Review

Novel Antiandrogenic 5α-Reductase Inhibitors with Antioxidant Activity

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Abstract

Background. Steroids are biomolecules with a basic structure made up of cyclopentanoperhydrophenanthrene. Two steroids derived from cholesterol are testosterone, as natural androgen, and progesterone, as natural antiandrogen. Reactive oxygen species (ROS) may act as a metabolic signal-mediating response to changes in glucose, and hormones. Antiandrogens can be prescribed to treat an array of diseases and disorders as Gender dysphoria. In men, antiandrogens are most frequently used to treat prostate cancer and hyperplasia. Methods.The present study has the aim of pharmacological evaluation of several new steroid derivatives that were prepared from the commercially available 16-dehydropregnenolone acetate. The biological activity of the new steroidal derivatives was determined. The neuroprotection effect of the steroids was demostrated using the biomarkers of oxidative stress on male rat brain and liver with hypoglycemia induced. Enzyme kinetics was demostrated by the inhibition of 5α-reductase enzyme on myelin of brain. Conclusion. This study suggest that steroid 12 derivatives with an electrophilic center can interact more efficiently with the 5α-reductase enzyme and then induce neuroprotection in hypoglycemia animal model. Further research with clinically meaningful endpoints is needed to optimize the use of antiandrogens in these hormonal therapies.

Keywords: antiandrogen agent; 5α-reductase inhibitors; myelin; antioxidant; neuroprotection

Introduction

An anti-androgen is a compound that blocks the androgen receptors. 5 alpha reductase inhibitors cannot be considered as anti-androgens: e.g., even blocking 5aR would leave testosterone available to act on AR. Steroids are biomolecules with a basic structure made up of cyclopentanoperhydrophenanthrene [1]. Two steroids derived from cholesterol are testosterone (Figure 1), as natural androgen, and progesterone (Figure 2), as natural antiandrogen [2]. Tradicionally finasteride and dutasteride, the two synthetic inhibitors used in clinics, as well, epigallo catechin gallate as a natural inhibitor of 5aR.

Figure 1. Testosterone.

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Figure 2. Progesterone.

Antiandrogens or androgen antagonists alter the androgen pathway by blocking the appropriate receptors, competing for binding sites on the cell surface or affecting androgen production [3]. Endogenous sex hormones may differentially modulate glycemic status and it is associated with higher risk of type 2 diabetes [4]. However, reactive oxygen species (ROS) may act as a metabolic signal-mediating responses to changes in glucose and hormones [5]. Low levels of circulating androgens should be considered as a significant risk factor for the development of neurodegenerative disorders [6]. In this regard, numerous neurotransmitters have been implicated in the pathogenesis of these disorders, with dopamine and serotonin playing a crucial role in the neural reward pathways [7].

Antiandrogens can be prescribed to treat an array of diseases and disorders as gender dysphoria. In men, antiandrogens are most frequently used to treat prostate hyperplasia and cancer [8]. In many tissues sulfonated steroids exceed the concentration of free steroids. Recently, these sulfonated steroids were also shown to fulfill important physiological functions. It was suggested that cholesterol sulfate (CS) is converted by CYP11A1 to pregnenolone sulfate (PregS), which is metabolized to 17OH-PregS; thus, strengthening the potential physiological meaning of a pathway for sulfonated steroids [9].

Antiandrogens present in the environment have become a topic of concern. Certain plant species have also been found to produce antiandrogens as dehydropregnenolone acetate (figure 3), and inhibit circulating androgens by blocking androgen receptors, suppressing androgen synthesis, or acting in both ways [10].

Figure 3. 16-Dehydropregnenolone Acetate.

Target Cell Action

The most common antiandrogens are androgen receptor (AR) antagonists, which act on the target cell level and competitively bind to androgen receptors [11]. Antiandrogenic drugs are used for hormone therapy. This therapy is called androgen deprivation therapy (ADT). The main goal of ADT is to produce a state of competition between them and the circulating androgens for binding sites on prostate cell receptors. In this way, they can inhibit prostate cancer growth and promote their apoptosis [12]. Antiandrogen monotherapy generally causes fewer side effects in males; however, they are less effective in blocking androgen when compared with combined therapies. Monotherapy is often preferred by men as it is less likely to diminish libido than combined therapies [13]. In fact, antiandrogens are 5α-reductase inhibitors and prevent the conversion of testosterone to DHT [14], by directly binding on hydrogen in C-5 (Figure 4).

Figure 4. 5α-reductase inhibits the conversion of testosterone to DHT.

DHT is 3-5 times more potent than testosterone or other androgens. They are unique because they do not counteract the effects or production of other androgens other than DHT. Dihydrotestosterone is necessary for development of both external male sex organs and the prostate [15].

However, 5α-reductase enzyme has several isoforms and is expressed in various tissues as well as in the epithelium and myelin [16]. Therefore, the circulating and intraprostatic DHT could be further reduced by a more effective dual 5-alpha-reductase inhibitor, which would be efficacious in the treatment of benign prostate hyperplasia and other DHT-related disorders, as gender dysphoria. Antiandrogens act by various mechanisms to decrease the production or effects of testosterone, but it is unclear which antiandrogen is most effective at feminization [17], although, Spironolactone is commonly used in feminizing hormone therapy to achieve the goal of female range testosterone level [18].

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A peptide antagonist interrupts androgen receptor protein interaction from the surface of the receptor. This approach is mechanism-based and has greater potential for blocking receptor activity than the traditional ligand-receptor binding approach [19].

Developing Novel Antiandrogen

On the other hand, some studies suggest that the modification of steroid B-ring or D-ring and lateral chain play an important role for the hormonal therapy [20]. There is much interest in developing chemical novel antiandrogen drugs that may help to prevent or ameliorate these clinical disorders; thus, suggesting the introduction of aromatic or aliphatic structures in steroid B-ring and D-ring [21].

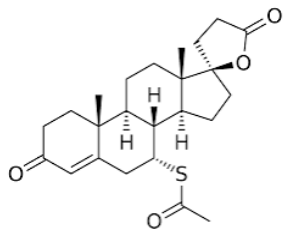
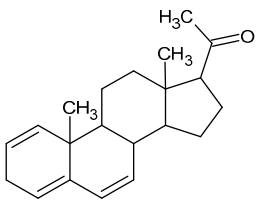
Methods

The present study has the aim of pharmacological evaluation of several new steroid derivatives that were prepared from the commercially available 16-dehydropregnenolone acetate. The biological activity of the new steroidal derivatives was determined. The neuroprotection effect of the steroids was demostrated using the biomarkers of oxidative stress on male rat brain and liver with hypoglycemia induced.

Results and Discusion

Enzyme kinetics was demostrated by the inhibition of 5α-reductase enzyme on myelin of brain. This study suggest that steroid 12 derivatives with an electrophilic center can interact more efficiently with the 5α-reductase enzyme and then induce neuroprotection in hypoglycemia animal model. Further research with clinically meaningful endpoints is needed to optimize the use of antiandrogens in these hormonal therapies.

Table 1. Novel synthetic steroidal structures and *in vitro* result assessments.

Steroid structure	5α-Reductase ± SD	Km ± SD	Vmax ± SD
 Spironolactone $C_{24}H_{32}O_4S$ PM 416.57 g/mol	0.305 ± 0.006	0.0309 ± 0.006	0.304 ± 0.007
 1,4,6-tripregnen-20-one	0.739 ± 0.01	0.378 ± 0.03	0.767 ± 0.01

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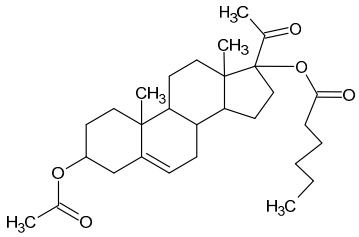
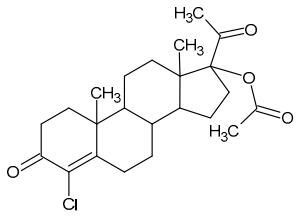
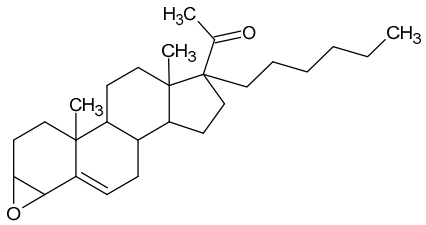
$C_{21}H_{28}O$ PM=296 g/mol			
 3β-acetoxy-5-pregnen-17α-hexanoiloxy-20-one $C_{29}H_{44}O_5$ PM=472 g/mol	0.615 ± 0.16	0.288 ± 0.27	0.607 ± 0.16
 4-chloro-5-pregnen-17α-etiloixy-3,20-dione $C_{23}H_{31}ClO_4$ PM=406.5 g/mol	0.369 ± 0.002	0.010 ± 0.008	0.370 ± 0.003
 3α.4α-epoxy-17α-hexyl-5-pregnen-20-one $C_{27}H_{42}O_2$ PM=398 g/mol	0.406 ± 0.008	0.063 ± 0.009	0.409 ± 0.006

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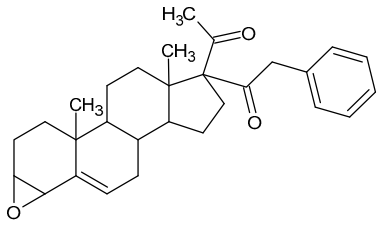
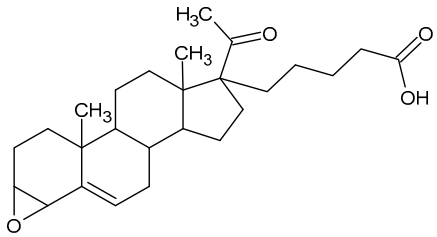
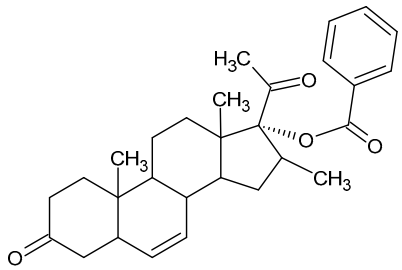
 3α,4α-epoxy-17α-phenylacetyl-5-pregnen-20-one $C_{29}H_{36}O_3$ PM=432 g/mol	0.434 ± 0.007	0.075 ± 0.01	0.434 ± 0.007
 3α,4α-epoxy-17α-valeryl-5-pregnen-20-one $C_{26}H_{38}O_4$ PM=414 g/mol	0.390 ± 0.008	0.020 ± 0.01	0.392 ± 0.008
 16α-methyl-6-pregnen-17α-benzoiloxy-20-one $C_{29}H_{36}O_4$ PM=448 g/mol	0.381 ± 0.01	0.142 ± 0.014	0.382 ± 0.01

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The complete data of this study showed very clearly that all compounds are good inhibitors for the 5alpha-reductase enzyme. Probing the efficacy of these novel steroids with respect to spironolactone *in vitro* assay, appears that they would be promising compounds for future hormonal therapy in patients

Conclusion

The future of antiandrogenic steroid drugs is believed to be antagonists due mainly to cyclopentanoperhydrophenanthrene structure. Androgen receptor antagonists act in an alternative manner and this may be one of the mechanisms underlying the benefits of these drugs. In addition, this response between antiandrogens and clinical disorders is expected in adult people. In general, the analysis of novel synthetic steroid suggests that it is favorable to insert aromatic or ester-aliphatic groups in ring D, chloride in ring B, and aliphatic group with carbonyl in ring A. The aromatic groups inserted in ring D, activated 5α-reductase enzyme. These antioxidant molecules provide the scientific basis to design clinical trials aimed at reducing the oxidative stress, and probably the CNS changes elicited by hormonal therapy in patients. This is important when we consider that it is still unclear which antiandrogen is most effective at achieving feminization. However, there are insufficient evidence to determine the efficacy or safety of hormonal treatment approaches for transgender women in transition, or prostate cancer. Hence, further research with clinically meaningful endpoints is needed to optimize the use of antiandrogens in these hormonal therapies.

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Availability of Data: **Any data** used in this study **are available** on request to the **correspondence author.**

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Conflict of Interest: The authors declare that they have no conflict of interest.

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