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The impact of nandrolone decanoate and growth hormone on biosynthesis of steroids in rats

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ABSTRACT

Growth hormone (GH) and anabolic androgenic steroids (AAS) are commonly used in sports communities. Several studies have suggested an association between GH and AAS. We have investigated the impact of GH in rats treated with nandrolone decanoate (ND). Male Wistar rats received ND (15 mg/kg) every third day during three weeks and were subsequently treated with recombinant human GH (1.0 IU/kg) for ten consecutive days. Plasma samples were collected and peripheral organs (i.e. heart, liver, testis and thymus) were dissected and weighed. Concentration of thirteen endogenous steroids was measured in the rat plasma samples using high specificity LC–MS/MS methods. Seven steroids were detected and quantified, and concentrations of estrone, testosterone, and androstenedione were significantly different among the groups, while concentrations of pregnenolone, DHEA, 17-hydroxyprogesterone and corticosterone were not altered. Administration of rhGH alone altered the plasma steroid distribution, and the results demonstrated significantly increased concentrations of plasma estrone as well as decreased concentrations of testosterone and androstenedione in the ND-treated rats. Administration of rhGH to ND-pretreated rats did not reverse the alteration of the steroid distribution induced by ND. Administration of ND decreased the weight of the thymus, and addition of rhGH did not reverse this reduction. However, rhGH administration induced an enlargement of thymus. Taken together, the plasma steroid profile differed in the four groups, i.e. control, AAS, rhGH and the combination of AAS and rhGH treatment.

1. Introduction

Growth hormone (GH) is an endocrine polypeptide with a wide variety of biological functions e.g. in processes associated with metabolism and growth. In addition, GH plays an important role in the function of the central nervous system (CNS). GH and insulin-like growth factor 1 (IGF1), a peptide mediating many of the effects of GH, have been shown to increase neurogenesis, be involved in neuroprotection and counteract opioid-induced apoptosis in cells from mouse hippocampus [1,2]. In addition, it was shown that GH is involved in cognition and memory functions [3]. Studies suggest that GH affects function of the hypothalamic–pituitary–adrenal (HPA) axis [4] and influence the adrenal androgen secretion [5]. GH signaling is also believed to affect the hypothalamic–pituitary–gonadal (HPG) axis (for review see [6]). However, to the best of our knowledge, the influence of GH on concentrations of plasma steroids in intact rodents has not been examined.

GH is known to be abused in sports communities, and is recognized as an anabolic agent. Another group of substances, which are abused for their anabolic attributes are the anabolic androgenic steroids (AAS), among which one of the most frequently abused is nandrolone decanoate (ND). ND has a structure very similar to testosterone, the main endogenous androgen biosynthesized in the testis (Fig. 1). Today the AAS abuse has spread beyond the world of sports and is commonly used by adolescents and young adults [7]. AAS effect many functions in the body, including the CNS, and are known to cause a range of adverse effects, including gynecomastia, increased risk of cardiovascular and hepatic disease, aggression, depression, and impaired cognitive functions [8,9].

Users of AAS often combine the steroid intake with other drugs of abuse and various pharmaceuticals, and combination with GH is common [10]. It is known that GH and IGF1 effects biosynthesis of sex steroids [6,11] and that gonadal steroids to some extent regulate GH secretion [12]. A recent study demonstrated that treatment

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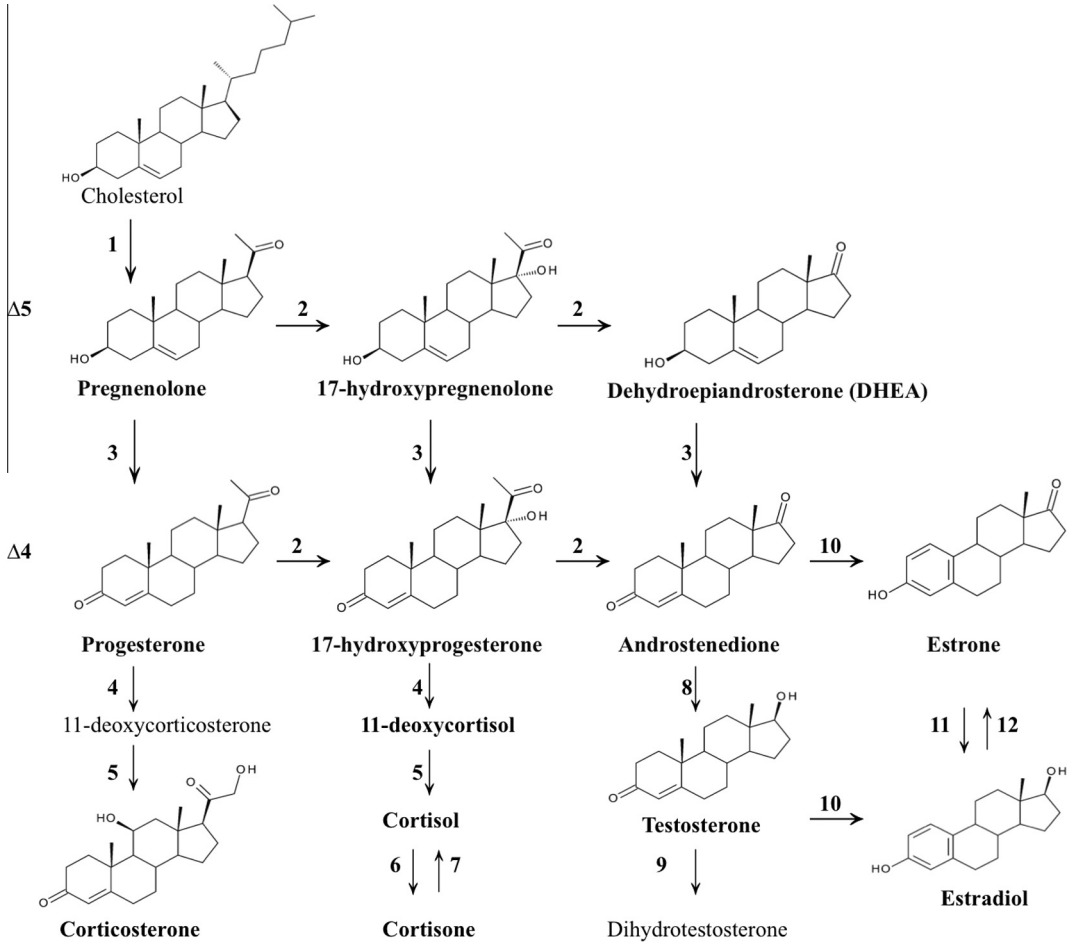

Fig. 1. Pathway of steroid biosynthesis. The most common pathway of steroidogenesis in rodents is believed to be the Δ4-pathway, involving progesterone, 17-hydroxyprogesterone and androstenedione [57], whereas the Δ5-pathway is more common in humans [58]. (1) Cholesterol side chain cleavage enzyme (CYP11A), (2) Cytochrome P450 17α-hydroxylase, 17,20 lyase (CYP17A1), (3) 3β-hydroxysteroid dehydrogenase 1 (3β-HSD1), (4) 21α-hydroxylase (CYP21), (5) 11β-hydroxylase 1 (CYP11B1), (6) 11β-hydroxysteroid dehydrogenase 2 (11β-HSD2), (7) 11β-hydroxysteroid dehydrogenase 1 (11β-HSD1), (8) 17β-hydroxysteroid dehydrogenase 3 (17β-HSD3), (9) 5α-reductase, (10) aromatase (CYP19), (11) 17β-hydroxysteroid dehydrogenase 1 (17β-HSD1), (12) 17β-hydroxysteroid dehydrogenase 2 (17β-HSD2). Steroids in bold were measured in the present study.

with ND lead to decreased plasma concentrations of IGF1 [13], however, the mechanism behind the interaction between the two systems is not known. It is well known that AAS in humans induce a negative feedback function on the HPG-axis causing a decrease of concentrations of endogenous androgens [14–16]. GH is less studied in this context, but was shown to be present in the testis and play an important role for steroidogenesis and gametogenesis [17,18]. Kanzaki et al., showed that GH stimulates steroidogenesis in rat Leydig cells [19]. Notably, there have been no reports on combined effect of administration of ND and GH on the plasma steroid profile in intact animals. As mentioned above, both substances target functions of several peripheral organs, the effect of concurrent administration of ND and GH on organ weight have however not previously been studied in rats.

The aims of this study were to examine the effects of GH and AAS on biosynthesis of endogenous steroids, and to investigate the effect of a supraphysiologic three-week long intake of ND, and a subsequent ten day long treatment with rhGH in intact rats. Furthermore, the impact of AAS and GH treatment on weight of liver, thymus, testis and heart was investigated. Concentration of thirteen steroids was measured in rat plasma using validated liquid chromatography tandem mass spectrometry (LC–MS/MS) methods.

2. Experimental

2.1. Animals

In this study, 48 male Wistar rats obtained from Taconic Farms (Ejeby, Denmark) were allowed to adapt to the laboratory environment for approximately two weeks. The rats were ten weeks old, with a body weight of 316.1 ± 2.5 g, at the start of the experiment. All rats were group-housed (three in each cage) with free access to water and food and kept in an air-conditioned room with controlled temperature (22–24 °C) and humidity (50–60%). The rats were kept under a reversed 12-h dark/light cycle with lights off at 7 a.m. The animal experiments were performed under a protocol approved by the Uppsala Animal Ethical Committee. Data from these rats are also included in an earlier publication on effects of GH and ND on spatial memory, for further details see Ref. [13].

2.2. Treatment

Subcutaneous (s.c.) injections with ND (Deca-Durabol, Organon, Oss, Netherlands), 15 mg/kg, or peanut oil (Apoteket AB, Umeå, Sweden) was performed every third day during three weeks, starting from day 1 to 21 of the experiment. During days 22–31, the rats