A commentary on the origins of 11-ketotestosterone

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Over the past decade, 11-oxygenated androgens, and in particular 11-ketotestosterone (11KT), have been recognised as important contributors to the androgen pool in human physiology. Indeed, 11KT is now widely accepted as a physiologically relevant androgen in humans with the ability to bind to and activate the human androgen receptor in a manner similar to testosterone (1, 2, 3). Notably, 11KT has been implicated as a role player in several disease states, including congenital adrenal hyperplasia, polycystic ovarian syndrome and castration-resistant prostate cancer (1, 2).

Although 11KT is considered an adrenal-derived androgen, its biosynthesis is dependent on the adrenal as well as peripheral tissues, with only minor direct biosynthesis in the adrenal cortex. Kitamura et al. (4) report on a rare case of adrenocortical adenoma that resulted in mild autonomous cortisol secretion and hyperandrogenemia due to elevated 11-oxygenated androgen biosynthesis, resulting in excessive circulating 11KT levels. Immunohistochemical analysis of the resected tumour illustrate the key roles played by the enzymes 11β-hydroxysteroid dehydrogenase type 2 (HSD11B2) and aldo-keto reductase 1C3 (AKR1C3) in the biosynthesis of 11KT, and provide insights into why 11KT owes its origins to the peripheral conversion of adrenal derived 11β-hydroxyandrostenedione in healthy individuals.

The first step in the biosynthesis of 11-oxygenated androgens is the 11β-hydroxylation of androstenedione or testosterone by the adrenal-specific enzyme cytochrome P450 11β-hydroxylase (CYP11B1). Both products, 11β-hydroxyandrostenedione (11OHA4) and 11β-hydroxytestosterone (11OHT), can subsequently be converted to 11KT (Fig. 1). The biosynthesis of 11KT from 11OHA4 requires the HSD11B2-catalysed conversion of 11OHA4 to 11-ketoandrostenedione (11KA4) followed by the AKR1C3-mediated conversion of 11KA4 to 11KT, whereas the conversion of 11OHT to 11KT requires only HSD11B2. Given that only a single step is required and that the circulating levels of 11OHT and 11KT correlate strongly, it is often suggested that circulating 11KT primarily owes its origins to adrenal-derived 11OHT. However, a closer inspection of the expression levels and characteristics of the enzymes involved reveals that this is likely not the case.

The adrenocortical adenoma reported by Kitamura et al. (4) expressed all enzymes required for the biosynthesis of 11OHA4 and 11OHT. In addition, immunohistochemical analysis of the resected tumour revealed the extensive expression of both HSD11B2 and AKR1C3, thus allowing for the intratumoural biosynthesis of 11KT from 11OHA4 or 11OHT. A key enzyme in both routes is AKR1C3. Although not required for the conversion of 11OHT to 11KT, AKR1C3 is essential for the conversion of androstenedione to testosterone prior to the 11β-hydroxylation of testosterone by CYP11B1. This testosterone must be produced at the site of CYP11B1 expression in order to generate 11OHT (5). Moreover, 11OHT is not generated directly from 11OHA4 as 11OHA4 is not a substrate for AKR1C3 (6). When comparing the contributions of the two possible routes to 11KT (Fig. 1), one must consider that the AKR1C3-catalysed conversion of androstenedione to testosterone is relatively inefficient (6, 7). This was reflected by the >7-fold higher levels of androstenedione (11.6 nM) when compared to testosterone (1.6 nM), as well as the observation that the concentration of 11OHT (4.0 nM) was the lowest of the 11-oxygenated androgens measured by a >2-fold margin. On the other hand, 11OHA4 was by far the most abundant 11-oxygenated androgen (46.1 nM) produced, demonstrating substantially more 11β-hydroxylation of
Origins of 11-ketotestosterone

K-H Storbeck

When taking into account that the same enzymes are involved in the biosynthesis of 11KT in healthy individuals, the same concepts must apply. The only difference is that not all the enzymes are highly expressed in the adrenal cortex and as such the biosynthesis of 11KT is dependent on the expression of key enzymes in peripheral tissues. Although AKR1C3 is moderately expressed in the zona reticularis of the healthy adrenal cortex, the inefficient conversion of androstenedione to testosterone limits the levels of 11OHT produced (6, 8, 9). Unsurprisingly, the amount of 11OHT produced by the adrenal cortex pales in comparison to that of the 11OHA4 produced, with levels of 11OHA4 reported to be >300 fold higher than that of 11OHT, thus providing a substantially larger substrate pool for peripheral 11KT biosynthesis (8). Moreover, while 11OHA4 is the most abundant 11-oxygenated androgen in the adrenal vein with levels reported to be 84-fold higher than that of 11KA4, 11OHT and 11KT combined, 11OHA4 levels are only approximately 2-fold that of the combination of 11KA4, 11OHT and 11KT in peripheral circulation, thus suggesting extensive peripheral conversion (1, 8).

Once in circulation, 11OHA4 is undoubtedly converted to 11KA4 by HSD11B2, which is highly expressed in mineralocorticoid target tissues such as the kidney (10). The resulting 11KA4 is in turn a substrate for AKR1C3 expressed in peripheral tissues such as adipose yielding 11KT (6, 11). Peripheral expression of AKR1C3 plays an important role in the peripheral activation of androgen precursors, especially in women (12). Therefore, peripheral conversion of adrenal-derived 11OHA4 most likely makes the largest contribution to 11KT in circulation with a smaller contribution due to the conversion of adrenal-derived 11OHT to 11KT by peripherally expressed HSD11B2.

Notably, 11β-hydroxysteroid dehydrogenase type 1 (HSD11B1) expressed in peripheral tissues, such as adipose, catalyses the conversion of 11KA4 and 11KT to 11OHA4 and 11OHT, respectively (13). Therefore, the peripheral metabolism of 11KT must also contribute to the circulating 11OHT pool. Much like with the glucocorticoids cortisol and cortisone, for which there is a constant

Figure 1
Biosynthesis of 11-ketotestosterone (11KT). The preferred route of 11KT biosynthesis in the 11-oxygenated androgen-producing adrenocortical adenoma reported by Kitamura et al. (4) (A) and in healthy individuals (B) is shown by the bold arrows. 11KT biosynthesis in healthy individuals includes enzymes expressed in multiple peripheral tissues with the result that there is a constant interconversion of 11KT and other 11-oxygenated androgens as indicated by the dashed arrows. It should be noted that peripheral metabolism would have contributed to the elevated serum levels of 11KT measured by Kitamura et al. (4) prior to the resection of the adrenocortical adenoma.
interconversion due to the expression of HSD11B2 and HSD11B1 in different peripheral tissues, one can imagine that there is a constant interconversion of the 11β-hydroxy and 11-keto forms of the 11-oxygenated androgens which would account for the strong correlations observed for the levels of 11KT and 11OHT as well as for 11KA4 and 11OHA4, respectively (14).

There are, however, pathological conditions in which the direct 11β-hydroxylation of testosterone plays a bigger role. In 21-hydroxylase deficiency (21OHD), steroid precursors are shunted towards androgen biosynthesis in the adrenal cortex with the result that adrenal testosterone biosynthesis and subsequent 11OHT production are significantly increased (14). However, 11OHA4 and 11KA4 levels are also increased in 21OHD and thus would still contribute to 11KT biosynthesis through the peripheral AKR1C3-catalysed conversion of 11KA4 to 11KT. Conversely, in testicular adrenal rest tumours (TARTs), 11OHT and 11KT are the most abundant 11-oxygenated androgens released into the spermatic vein, which can be accounted for by the unusual co-expression of testicular and adrenal enzymes, thus allowing for testosterone biosynthesis and subsequent 11β-hydroxylation to 11OHT (15, 16).

Taken together, it is clear that the biosynthesis and metabolism of 11-oxygenated androgens is complex. Analysis of the enzymes and their substrate preferences clearly indicates that two routes occur in healthy individuals, but that the majority of 11KT in circulation is in all probability produced from peripheral conversion of adrenal-derived 11OHA4. While adrenal-derived 11OHT must feed into this pathway, current evidence suggests that this contribution is low under normal conditions but makes a larger contribution in pathological conditions such as in 21OHD and TARTs.

Declaration of interest
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References
3 Pretorius E, Africander DJ, Vlok M, Perkins MS, Quanson JL & Storbeck KH. 11-Ketotestosterone and 11-ketohydrotestosterone in castration resistant prostate cancer: potent androgens which can no longer be ignored. PLoS ONE 2016 11 e0159667. (https://doi.org/10.1371/journal.pone.0159667)
14 Turcu AF, Nanba AT, Chomic R, Upadhyay SK, Giordano TJ, Shields JJ, Merke DP, Rainey WE & Auchus RJ. Adrenal-derived 11-oxygenated 19-carbon steroids are the dominant androgens in classic


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