Differential Male and Female Adrenal Cortical Steroid Hormone and Cortisol Responses to Interleukin-6 in Humans

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ABSTRACT: **Evidence from experimental animal studies show that sex hormones influence the glucocorticoid response to a variety of inflammatory and noninflammatory stimuli. In this study we assessed gender differences in the response of ACTH and cortisol in normal young male and female humans following intravenous infusion of human IL-6 in various dosages. Males presented a significantly stronger ACTH production in response to IL-6 than females. Peak cortisol response, however, was similar in males and females. Cortisol/ACTH ratios were significantly higher in females than in males, both at baseline and after each of the IL-6 dosages. These results suggest that an effective glucocorticoid response requires similar levels of IL-6 in males and females. However, they also suggest that the adrenals of males and females have different sensitivities to ACTH (higher in females) and possibly also to direct IL-6 stimulation.**

KEYWORDS: **gender; sex steroids; cortisol, IL-6**

INTRODUCTION

In both humans and experimental animals, females are more commonly and severely affected by autoimmune diseases. This has been related to the differential effects of sex hormones on the immune system: generally estrogens in physiologic concentrations enhance humoral immune responses and depress cellular-mediated responses, whereas androgens tend to suppress both types of mechanisms.[1]

Although receptors for sex steroids have been shown in a variety of immune competent cells, most of the influence of sex hormones on the immune system *in vivo* may be mediated through indirect mechanism,[2] with emphasis on the modulation of the hypothalamus–pituitary–adrenal (HPA) axis to immune and inflammatory challenge.

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In experimental animals, females have been shown to mount a stronger glucocorticoid response to a variety of stress conditions and inflammatory mediators.[3,4] This is dependent on the effects of sex hormones on several levels of the HPA axis.

Indirect evidence suggests that similar interactions may occur in humans, but this hypothesis has never been directly tested. Thus, we aimed to study adrenal cortical steroid hormone (ACTH) and cortisol levels after a continuous intravenous infusion of interleukin (IL)–6 in healthy adult males and females.

METHODS

The study included 22 volunteers (11 females and 11 males), aged 20 to 41 years. Approval was obtained from the university hospital ethical committee, and informed consent was obtained from all participants. All females were in the late follicular phase of the menstrual cycle.

IL-6 (Sigosix®; Serono Pharmaceuticals, Rome, Italy) was infused intravenously; diluted in saline with 0.2% human albumin, for 60 minutes (between 8:00 and 9:00 A.M.) after overnight rest in hospital, in a dose range of 0.03, 0.1, or 0.3 μg/kg (n = 6, 10, and 6 for each dose group, respectively). Blood samples were collected at baseline 60, 120, and 240 minutes after start of the infusion, using an indwelling catheter. Participants were kept in bed during the full duration of the study. Plasma ACTH was measured by a sensitive enzyme immunoassay (Sangui BioTech, Inc., Santa Ana, CA, via IBL, Hamburg, Germany; detection limit: 0.1 pmol/l). A radioimmunometric assay was used for the quantitative determination of serum cortisol (Coulter Immunotech, Marseille, France; detection limit: 10 nmol/l). Serum IL-6

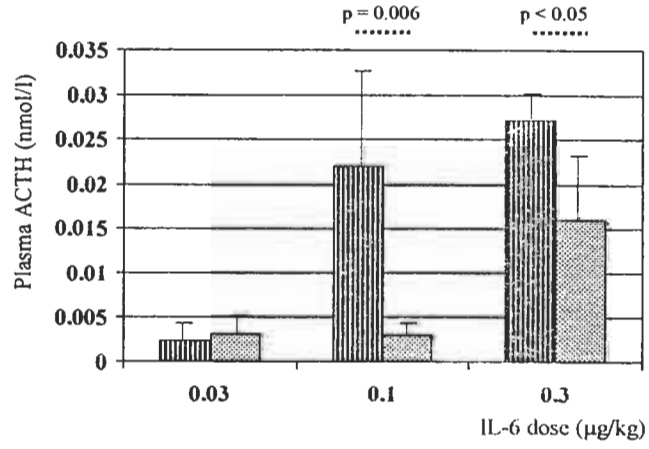

FIGURE 1. Plasma ACTH after one-hour IL-6 intravenous infusion. Males (*striped bars*); females (*dotted bars*).

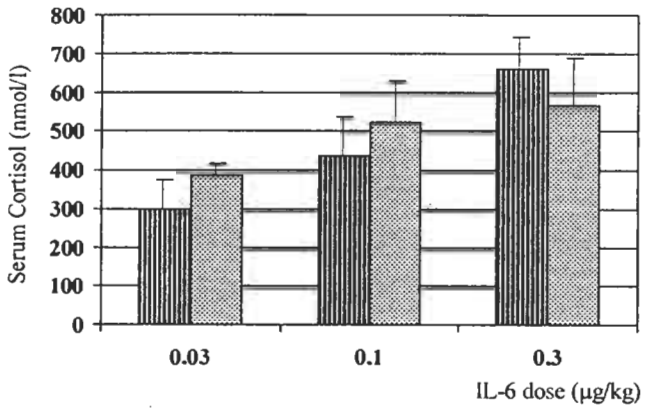

FIGURE 2. Serum cortisol after one-hour IL-6 intravenous infusion. Males (*striped bars*); females (*dotted bars*).

was measured by means of an enzyme immunoassay (high sensitivity Quantikine; R&D Systems, Minneapolis, MN; detection limit: 0.2 pg/ml). Intraassay and interassay coefficients of variation were below 10% in each test.

Mean values were compared by the nonparametric Mann–Whitney test.

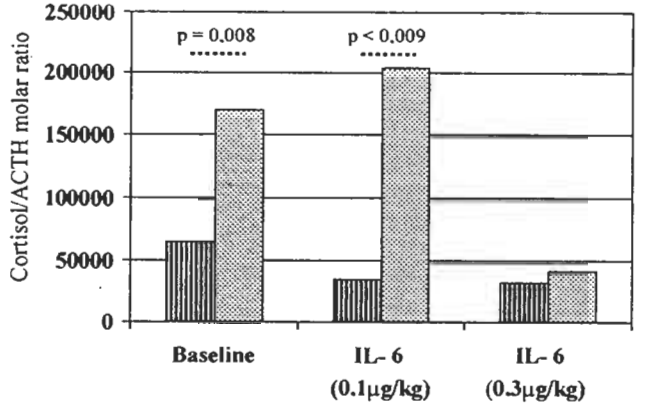

FIGURE 3. Cortisol/ACTH molar ratios in males (*striped bars*) and females (*dotted bars*) at baseline (n = 11 per group), after one-hour IL-6 intravenous infusion at 0.1 µg/kg (n = 5 per group) and 0.3 µg/kg (n = 3 per group).

RESULTS

Males presented a significantly stronger ACTH production in response to IL-6 than females. The difference between the genders reached statistical significance for IL-6 at 0.1 and 0.3 µg/kg (see FIGURE 1). Peak cortisol response, however, was similar in males and females (see FIGURE 2). A significant increase of cortisol from baseline values was observed only with the highest dose of IL-6 (data not shown). In males, ACTH concentrations reached maximum values in the group receiving 0.1 µg/kg IL-6, but this was not associated with a cortisol increase. ACTH values did not rise beyond 0.3 µg/kg IL-6. In females, a significant rise of ACTH was only achieved with 0.3 µg/kg IL-6, coinciding with a significant increase of cortisol. Overall, maximum cortisol levels correlated significantly with serum IL-6 levels in males, but not in females, whereas the opposite was seen for the correlation between ACTH and cortisol values. Furthermore, cortisol/ACTH (see FIGURE 3) ratios were significantly higher in females than in males, both at baseline (n = 22) and after each of the IL-6 doses.

CONCLUSIONS

These results suggest that an effective glucocorticoid response requires similar levels of IL-6 in males and females. However, we observed significant intergender differences in the response of ACTH and cortisol to the various doses of IL-6. This suggests that the adrenal of males and females have different sensitivities to ACTH (higher in females) and possibly also to direct IL-6 stimulation.

Interestingly, the classic concept that the ACTH test does not differ between the genders has been questioned by a recent article in which females presented stronger glucocorticoid responses to synacthen.[5] It has been established that, further to stimulating CRH production in the hypothalamus, IL-6 has direct stimulating effects on adrenal steroidogenesis.[6] On the other hand, sex hormones have been shown to influence IL-6 actions on different target cells.[7,8] Our results open the possibility that this is also the case in the adrenal, given the intergender differences in cortisol/ACTH ratio. In a recent report, Puder *et al.*[9] studied the production of cytokines, ACTH, and cortisol after endotoxin administration in six postmenopausal females, before and after estradiol replacement. Analysis of published data shows that all the patients presented a marked increase in the cortisol: ACTH ratio after estradiol, ranging from 16.5% to 266%, mean, 111.5%. In a recent study, we demonstrated that premenopausal female subjects have significantly higher ratios of serum cortisol:plasma ACTH as compared with postmenopausal women, which may indicate that high estradiol levels may be necessary to maintain high cortisol levels in relation to plasma ACTH. This latter study also demonstrated that no changes of this particular ratio occurred in male patients with increasing ages.[10] Our results are also in line with experimental observations that testosterone reduces the adrenal response to ACTH.[11]

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