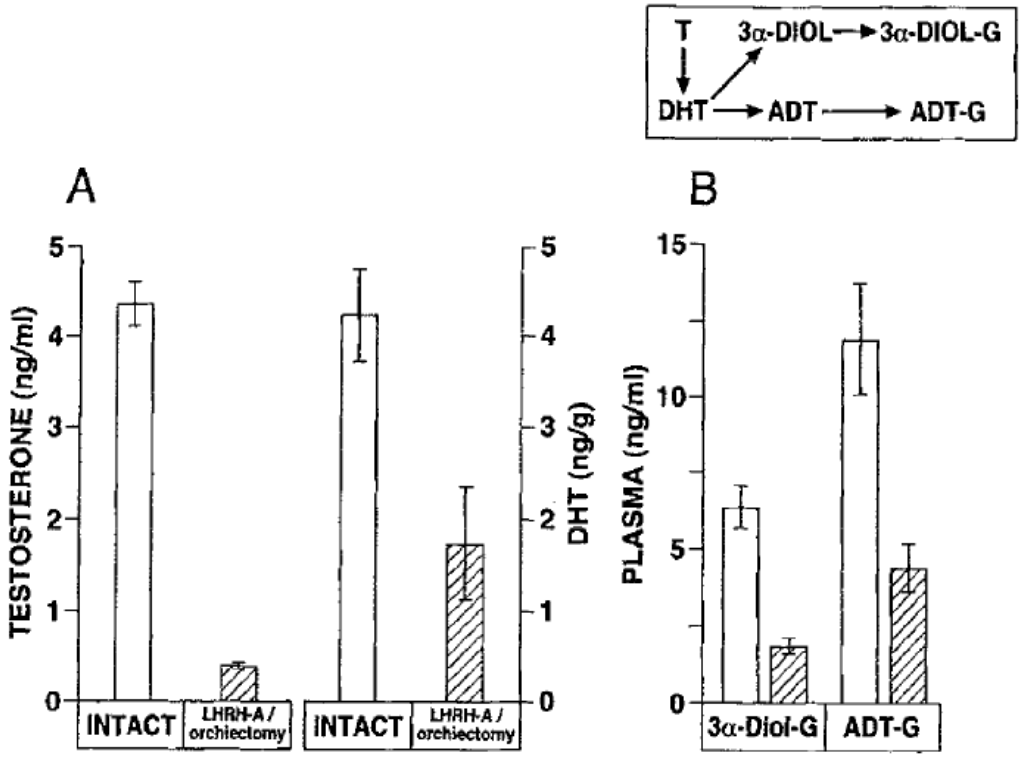


FIG. 2. (A) Effect of castration on the serum levels of testosterone (T), on one hand, and on the concentration of the active androgen 5α-dihydrotestosterone (DHT) remaining in prostatic cancer tissue after castration, on the other hand. Note the relatively small effect (approximately 60%) of castration on intraprostatic DHT concentration as compared with the 90% fall in serum T. LHRH-A = luteinizing hormone-releasing hormone agonist.[63] (B) Plasma concentrations of androstane-3α, 17β-diol glucuronide (3α-Diol-G), and androsterone glucuronide (ADT-G) in 20 intact (□) and 18 castrated (▨) men with prostate cancer. Patients were of similar ages.[13]

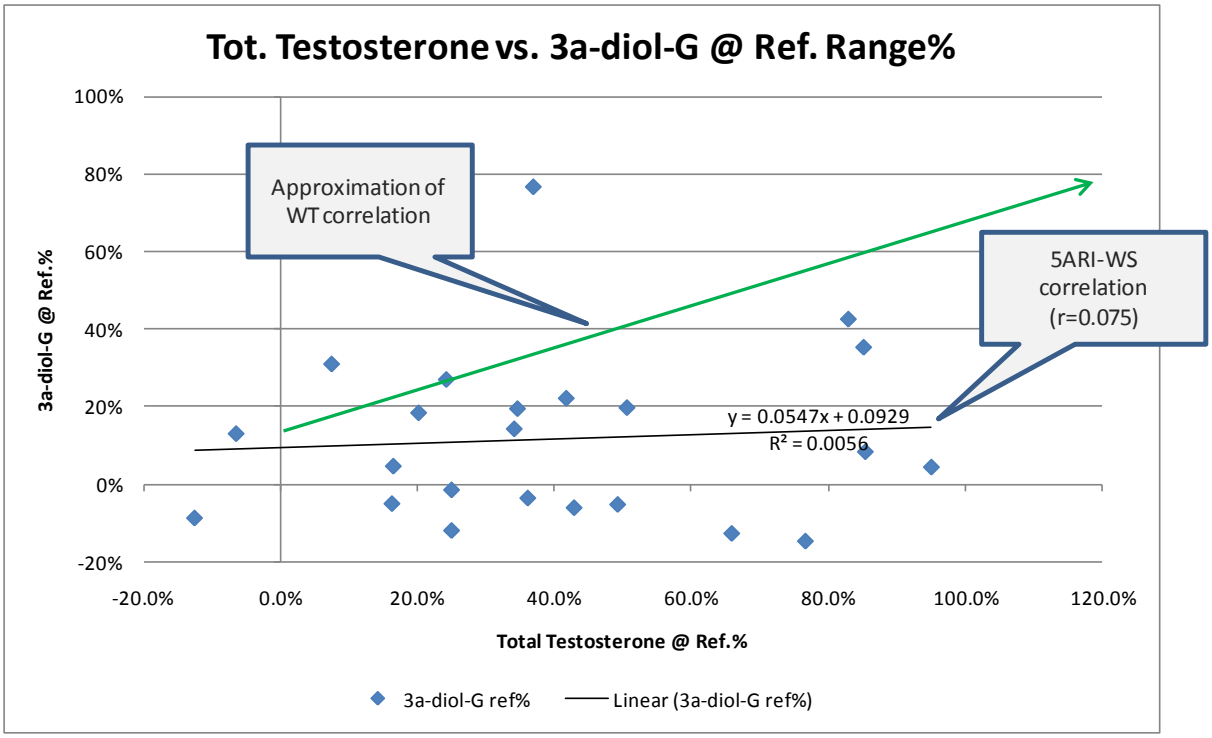
The correlation analysis between testosterone and 3α-diol-G levels from 5ARI-WS patients would allow us to verify this hypothesis.

We thus proceeded to analyze available patient data from the Propeciahelp.com website with the help of a scatter plot diagram, where 3α-diol-G was plotted against total testosterone levels. All values were normalized against reference ranges using following formula:

norm(Val) = (Val – Lr) / (Ur – Lr)

Where Val = assay value, Lr = lower ref. range limit, Ur = upper ref. range limit

The result was as follows:



The data from 5ARI-WS patients (90) yielded an almost flat linear regression line, indicating practically no correlation between testosterone and 3α-diol-G values. This is in strong contrast with the above cited studies. Given that low 3α-diol-G values suggest low AR gene expression, this graph strongly supports the notion that higher testosterone values seem to have little to no effect on (functional) AR gene expression.

Linking low 3α-HSD induction to mental symptoms (Depression, Cognitive difficulties)

To date, four functional isoforms of 3α-HSD [i.e. types 1, 2 and 3 3α-HSD, and 20α(3α)-HSD] have been characterized on the basis of their affinity for 5α-DHT. Types 2, 3 and 20α(3α)-HSD are also actively expressed in the human brain, whereas 3a-HSD type 1 is not (104). All types are likely to be induced by AR gene expression.

5ARI-WS often suffer from a wide array of mental symptoms which include:

- Various degrees of depression (from light to suicidal)
- Slowdown of cognitive function, often described as “brain fog”. This is sometimes accompanied by slurring of speech.
- Impairment of short and long-term memory
- Disturbed sleeping patterns and insomnia
- Social phobia
- Anxiety
- Emotional flatness
- Lack of libido, loss of sexual interest
- Loss of aggression and self-confidence

Scientific publications (105) (104) (106) have shown that:

- Major depressive disorders are linked to a deficiency in 3α-reduced neuroactive steroids, specifically allopregnanolone (3α,5α-THP).
- Allopregnanolone (3α,5α-THP), pregnanolone(3α,5β-THP), and allotetrahydrodeoxycorti-costerone(THDOC) are perhaps the most potent positive modulators of GABAA receptor function.
- Neurosteroids may regulate their own biosynthesis through modulation of GABAA receptor function.
- Neurosteroids are also involved in the control of a number of behavioral, neuroendocrine and metabolic processes such as regulation of food intake, locomotor activity, sexual activity, aggressiveness, anxiety, depression, body temperature and blood pressure.
- Dysfunction in GABAergic neurotransmission may contribute to the pathophysiology of mood disorders.
- Insomnia has been linked to a deficiency GABAA-receptor-active 3α-reduced neurosteroids.
- Allopregnanolone (3α,5α-THP) plays a key role in inhibiting fear and anxiety.
- Allopregnanolone is reduced in prefrontal cortex in Alzheimer's disease patients.

The above listed publications (105) (104) are summaries of current knowledge and understanding of neurosteroid metabolism and action. From these it is clear that the most potent neurosteroids (allopregnanolone, pregnanolone and THDOC) are synthesized by 3α-HSD. These are especially important because of their central role in modulating GABAA receptor function and regulation of other neurosteroids.

Given that AR gene expression seems to have an important influence on 3α-HSD induction, as previously evidenced by low 3α-diol-G levels with CAIS patients, it becomes plausible that a silencing of the AR signal would cause a sharp drop in 3α-HSD metabolized neurosteroids which in turn would lead to an onset of the above listed symptoms.

It must be noted that 3α-HSD already gets downregulated while on the 5ARI (due to reduction in DHT, and as evidenced through concomitant decrease in 3α-diol-G values during usage of finasteride) (107). We propose that the presumably abrupt silencing of the AR signal, once DHT returns and binds to hypersensitive AR, would lead to a further, strong downregulation of 3α-HSD. This in turn would cause a sharp drop in these neurosteroids, resulting in an almost immediate onset of strong

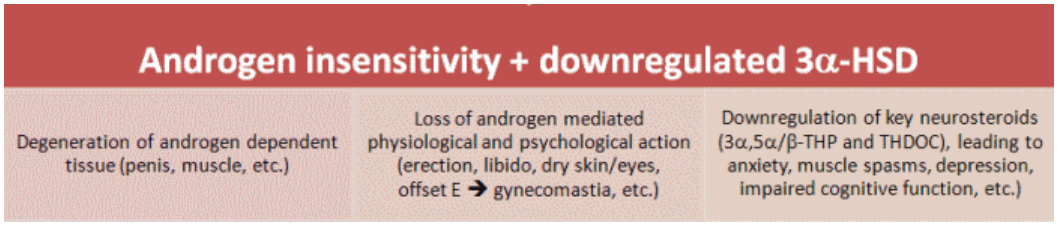
depression and anxiety in many of the affected persons. We regard this as a plausible explanation for the suicides which have occurred in relation with isotretinoin (Accutane) and in certain cases with finasteride use. Such negative changes in neurosteroid levels, particularly significantly reduced allopregnanlone values, provide a likely explanation for the many cognitive issues ("brain fog") which are often reported by affected individuals.

Conciliating AR hypersensitivity with LH downregulation in the light of silenced AR signal:

As previously mentioned in this paper, an important clinical observation after quitting the 5ARI is a sharp drop in LH and testosterone levels, leading to tertiary hypogonadism in affected patients. At first thought, this effect is not consistent with the concept of a silenced AR signal – that is, in the case of a silenced AR signal, the lack of AR induction at the hypothalamus would lead to a surge in LH and thus testosterone, not a drop. Therefore, for the manifestation of hypogonadal LH and testosterone values to be compatible with the concept of a silenced AR signal, the signaling mechanism at the hypothalamic site must be uniquely different.

A unique mode of AR action at the hypothalamic site seems to be the case. Recent research has demonstrated that the regulation of GnRH by AR is not only through genomic mechanisms but rather differentially through a novel form of non-genomic membrane mediated AR signal transduction (61). This difference in the AR signaling mechanism between the hypothalamus and the rest of the body makes it plausible that AR autoregulation functions differently or does not apply at the hypothalamic site.

Step 5



Following the silencing of the AR signal described in STEP 4, which results from the returning DHT binding hypersensitive androgen receptors, a persistent form of AR silencing likely results. This is evidenced by the regression of androgen mediated tissue (penile, muscle and prostate tissue) as well as androgen mediated physiological processes (libido, erectile function, spermatogenesis, sebum production, etc.).

The genotype of the affected persons is currently unknown. There is much case evidence that not all individuals are affected in the same way or to the same degree. Some get only weak side effects such as mild ED or depression, while others get a very broad range of serious side effects, which include strong muscle wasting, life threatening depression, metabolic syndrome, osteopenia/osteoporosis, destruction of penile structure and complete loss of all sexual function. There appears to be a broad range between those end-points, which presumably is determined by individual genetics and the 5AR isoform which was inhibited.

We have observed that 5ARI-WS resulting from isoform specific inhibition of 5AR (finasteride = type 2 vs. isotretinoin = type 1) tends to more severely affect areas in which the inhibited isoform is strongly expressed (5AR1 vs. 5AR2 tissue).

5. Discussion: Explaining persistency

As stated earlier in this paper, hypogonadal LH and T values, while coming off the drug, are proposed to be caused by AR hypersensitivity. In some affected individuals, LH and T values appear to increase somewhat over time, but in most cases never reach normal levels again. It thus must be assumed that AR remains persistently hypersensitive to a certain degree in numerous affected men.

Why AR hypersensitivity would remain persistent, even after restoration of baseline DHT values, is unclear. Scientific literature describes AR hypersensitivity to be reversible upon restoration of normal androgen levels (63). This is often not the case as evidenced by affected men. Therefore, how could such a persistent state be explained?

- As stated prior, persistency generally could be explained by an epigenetic alteration of homeostasis through direct (chromatin remodeling, DNA methylation) or indirect mechanisms (alteration of transcription factor activity at gene promoters) (85).

- As another possible explanation, a recent study found that cell turnover rate increases with decreasing androgen levels, which may increase the rate of mutation and malignant evolution. The study further found that low androgen environments, caused either by low serum testosterone or by reduced 5a-reductase activity, select more strongly for elevated AR expression than do normal environments (108). Such a change would presumably be slow to reverse.

- Of particular note is a study in which the AR within animal model prostates, which were subjected to finasteride, remained hypersensitive even 21 days after stopping the treatment. This is a further indication that persistency of AR hypersensitivity is possible, probably due to a persistent change at the transcriptional level given the observed nuclearisation of the AR (109).

- Another noteworthy parallel to the 5ARI-WS is the Anti Androgen Withdrawal Syndrome (AAWS). Discontinuation of an anti-androgen treatment sometimes results in prostate-specific antigen (PSA) decline, often associated with clinical improvement of the prostate tumor (110) (111). These benefits typically persist for a number of months. It is known that both PSA and prostate cancer tumors are androgen dependant (112). The reduction of both could suggest that some form of AR silencing has occurred, similar to the phenomenon we are describing in this paper. With AAWS we basically have the same sequence of events as with 5ARI-WS: Androgen depletion, AR hypersensitivity, returning androgens binding hypersensitive AR, the result of which is that androgen mediated processes stop working. It is interesting to note that PSA values of many affected men are in the low range (< 0.5). It would be very interesting to test 3α-diol-G values and determine if AAWS patients also suffer from 5ARI-WS symptoms in order to confirm a possible link.

Consequences of the Hypothesis and Summary

We the authors have attempted to present here in this paper, for the first time, a unified theory to explain the 5ARI-Withdrawal Syndrome, reasons for persistent side effects, and mechanisms behind the acquired hypogonadism and androgen resistance which can result from 5ARI use. In this regard, we believe that the links which we have hypothesized between the covered 5ARI substances and the combined molecular mechanisms of the observed adverse reactions is ground breaking, in that:

- Androgen depletion through 5ARI use can result in persistent AR hypersensitivity. Hypogonadal LH/FSH and testosterone levels, commonly observed with 5ARI-WS patients, are the result of excess negative GnRH regulation due to AR hypersensitivity at the hypothalamic site.
- Withdrawal of 5ARI and return of DHT to baseline levels typically represents an increase of over 330% compared with suppressed levels while on the 5ARI (finasteride). The combination of AR hypersensitivity and a threefold increase in hormonal levels can lead to extreme AR negative autoregulatory response which involves epigenetic silencing mechanisms such as DNA methylation.
- In light of AR hypersensitivity and a misguided negative AR autoregulation, attempts to supplement testosterone and DHT can further exasperate the situation, leading to a persistent worsening of the symptoms, possibly through further methylation.
- Downstream AR mediated processes, such as the induction of 3α-HSD, become substantially downregulated as a result of the silenced AR signal. This is evidenced by low 3α-diol-G values with affected patients. Failed induction of 3α-HSD results in a critical drop in neurosteroid levels which in turn is causative for depression, impaired cognitive function and loss of sexual desire (libido).
- Androgen dependent tissues such as penile tissue, muscle and the prostate gland undergo apoptosis as a result of the silenced AR signal.
- Androgen dependent physiological processes such as the male erection are impaired. Lacking activation of sebaceous glands as a result of a silenced AR signal typically leads to dry skin and other skin problems.
- Commonly diagnosed low vitamin D3 values amongst 5ARI-WS patients suggests that the pathway for vitamin D3 synthesis is in part dependent on AR signaling. The abundance of 5AR1 in the skin gives support to this hypothesis.
- Bone metabolism, which is also androgen-mediated, can become impaired, leading to osteopenia or even osteoporosis. Low Vitamin D values further aggravate this risk factor.

We believe this problem has possible implications for the development of new therapy options for prostate cancer. The problem described in this paper makes it plausible that there are ways to persistently silence AR gene expression without continued chemical intervention. If it is possible to determine the involved molecular mechanisms, and if it would be possible to apply these locally in the prostate, medicine could potentially have a completely new set of therapy options to work with – without the risk of AR antagonists turning into agonists during therapy.

Likewise, it is only through an in-depth scientific research study on persons affected by 5ARI-WS that the molecular mechanisms of this problem will likely ever be verified. Such a study involving research scientists, androgen receptor specialists and geneticist could be coordinated and accomplished, if the medical community and health authorities were to realize the severity and broad implications of this

problem. The obtained results could shed groundbreaking new insights into the mechanisms of androgen metabolism, AR function and implications of 5AR inhibition. Genetic profiling of the affected patients would allow for the development of genetic testing prior to taking anti-androgens/5AR inhibitors to determine genetic risk factors.

To that end, the authors propose such a study involve the following areas of investigation:

- Assessment of the methylation status of the androgen receptor promoter CpG island
- Possibly genome wide assessment of methylation status
- Testing for AR post-translational modification or protein-protein interactions (gene silencing) via mass spectrometry protein profiling techniques
- Testing for AR gene expression via microarray tests
- Genetic profiling of 5ARI-WS patients in order to develop a genetic testing for determining the 5ARI-WS risk factor before 5ARI treatment
- Testing of 3α-diol-G levels with AAWS patients (pre and post withdrawal) in order to verify the link between 5ARI-WS and AAWS, and to confirm 3α-diol-G as a marker of AR gene expression.
- Study of the induction of 3α-HSD (AKR superfamily) to verify induction by AR signaling and thus further confirm 3α-diol-G as a marker of in vivo AR gene expression.

Considering the fact manufacturers of such 5ARI substances continue to market these medications as overtly safe and reversible in terms of side effects, ensures the vast majority of consumers and the medical community remain unaware of the potential life-altering or even life-threatening consequences consumers risk by using a 5ARI for hair loss or for acne treatment.

In the end, consumers can only make a decision based on the information they are given, and it is the authors' opinions that current official prescribing literature for agents such as finasteride, dutasteride, isotretinoin and saw palmetto do not provide strong enough warning labels regarding the possibility of permanent side effects from use.

Ultimately, determining the molecular mechanisms involved in 5ARI-WS will help science better understand the complex functioning of the androgen receptor and its autoregulatory mechanisms. The knowledge gained will enable safer application of and therapies with 5ARI agents, possibly shedding new insights into prostate cancer research and potentially, offering new therapy options in the future. Potential broad scale medical, media and social awareness regarding the potential risks related to 5ARI use would inform both those who prescribe 5ARI medications as well as those who consume them for cosmetic purposes, BPH and prostate cancer, so that each can make a more informed choice in the end.

The authors of this paper can be contacted as follows:

research@propeciahelp.com

6. References

1. **Leiber C, Wetterauer U, Berner M.** Testosterone and psyche. *Urologe A.* Jan 49(1), 2010, pp. 43-6 (http://www.ncbi.nlm.nih.gov/pubmed/20057987).

2. **Traish AM, Guay AT.** Are androgens critical for penile erections in humans? Examining the clinical and preclinical evidence. *J Sex Med.* May 3(3), 2006, pp. 382-404 (http://www.ncbi.nlm.nih.gov/pubmed/16681465).

3. **Kindlundh AM, Rahman S, Lindblom J, Nyberg F.** Increased dopamine transporter density in the male rat brain following chronic nandrolone decanoate administration. *Neurosci Lett.* Feb 12;356(2), 2004, pp. 131-4 (http://www.ncbi.nlm.nih.gov/pubmed/14746881).

4. **de Souza Silva MA, Mattern C, Topic B, Buddenberg TE, Huston JP.** Dopaminergic and serotonergic activity in neostriatum and nucleus accumbens enhanced by intranasal administration of testosterone. *Eur Neuropsychopharmacol.* Jan 19(1), 2009, pp. 53-63 (http://www.ncbi.nlm.nih.gov/pubmed/18818056).

5. **Zarrouf FA, Artz S, Griffith J, Sirbu C, Kommor M.** Testosterone and depression: systematic review and meta-analysis. *J Psychiatr Pract.* Jul 15(4), 2009, pp. 289-305 (http://www.ncbi.nlm.nih.gov/pubmed/19625884).

6. **Markianos M, Tripodianakis J, Istikoglou C, Rouvali O, Christopoulos M, Papageorgopoulos P, Seretis A.** Suicide attempt by jumping: a study of gonadal axis hormones in male suicide attempters versus men who fell by accident. *Psychiatry Res.* Nov 30;170(1), 2009, pp. 82-5 (http://www.ncbi.nlm.nih.gov/pubmed/19781790).

7. **Matousek RH, Sherwin BB.** Sex steroid hormones and cognitive functioning in healthy, older men. *Horm Behav.* Mar 57(3), 2010, pp. 352-9 (http://www.ncbi.nlm.nih.gov/pubmed/20079740).

8. **Blute M, Hakimian P, Kashanian J, Shteynshluyger A, Lee M, Shabsigh R.** Erectile dysfunction and testosterone deficiency. *Front Horm Res.* 37 2009, pp. 108-22 (http://www.ncbi.nlm.nih.gov/pubmed/19011292).

9. **AM, Traish.** Androgens play a pivotal role in maintaining penile tissue architecture and erection: a review. *J Androl.* Jul-Aug 30(4), 2009, pp. 363-9 (http://www.ncbi.nlm.nih.gov/pubmed/18802199).

10. **Davis-Dao CA, Tuazon ED, Sokol RZ, Cortessis VK.** Male infertility and variation in CAG repeat length in the androgen receptor gene: a meta-analysis. *J Clin Endocrinol Metab.* Nov 92(11), 2007, pp. 4319-26 (http://www.ncbi.nlm.nih.gov/pubmed/17684052).

11. **Welsh M, Saunders PT, Atanassova N, Sharpe RM, Smith LB.** Androgen action via testicular peritubular myoid cells is essential for male fertility. *FASEB J.* Dec 23(12), 2009, pp. 4218-30 (http://www.ncbi.nlm.nih.gov/pubmed/19692648).

12. **Aksglaede L, Jørgensen N, Skakkebaek NE, Juul A.** Low semen volume in 47 adolescents and adults with 47,XXY Klinefelter or 46,XX male syndrome. *Int J Androl.* Aug 32(4), 2009, pp. 376-84 (http://www.ncbi.nlm.nih.gov/pubmed/19515177).

13. **Bhasin S, Woodhouse L, Storer TW.** Proof of the effect of testosterone on skeletal muscle. *J Endocrinol.* Jul 170(1), 2001, pp. 27-38 (http://www.ncbi.nlm.nih.gov/pubmed/11431134).

14. **F, Kadi.** Cellular and molecular mechanisms responsible for the action of testosterone on human skeletal muscle. A basis for illegal performance enhancement. *Br J Pharmacol.* Jun 154(3), 2008, pp. 522-8 (http://www.ncbi.nlm.nih.gov/pubmed/18414389).

15. **Brown D, Hikim AP, Kovacheva EL, Sinha-Hikim I.** Mouse model of testosterone-induced muscle fiber hypertrophy: involvement of p38 mitogen-activated protein kinase-mediated Notch signaling. *J Endocrinol.* Apr 201(1), 2009, pp. 129-39 (http://www.ncbi.nlm.nih.gov/pubmed/19144735).

16. **Mathur R, Braunstein GD.** Gynecomastia: pathomechanisms and treatment strategies. *Horm Res.* 1997, pp. 95-102 (http://www.ncbi.nlm.nih.gov/pubmed/11546925).

17. **CC, Zouboulis.** Acne vulgaris. The role of hormones. *Hautarzt.* Feb 61(2), 2010, pp. 107-8, 110-4 (http://www.ncbi.nlm.nih.gov/pubmed/20107754).

18. **Schneider MR, Paus R.** Sebocytes, multifaceted epithelial cells: lipid production and holocrine secretion. *Int J Biochem Cell Biol.* Feb 42(2), 2010, pp. 181-5 (http://www.ncbi.nlm.nih.gov/pubmed/19944183).

19. **Schirra F, Seitz B, Knop N, Knop E.** Sex hormones and dry eye. *Ophthalmologe.* Nov 106(11), 2009, pp. 988-94.

20. **F, Saad.** The role of testosterone in type 2 diabetes and metabolic syndrome in men. *Arq Bras Endocrinol Metabol.* Nov 53(8), 2009, pp. 901-7 (http://www.ncbi.nlm.nih.gov/pubmed/20126841).

21. **Ashida K, Akehi Y, Kudo T, Yanase T.** Bone and Men's Health. The role of androgens in bone metabolism. *Clin Calcium.* Feb 20(2), 2010, pp. 165-73 (http://www.ncbi.nlm.nih.gov/pubmed/20118507).

22. **Muñoz-Torres M, Jódar E, Quesada M, Escobar-Jiménez F.** Bone mass in androgen-insensitivity syndrome: response to hormonal replacement therapy. *Calcif Tissue Int.* Aug 57(2), 1995, pp. 94-6.

23. **Bitran D, Kellogg CK, Hilvers RJ.** Treatment with an anabolic-androgenic steroid affects anxiety-related behavior and alters the sensitivity of cortical GABAA receptors in the rat. *Horm Behav.* Dec 27(4), 1993, pp. 568-83.

24. **Bitran D, Hilvers RJ, Frye CA, Erskine MS.** Chronic anabolic-androgenic steroid treatment affects brain GABA(A) receptor-gated chloride ion transport. *Life Sci.* 1996, pp. 573-83 (http://www.ncbi.nlm.nih.gov/pubmed/8632710).

25. **Griebel G, Perrault G, Simiand J, Cohen C, Granger P, Depoortere H, Françon D, Avenet P, Schoemaker H, Evanno Y, Sevrin M, George P, Scatton B.** SL651498, a GABAA receptor agonist with subtype-selective efficacy, as a potential treatment for generalized anxiety disorder and muscle spasms. *CNS Drug Rev.* Spring 9(1), 2003, pp. 3-20.

26. **Wehr E, Pilz S, Boehm BO, März W, Obermayer-Pietsch B.** Association of vitamin D status with serum androgen levels in men. *Clin Endocrinol (Oxf).* Dec 29, 2009, p. http://www.ncbi.nlm.nih.gov/pubmed/20050857.

27. **JC, Nickel.** Comparison of clinical trials with finasteride and dutasteride. *Rev Urol.* 6 Suppl 9 2004, S. 31-9 (http://www.ncbi.nlm.nih.gov/pubmed/16985923).

28. **Clark RV, Hermann DJ, Cunningham GR, Wilson TH, Morrill BB, Hobbs S.** Marked suppression of dihydrotestosterone in men with benign prostatic hyperplasia by dutasteride, a dual 5alpha-reductase inhibitor. *J Clin Endocrinol Metab.* May 89(5), 2004, pp. 2179-84 (http://www.ncbi.nlm.nih.gov/pubmed/15126539).

29. **Keam SJ, Scott LJ.** Dutasteride: a review of its use in the management of prostate disorders. *Drugs.* 68(4) 2008, pp. 463-85 (http://www.ncbi.nlm.nih.gov/pubmed/18318566).

30. **Boudou P, Soliman H, Chivot M, Villette JM, Vexiau P, Belanger A, Fiet J.** Effect of oral isotretinoin treatment on skin androgen receptor levels in male acneic patients. *J Clin Endocrinol Metab.* Apr 80(4), 1995, pp. 1158-61 (http://www.ncbi.nlm.nih.gov/pubmed/7714084).

31. **Boudou P, Chivot M, Vexiau P, Soliman H, Villette JM, Julien R, Belanger A, Fiet J.** Evidence for decreased androgen 5 alpha-reduction in skin and liver of men with severe acne after 13-cis-retinoic acid treatment. *J Clin Endocrinol Metab.* May 78(5), 1994, pp. 1064-9 (http://www.ncbi.nlm.nih.gov/pubmed/8175961).

32. **Lookingbill DP, Demers LM, Tigelaar RE, Shalita AR.** Effect of isotretinoin on serum levels of precursor and peripherally derived androgens in patients with acne. *Arch Dermatol.* Apr 124(4), 1988, pp. 540-3 (http://www.ncbi.nlm.nih.gov/pubmed/2965551).

33. **Palatsi R, Ruokonen A, Oikarinen A.** Isotretinoin, tetracycline and circulating hormones in acne. *Acta Derm Venereol.* Sep 77(5), 1997, pp. 394-6 (http://www.ncbi.nlm.nih.gov/pubmed/9298137).

34. **Rademaker M, Wallace M, Cunliffe W, Simpson NB.** Isotretinoin treatment alters steroid metabolism in women with acne. *Br J Dermatol.* Apr 124(4), 1991, pp. 361-4 (http://www.ncbi.nlm.nih.gov/pubmed/1827343).

35. **Abe M, Ito Y, Oyunzul L, Oki-Fujino T, Yamada S.** Pharmacologically relevant receptor binding characteristics and 5alpha-reductase inhibitory activity of free Fatty acids contained in saw palmetto extract. *Biol Pharm Bull.* Apr 32(4), 2009, pp. 646-50 (http://www.ncbi.nlm.nih.gov/pubmed/19336899).

36. **McPartland JM, Pruitt PL.** Benign prostatic hyperplasia treated with saw palmetto: a literature search and an experimental case study. *J Am Osteopath Assoc.* Feb 100(2), 2000, pp. 89-96.

37. **Veltri RW, Marks LS, Miller MC, Bales WD, Fan J, Macairan ML, Epstein JI, Partin AW.** Saw palmetto alters nuclear measurements reflecting DNA content in men with symptomatic BPH: evidence for a possible molecular mechanism. *Urology.* Oct 60(4), 2002, pp. 617-22 (http://www.ncbi.nlm.nih.gov/pubmed/12385921).

38. **Marks LS, Hess DL, Dorey FJ, Luz Macairan M, Cruz Santos PB, Tyler VE.** Tissue effects of saw palmetto and finasteride: use of biopsy cores for in situ quantification of prostatic androgens. *Urology.* May 57(5), 2001, pp. 999-1005 (http://www.ncbi.nlm.nih.gov/pubmed/11337315).

39. **Abe M, Ito Y, Suzuki A, Onoue S, Noguchi H, Yamada S.** Isolation and pharmacological characterization of fatty acids from saw palmetto extract. *Anal Sci.* Apr 25(4), 2009, pp. 553-7 (http://www.ncbi.nlm.nih.gov/pubmed/19359798).

40. **Avins AL, Bent S, Staccone S, Badua E, Padula A, Goldberg H, Neuhaus J, Hudes E, Shinohara K, Kane C.** A detailed safety assessment of a saw palmetto extract. *Complement Ther Med.* 16(3). Jun 2008, S. 147-54 (http://www.ncbi.nlm.nih.gov/pubmed/18534327).

41. **Crisler, Dr. John.** Speaking about Propecia at a medical conference. *Youtube.* [Online] http://www.youtube.com/watch?v=BEGCTMtlgoc.

42. **Jacobs, Dr. Alan.** Alan Jacobs md's blog. [Online] 04. 06 2010. http://blog.alanjacobsmd.com/alan-jacobs-mds-blog/2010/04/a-neuroendocrine-approach-to-finasteride-side-effects-in-men.html.

43. **Wysowski DK, Pitts M, Beitz J.** An analysis of reports of depression and suicide in patients treated with isotretinoin. *J Am Acad Dermatol.* Oct 45(4), 2001, pp. 515-9.

44. **Iehlé C, Délos S, Guirou O, Tate R, Raynaud JP, Martin PM.** Human prostatic steroid 5 alpha-reductase isoforms--a comparative study of selective inhibitors. *J Steroid Biochem Mol Biol.* Sep 1995, pp. 273-9 (http://www.revivogen.gen.tr/pdf/J%20Biochem%20Vol.%2054%20No.%2056.pdf).

45. **Uemura M, Tamura K, Chung S, Honma S, Okuyama A, Nakamura Y, Nakagawa H.** Novel 5 alpha-steroid reductase (SRD5A3, type-3) is overexpressed in hormone-refractory prostate cancer. *Cancer Sci.* Jan 99(1), 2008, pp. 81-6 (http://www.ncbi.nlm.nih.gov/pubmed/17986282).

46. **G Aumüllera, W. Eichelera, H. Renneberga, K. Adermannb, P. Viljab, W.-G. Forssmannb.** Immunocytochemical Evidence for Differential Subcellular Localization of 5alpha-Reductase Isoenzymes in Human Tissues. *Acta Anatomica.* 156 1996, pp. 241-252 (http://content.karger.com/ProdukteDB/produkte.asp?doi=10.1159/000147852).

47. **Melanie Samson, Fernand Labrie and Van Luu-The.** Sequential transformation of 4-androstenedione into dihydrotestosterone in prostate carcinoma (DU-145) cells indicates that 4-androstenedione and not testosterone is the substrate of 5a-reductase. *Horm Mol Biol Clin Invest.* 1(2) 2010, pp. 67–72 (http://www.fernandlabrie.com/files/documents/5s/c7/samsonm-2010-sequential-transformation-of.pdf).

48. **Erdemir F, Harbin A, Hellstrom WJ.** 5-alpha reductase inhibitors and erectile dysfunction: the connection. *J Sex Med.* 5(12). Dec 2008, S. 2917-24 (http://www.ncbi.nlm.nih.gov/sites/pubmed/19090946).

49. **Cunningham GR, Hirshkowitz M.** Inhibition of steroid 5 alpha-reductase with finasteride: sleep-related erections, potency, and libido in healthy men. *J Clin Endocrinol Metab.* Jun 80(6), 1995, pp. 1934-40 (http://www.ncbi.nlm.nih.gov/sites/pubmed/7775644).

50. **Uygur MC, Gür E, Arik AI, Altuğ U, Erol D.** Erectile dysfunction following treatments of benign prostatic hyperplasia: a prospective study. *Andrologia.* Feb-Mar 30(1), 1998, pp. 5-10 (http://www.ncbi.nlm.nih.gov/sites/pubmed/9567164).

51. **SR, King.** Emerging roles for neurosteroids in sexual behavior and function. *J Androl.* Sep-Oct 29(5), 2008, pp. 524-33 (http://www.ncbi.nlm.nih.gov/pubmed/18567641).

52. **Michaela Dušková, Martin Hill, Luboslav Stárka.** The influence of low dose finasteride, a type II 5α-reductase inhibitor, on circulating neuroactive steroids. *Hormone Molecular Biology and Clinical Investigation.* Jan Volume 1, Issue 2, 2010, pp. 95–102 (http://www.reference-global.com/doi/abs/10.1515/HMBCI.2010.010?journalCode=hmbci).

53. **Rahimi-Ardabili B, Pourandarjani R, Habibollahi P, Mualeki A.** Finasteride induced depression: a prospective study. *BMC Clin Pharmacol.* Oct 7, 2006, p. 6:7 (http://www.ncbi.nlm.nih.gov/sites/pubmed/17026771).

54. **Dusková M, Hill M, Hanus M, Matousková M, Stárka L.** Finasteride treatment and neuroactive steroid formation. *Prague Med Rep.* 2009, pp. 222-30 (http://www.ncbi.nlm.nih.gov/pubmed/19655698).

55. **Uygur MC, Arik AI, Altuğ U, Erol D.** Effects of the 5 alpha-reductase inhibitor finasteride on serum levels of gonadal, adrenal, and hypophyseal hormones and its clinical significance: a prospective clinical study. *Steroids.* Apr 63(4), 1998, pp. 208-13 (http://www.ncbi.nlm.nih.gov/sites/pubmed/9589555).

56. **Imperato-McGinley J, Shackleton C, Orlic S, Stoner E.** C19 and C21 5 beta/5 alpha metabolite ratios in subjects treated with the 5 alpha-reductase inhibitor finasteride: comparison of male pseudohermaphrodites with inherited 5 alpha-reductase deficiency. *J Clin Endocrinol Metab.* Mar 70(3), 1990, pp. 777-82.

57. **Bayhan G, Bahçeci M, Demirkol T, Ertem M, Yalinkaya A, Erden AC.** A comparative study of a gonadotropin-releasing hormone agonist and finasteride on idiopathic hirsutism. *Clin Exp Obstet Gynecol.* 27(3-4) 2000, pp. 203-6 (http://www.ncbi.nlm.nih.gov/sites/pubmed/11214952).

58. **Harman SM, Metter EJ, Tobin JD, Pearson J, Blackman MR.** Longitudinal effects of aging on serum total and free testosterone levels in healthy men. Baltimore Longitudinal Study of Aging. *J Clin Endocrinol Metab.* Feb 86(2), 2001, pp. 724-31 (http://www.ncbi.nlm.nih.gov/pubmed/11158037).

59. **FM., Köhn.** Diagnosis and therapy of hypogonadism in adult males. *Urologe A.* Dec 2004, S. 1563-81 (http://www.ncbi.nlm.nih.gov/pubmed/15645552).

60. **Kerrigan JR, Veldhuis JD, Rogol AD.** Androgen-receptor blockade enhances pulsatile luteinizing hormone production in late pubertal males: evidence for a hypothalamic site of physiologic androgen feedback action. *Pediatr Res.* Jan 35(1), 1994, pp. 102-6 (http://www.ncbi.nlm.nih.gov/pubmed/8134186).

61. **Shakil T, Hoque AN, Husain M, Belsham DD.** Differential regulation of gonadotropin-releasing hormone secretion and gene expression by androgen: membrane versus nuclear receptor activation. *Mol Endocrinol.* Nov 2002, pp. 2592-602 (http://www.ncbi.nlm.nih.gov/pubmed/12403848).

62. **Kawata H, Ishikura N, Watanabe M, Nishimoto A, Tsunenari T, Aoki Y.** Prolonged treatment with bicalutamide induces androgen receptor overexpression and androgen hypersensitivity. *Prostate.* May 2010, S. 745-54 (http://www.ncbi.nlm.nih.gov/pubmed/20058237).

63. **Culig Z, Steiner H, Bartsch G, Hobisch A.** Mechanisms of endocrine therapy-responsive and -unresponsive prostate tumours. *Endocr Relat Cancer.* Jun 2005, pp. 229-44 (http://www.ncbi.nlm.nih.gov/pubmed/15947099).

64. **Gregory CW, Johnson RT Jr, Mohler JL, French FS, Wilson EM.** Androgen receptor stabilization in recurrent prostate cancer is associated with hypersensitivity to low androgen. *Cancer Res.* Apr 2001, pp. 2892-8 (http://www.ncbi.nlm.nih.gov/pubmed/11306464).

65. **Fujimoto N, Miyamoto H, Mizokami A, Harada S, Nomura M, Ueta Y, Sasaguri T, Matsumoto T.** Prostate cancer cells increase androgen sensitivity by increase in nuclear androgen receptor and androgen receptor coactivators; a possible mechanism of hormone-resistance of prostate cancer cells. *Cancer Invest.* Feb 25(1), 2007, pp. 32-7 (http://www.ncbi.nlm.nih.gov/sites/pubmed/17364555).

66. **Klocker H, Culig Z, Eder IE, Nessler-Menardi C, Hobisch A, Putz T, Bartsch G, Peterziel H, Cato AC.** Mechanism of androgen receptor activation and possible implications for chemoprevention trials. *Eur Urol.* 35(5-6) 1999, S. 413-9 (http://www.ncbi.nlm.nih.gov/sites/pubmed/10325498).

67. **Chen CD, Welsbie DS, Tran C, Baek SH, Chen R, Vessella R, Rosenfeld MG, Sawyers CL.** Molecular determinants of resistance to antiandrogen therapy. *Nat Med.* Jan 2004, pp. 33-9 (http://www.ncbi.nlm.nih.gov/pubmed/14702632).

68. **Ai J, Wang Y, Dar JA, Liu J, Liu L, Nelson JB, Wang Z.** HDAC6 regulates androgen receptor hypersensitivity and nuclear localization via modulating Hsp90 acetylation in castration-resistant prostate cancer. *Mol Endocrinol.* Dec 23(12), 2009, pp. 1963-72 (http://www.ncbi.nlm.nih.gov/pubmed/19855091).

69. **Bolton EC, So AY, Chaivorapol C, Haqq CM, Li H, Yamamoto KR.** Cell- and gene-specific regulation of primary target genes by the androgen receptor. *Genes Dev.* Aug 2007, pp. 2005-17 (http://www.ncbi.nlm.nih.gov/pubmed/17699749).

70. Wikipedia. *Introduction to genetics.* [Online] 3. June 2010. http://en.wikipedia.org/wiki/Introduction_to_genetics#Genes_make_proteins.

71. **Mora GR, Mahesh VB.** Autoregulation of the androgen receptor at the translational level: testosterone induces accumulation of androgen receptor mRNA in the rat ventral prostate polyribosomes. *Steroids.* Sep 1999, pp. 587-91 (www.ncbi.nlm.nih.gov/pubmed/10503713).

72. **Wolf DA, Herzinger T, Hermeking H, Blaschke D, Hörz W.** Transcriptional and posttranscriptional regulation of human androgen receptor expression by androgen. *Mol Endocrinol.* Jul 1993, pp. 924-36 (http://www.ncbi.nlm.nih.gov/pubmed/8413317).

73. **NH., Ing.** Steroid hormones regulate gene expression posttranscriptionally by altering the stabilities of messenger RNAs. *Biol Reprod.* Jun 2005, pp. 1290-6 (http://www.ncbi.nlm.nih.gov/pubmed/15728791).

74. **Bagamasbad P, Denver RJ.** Mechanisms and significance of nuclear receptor auto- and cross-regulation. *Gen Comp Endocrinol.* Mar 23, 2010, p. Epub ahead of print (http://www.ncbi.nlm.nih.gov/pubmed/20338175).

75. **Chen PH, Tsao YP, Wang CC, Chen SL.** Nuclear receptor interaction protein, a coactivator of androgen receptors (AR), is regulated by AR and Sp1 to feed forward and activate its own gene expression through AR protein stability. *Nucleic Acids Res.* Jan 2008, S. 51-66 (http://www.ncbi.nlm.nih.gov/pubmed/17984071).

76. **Zeiser S, Franz U, Müller J, Liebscher V.** Hybrid modeling of noise reduction by a negatively autoregulated system. *Bull Math Biol.* May 2009, pp. 1006-24 (http://www.ncbi.nlm.nih.gov/pubmed/19159985).

77. **Auboeuf D, Batsché E, Dutertre M, Muchardt C, O'Malley BW.** Coregulators: transducing signal from transcription to alternative splicing. *Trends Endocrinol Metab.* Apr 18(3), 2007, pp. 122-9 (http://www.ncbi.nlm.nih.gov/sites/pubmed/17320409).

78. **Rev., Endocr.** Androgen receptor (AR) coregulators: a diversity of functions converging on and regulating the AR transcriptional complex. *Heemers HV, Tindall DJ.* Dec 28(7), 2007, pp. 778-808 (http://www.ncbi.nlm.nih.gov/sites/pubmed/17940184).

79. **Babur O, Demir E, Gönen M, Sander C, Dogrusoz U.** Discovering modulators of gene expression. *Nucleic Acids Res.* May 13, 2010, p. [Epub ahead of print] (http://www.ncbi.nlm.nih.gov/pubmed/20466809).

80. **IJ., McEwan.** Molecular mechanisms of androgen receptor-mediated gene regulation: structure-function analysis of the AF-1 domain. *Endocr Relat Cancer.* Jun 2004, S. 281-93 (http://www.ncbi.nlm.nih.gov/pubmed/15163303).

81. **Rosenfeld MG, Lunyak VV, Glass CK.** Sensors and signals: a coactivator/corepressor/epigenetic code for integrating signal-dependent programs of transcriptional response. *Genes Dev.* Jun 2006, pp. 1405-28 (http://www.ncbi.nlm.nih.gov/pubmed/16751179).

82. Wikipedia. *Regulation of gene expression.* [Online] May 31, 2010. http://en.wikipedia.org/wiki/Gene_regulation.

83. **Heemers HV, Tindall DJ.** Androgen receptor (AR) coregulators: a diversity of functions converging on and regulating the AR transcriptional complex. *Endocr Rev.* Dec 28(7), 2007, pp. 778-808 (http://www.ncbi.nlm.nih.gov/sites/pubmed/17940184).

84. **Kininis M, Kraus WL.** A global view of transcriptional regulation by nuclear receptors: gene expression, factor localization, and DNA sequence analysis. *Nucl Recept Signal.* Feb 15, 2008, p. 6:e005 (http://www.ncbi.nlm.nih.gov/sites/pubmed/18301785).

85. **Csoka AB, Szyf M.** Epigenetic side-effects of common pharmaceuticals: a potential new field in medicine and pharmacology. *Med Hypotheses.* Nov 2009, S. 770-80 (http://www.ncbi.nlm.nih.gov/pubmed/19501473).

86. **Jarrard DF, Kinoshita H, Shi Y, Sandefur C, Hoff D, Meisner LF, Chang C, Herman JG, Isaacs WB, Nassif N.** Methylation of the androgen receptor promoter CpG island is associated with loss of androgen receptor expression in prostate cancer cells. *Cancer Res.* Dec, Dec 1;58(23), 1998, Vol. 1;58(23), pp. 5310-4 (http://www.ncbi.nlm.nih.gov/sites/pubmed/9850055).

87. **Liu ZJ, Wang G, Cai Y, Gu SZ, Zhang XB, Liu L, Gao X.** Androgen receptor CpG island methylation status in human leukemia cancer cells. *Cancer Invest.* Feb 27(2), 2009, pp. 156-62 (http://www.ncbi.nlm.nih.gov/sites/pubmed/19235587).

88. **Gmuender H, Kuratli K, Di Padova K, Gray CP, Keck W, Evers S.** Gene expression changes triggered by exposure of Haemophilus influenzae to novobiocin or ciprofloxacin: combined transcription and translation analysis. *Genome Res.* Jan 11(1), 2001, pp. 28-42 (http://www.ncbi.nlm.nih.gov/sites/pubmed/11156613).

89. Wikipedia. *Epigenetics.* [Online] May 27, 2010. http://en.wikipedia.org/wiki/Epigenetic_regulation.

90. propeciahelp.com. *Merck Website Archive.* [Online] http://www.propecia.com/finasteride/propecia/consumer/about-propecia-finasteride/possible_side_effects.jsp?WT.svl=2..

91. **Frye, Stephen V.** Inhibitors of 5a-Reductase. *Current Pharmaceutical Design.* Feb 1996, pp. 59-84 (http://books.google.ca/books?id=IYn4Va7wtAoC&pg=PA69&dq=finasteride&lr=&as_brr=3#v=onepage&q=&f=true).

92. **Jin Y, Penning TM.** Steroid 5alpha-reductases and 3alpha-hydroxysteroid dehydrogenases: key enzymes in androgen metabolism. *Best Pract Res Clin Endocrinol Metab.* Mar 15(1), 2001, pp. 79-94 (http://www.ncbi.nlm.nih.gov/pubmed/11469812).

93. **Zouboulis CC, Degitz K.** Androgen action on human skin -- from basic research to clinical significance. *Exp Dermatol.* 13 Suppl 4 2004, pp. 5-10 (http://www.ncbi.nlm.nih.gov/sites/pubmed/15507105).

94. **Lobo RA, Paul WL, Gentzschein E, Serafini PC, Catalino JA, Paulson RJ, Horton R.** Production of 3 alpha-androstanediol glucuronide in human genital skin. *J Clin Endocrinol Metab.* Oct 65(4), 1987, pp. 711-4 (http://www.ncbi.nlm.nih.gov/sites/pubmed/3654916).

95. **Rittmaster RS, Zwicker H, Thompson DL, Konok G, Norman RW.** Androstanediol glucuronide production in human liver, prostate, and skin. Evidence for the importance of the liver in 5 alpha-reduced androgen metabolism. *J Clin Endocrinol Metab.* Apr 76(4), 1993, pp. 977-82 (http://www.ncbi.nlm.nih.gov/pubmed/8473413).

96. **Guess HA, Friedman GD, Sadler MC, Stanczyk FZ, Vogelman JH, Imperato-McGinley J, Lobo RA, Orentreich N.** 5 alpha-reductase activity and prostate cancer: a case-control study using stored sera. *Cancer Epidemiol Biomarkers Prev.* Jan 6(1), 1997, pp. 21-4 (http://www.ncbi.nlm.nih.gov/pubmed/8993793).

97. **R, Horton.** Dihydrotestosterone is a peripheral paracrine hormone. *J Androl.* Jan-Feb 13(1), 1992, pp. 23-7 (http://www.ncbi.nlm.nih.gov/pubmed/1551803).

98. **Brian E. Henderson, Bruce A. J. Ponder, Ronald Keith Ross.** *Hormones, genes, and cancer (p. 23).* New-York : Oxford University Press, 2003.

99. **Xiong G, Maser E.** Regulation of the steroid-inducible 3alpha-hydroxysteroid dehydrogenase/carbonyl reductase gene in Comamonas testosteroni. *J Biol Chem.* Mar 276(13), 2001, pp. 9961-70.

100. **Penning TM, Jin Y, Rizner TL, Bauman DR.** Pre-receptor regulation of the androgen receptor. *Mol Cell Endocrinol.* 16. Jan 2008, S. 281(1-2):1-8 (http://www.ncbi.nlm.nih.gov/pubmed/18060684).

101. **Lee HJ, Mowszowicz I, Chang C.** The first detection of complete androgen insensitivity with no mutation in the coding sequence of the androgen receptor gene. *Front Biosci.* Jul 1, 1996, pp. 1:a34-8 (http://www.ncbi.nlm.nih.gov/sites/pubmed/9159192).

102. **Nahoul K, Adeline J, Roger M.** Age-related changes in plasma androstanediol glucuronide in men. *Pathol Biol (Paris).* Nov 38(9), 1990, pp. 928-34 (http://www.ncbi.nlm.nih.gov/pubmed/2277725).

103. **F, Labrie.** Combined androgen blockade: It's unique efficacy for the treatment of localized prostate cancer. *Principles & Practice of Oncology.* Vol. 13 Nr. 2, 1999, pp. 1-9 (http://www.fernandlabrie.com/files/documents/el/12/labrief-1999-combined-androgen-blockade.pdf).

104. **Do Rego JL, Seong JY, Burel D, Leprince J, Luu-The V, Tsutsui K, Tonon MC, Pelletier G, Vaudry H.** Neurosteroid biosynthesis: enzymatic pathways and neuroendocrine regulation by neurotransmitters and neuropeptides. *Front Neuroendocrinol.* Aug 30(3), 2009, pp. 259-301 (http://www.ncbi.nlm.nih.gov/sites/pubmed/19505496).

105. **Uzunova V, Sampson L, Uzunov DP.** Relevance of endogenous 3alpha-reduced neurosteroids to depression and antidepressant action. *Psychopharmacology (Berl).* Jun 186(3), 2006, pp. 351-61 (http://www.ncbi.nlm.nih.gov/sites/pubmed/16249906).

106. **Marx CE, Trost WT, Shampine LJ, Stevens RD, Hulette CM, Steffens DC, Ervin JF, Butterfield MI, Blazer DG, Massing MW, Lieberman JA.** The neurosteroid allopregnanolone is reduced in prefrontal cortex in Alzheimer's disease. *Biol Psychiatry.* Dec 15;60(12), 2006, pp. 1287-94 (http://www.ncbi.nlm.nih.gov/pubmed/16997284).

107. **FDA.** FDA. *Clinical Pharmacology Biopharmaceutics Review(s) of Finasteride.* [Online] http://www.accessdata.fda.gov/drugsatfda_docs/nda/97/20788_PROPECIA%20TABLETS,%201MG_BIOPHARMR.PDF.

108. **Eikenberry SE, Nagy JD, Kuang Y.** The evolutionary impact of androgen levels on prostate cancer in a multi-scale mathematical model. *Biol Direct.* Apr 2010, S. [Epub ahead of print] (http://www.ncbi.nlm.nih.gov/pubmed/20406442).

109. **Corradi LS, Góes RM, Vilamaior PS, Taboga SR.** Increased androgen receptor and remodeling in the prostatic stroma after the inhibition of 5-alpha reductase and aromatase in gerbil ventral prostate. *Microsc Res Tech.* Dec 72(12), 2009, pp. 939-50 (http://www.ncbi.nlm.nih.gov/pubmed/19484778).

110. **Miyamoto H, Rahman MM, Chang C.** Molecular basis for the antiandrogen withdrawal syndrome. *J Cell Biochem.* Jan 2004, pp. 3-12.

111. **Lau YK, Chadha MK, Litwin A, Trump DL.** A dramatic, objective antiandrogen withdrawal response: case report and review of the literature. *J Hematol Oncol.* 5. Nov 2008, S. 1-21 (http://www.ncbi.nlm.nih.gov/pubmed/18986533).

112. **Montgomery BT, Young CY, Bilhartz DL, Andrews PE, Prescott JL, Thompson NF, Tindall DJ.** Hormonal regulation of prostate-specific antigen (PSA) glycoprotein in the human prostatic adenocarcinoma cell line, LNCaP. *Prostate.* 21(1) 1992, pp. 63-73 (http://www.ncbi.nlm.nih.gov/pubmed/1379363).