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Review

Polycystic ovary syndrome: evidence for a primary disorder of ovarian steroidogenesis ☆

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Abstract

Polycystic ovary syndrome (PCOS) is a very common endocrinopathy of uncertain aetiology in which the most consistent biochemical abnormality is hypersecretion of androgens. In this review, evidence is presented to support the view that a primary abnormality of ovarian androgen biosynthesis provides the basis for the syndrome. PCOS is a familiar disorder and we demonstrate, in molecular genetic studies, that *CYP11a*, the gene coding for P450 side chain cleavage, is a key susceptibility locus for development of hyperandrogenism.

Contents

1. Introduction

The aetiology of polycystic ovary syndrome (PCOS)—the most common cause of hirsutism and of anovulatory infertility—remains uncertain but there is evidence for a major genetic basis for the syndrome [1–8]. Although the clinical and biochemical presentation in women with polycystic ovaries in variable, there are certain unifying features—in particular, evidence of ovarian hyperandrogenism—which are common to all groups of women with PCOS, irrespective of the mode of presentation [9].

Adrenal androgen concentrations in serum may also be elevated in PCOS but the weight of evidence favours the ovary as the main source of excess androgen production [10]. Suppression of the pituitary–ovarian axis by the chronic administration of agonist and analogues of gonadotrophin-releasing hormone (GnRH) results in a fall in circulating androstenedione and testosterone to levels which are similar to those in ovariectomized or menopausal women [11–13].

Our hypothesis, based on these observations, is that PCOS is characterised by a genetically-determined abnormality in the androgen biosynthetic pathway, affecting, principally the ovary.

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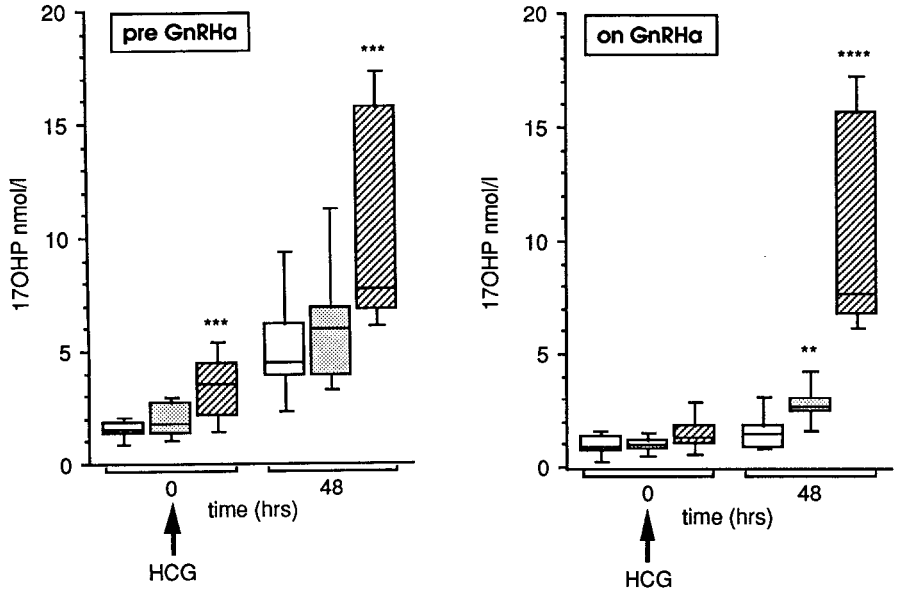

Fig. 1. Response of 17α-hydroxyprogesterone (17-OHP) to hCG (10,000 iu, im) before (pre GnRHa) and during (on GnRHa) suppression of endogenous LH secretion by a long-acting analogue of GnRH in three groups of subjects: normal women (open bars), women with PCO and regular cycles (stippled bars) and women with polycystic ovary *syndrome* (cross-hatched bars). Note, particularly, the exaggerated responsiveness of 17-OHP to hCG in both groups of women with PCO despite prior suppression of endogenous LH for up to four weeks. (adapted from ref [19]).

2. Clinical studies of ovarian and androgen production in PCOS

As a result of clinical studies of ovarian steroid production in women with hyperandrogenaemia, Rosenfield and colleagues have proposed that 'dysregulation' of cytochrome 450c17α (which comprises both 17α-hydroxylase and 17,20 lyase activities) is the central disorder in ovarian hyperandrogenism due to PCOS [14–16]. These workers observed that the rise in serum concentrations of 17α-hydroxyprogesterone (17-OHP) following the acute administration of a GnRH agonist analogue (GnRHa) was greater in women with PCOS than controls and (in PCOS subjects particularly) was proportionally greater than the coincident response of androstenedione.

Studies from our own group have confirmed these results in ovulatory as well as anovulatory women with hyperandrogenaemia and polycystic ovaries [17,18]. The obvious problem in interpretation of these data, is that the response of LH to GnRHa is also exaggerated in PCOS. Although many subjects with increased responsiveness of 17-OHP and androstenedione to GnRHa have normal levels of LH [14,17], these studies do not allow clear distinction to be made between pituitary and ovarian causes of hyperandrogenism. Data from two recent clinical studies, however, strongly suggest that the ovary is the site of the primary abnormality. Women with PCOS given an hCG challenge test produce more androstenedione and 17-OHP than normal subjects [18,19] (Fig. 1).

3. In vitro evidence for ovarian hyperandrogenism in polycystic ovary syndrome

Production of androstenedione and its precursors were examined in monolayer cultures of theca cells obtained from women with normal and polycystic ovaries [20]. Production of androstenedione (both basal and LH-stimulated) in theca cell-conditioned medium was much greater (median 20-fold) in cultures from polycystic compared with normal ovaries. Production of 17-OHP was also significantly (median 7-fold) higher than normal in theca cells cultured from polycystic ovaries, suggesting increased activity of both 17α-hydroxylase and 17,20 lyase.

However, it was also noted that progesterone production was 4–5 times greater by theca from polycystic than normal ovaries. The findings of greatly increased levels of androstenedione and 17-OHP support the concept of increased activity of 17α-hydroxylase/17,20 lyase as an intrinsic feature of PCO theca cells, but the overall data are more consistent with a more general increase in steroidogenesis by polycystic ovaries.

4. Molecular genetic basis of hyperandrogenism in PCOS

The studies of Rosenfield's group [14–16] pointed to *CYP17*, the gene encoding P450c17α as a prime candidate gene in the aetiology of PCOS. In studies conducted at this centre, a common polymorphism was

identified in the regulatory region of the *CYP17* gene (a single base change: a T to C substitution at -34 base pairs from the starting point of translation). The results of a preliminary case-control study indicated that this variant allele was associated with PCOS. This suggested that variation in the regulatory region of *CYP17* conferred increased susceptibility to PCOS. However linkage analysis in families with PCOS, excluded *CYP17* as the major cause of hyperandrogenism in these families [21]. Because of the disparity between these observations and because the preliminary findings were based on analysis of a relatively small population of both cases and controls, a further, follow-up study was performed. In this larger case control study, the variant allele was found to be no more common in women with PCOS than in the reference population and we were unable to find any correlation between the *CYP17* promoter polymorphism and serum testosterone levels [22]. A subsequent study from another centre has confirmed the presence of the -34 C allele as a common variant of *CYP17* but observed that the prevalence of this polymorphism was the same in hyperandrogenaemic subjects (with either PCOS or congenital adrenal hyperplasia) as that in control subjects [23].

The absence of an identifiable genetic abnormality does not, of course, exclude overactivity of P450c17α from having an important effect on ovarian (and, perhaps, adrenal) androgen production. An alternative view, however, is that accumulation of 17-OHP, in the circulation or in theca cells cultures, simply represents a marker of an overall increase in the steroidogenic activity of theca cells in PCOS [24]. It has been shown that 17-OHP is a poor substrate for 17,20 lyase in the human ovary, the delta-5 pathway (i.e. metabolism of 17-hydroxypregnenolone to dehydroepiandrosterone) being the preferred route for the generation of androgens from cholesterol [25]. Stimulation of the ovaries by LH (or hCG) will lead to a greater increase of all theca-derived steroids from polycystic compared with normal ovaries but the inefficiency of 17,20 lyase activity for 17-OHP will inevitably result in the relative abundance of 17-OHP in the circulation or theca-conditioned culture medium.

Because the data from theca cell cultures showed progesterone accumulation was increased in PCO conditioned medium, as well as that of 17-OHP and androstenedione [20], we studied *CYP11a*, the gene coding for P450 cholesterol side chain cleavage (P450scc). A polymorphic region was identified in the promoter region of *CYP11a* (a pentanecleotide repeat -528 bps 5′ to the start site of translation) and both case-control and linkage studies were performed. We found that alleles of *CYP11a* were indeed associated with both hirsutism and hyperandrogenaemia in women with PCO [26]. Importantly, in studies of families with multiple cases of PCOS, we found evidence that this region is a major susceptibility locus for hyperandrogenaemia in women with polycystic ovaries. Using non-parametric methods of analysis which make no assumptions about the mode of inheritance of PCOS, linkage to the *CYP11a* locus was demonstrated. As yet, it remains to be determined whether the -528 polymorphism represents a functional variant of *CYP11a* or whether it is simply in linkage disequilibrium with the disease locus.

It is unclear how this locus interacts with other genetic and environmental factors to influence the development and presentation of PCOS but recent studies from our group have shown evidence for involvement of the insulin gene regulatory region in the aetiology of PCOS [27]. The insulin gene variable number tandem repeat (*INS*-VNTR) appears to be a major susceptibility locus for hyperinsulinaemia and anovulation in women with polycystic ovaries. These findings support the hypothesis that PCOS is an oligogenic disorder in which the interaction of a small number of key genes with each other—and with environmental factors such as diet—can explain the typical clinical and biochemical heterogeneity [8].

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