

## Gender differences in serum cortisol levels among patients with major depressive disorder

Marsal Sanches<sup>1</sup>, Giovana B Zunta-Soares<sup>1</sup>, John P Hatch<sup>2</sup>, Jair C Soares<sup>1</sup>

<sup>1</sup>UT Center of Excellence on Mood Disorders, Department of Psychiatry and Behavioral Sciences, UT Medical School at Houston

<sup>2</sup>Department of Psychiatry, The University of Texas Health Science Center at San Antonio

Neuropsychiatry i Neuropsychologia 2013; 8, 3–4: 89–93

### Address for correspondence:

Marsal Sanches, MD, PhD  
UT Center of Excellence on Mood Disorders  
Department of Psychiatry and Behavioral Sciences  
UT Medical School at Houston  
e-mail: Marsal.Sanches@uth.tmc.edu

### Abstract

**Introduction:** A large amount of evidence supports the involvement of the hypothalamic-pituitary-adrenal axis (HPA) in the pathophysiology of mood disorders. These findings seem to reflect hyperactivity of the HPA axis in depression and are likely related to impaired negative feedback by endogenous glucocorticoids. However, any gender differences related to this involvement are unclear.

**Material and methods:** We collected serum cortisol samples of 71 unmedicated patients with major depressive disorder (21 males, 50 females; age =  $38.85 \pm 13.66$ ) and 60 healthy controls (19 males, 41 females; age =  $33.25 \pm 11.79$ ). All patients met the DSM-IV-R criteria for major depressive disorder, confirmed through the Structured Clinical Interview for DSM Disorders (SCID). Comparisons of the mean cortisol levels between groups were carried out using factorial analysis of covariance, with age and body weight as covariates.

**Results:** There was a statistically significant interaction between diagnosis and gender ( $F = 6.72$ , d.f. = 1/109,  $p = 0.01$ ). Male patients had significantly higher cortisol levels compared to male controls (patients =  $12.33 \pm 3.63 \mu\text{g/dl}$ , controls =  $10.16 \pm 3.44 \mu\text{g/dl}$ ;  $F = 6.25$ , d.f. = 1/35,  $p = 0.01$ ) whereas female patients had non-significantly lower cortisol levels compared to female controls (patients =  $9.88 \pm 3.03 \mu\text{g/dl}$ , controls =  $11.79 \pm 5.70 \mu\text{g/dl}$ ;  $F = 2.68$ , d.f. = 1/83,  $p = 0.10$ ).

**Conclusions:** Although preliminary, our findings suggest that gender may in part moderate HPA dysfunction in major depressive disorder. Further studies are necessary in order to replicate these findings and investigate their true implications.

**Key words:** mood disorders, major depressive disorder, affective disorders, cortisol, HPA axis, gender.

### Introduction

A large amount of evidence supports the involvement of the hypothalamic-pituitary-adrenal (HPA) axis in the pathophysiology of mood disorders. Many depressed patients show increased levels of salivary and plasmatic cortisol, and a lack of suppression of HPA axis activity by oral dexamethasone (negative dexamethasone suppression test) is a classical finding among patients with major depressive disorder (Pariente and Lightman 2008; Stokes 1995). These findings seem to reflect hyperactivity of the HPA axis in depression and are likely related to impaired negative feedback by endogenous glucocorticoids (Pariente and Lightman 2008).

This increased activity of the HPA axis among depressed patients has many potential consequences. Because of the modulating effects of cortisol on neurogenesis, hypercortisolism may be directly involved in the pathogenesis of several symptoms of depression, such as mood deregulation and memory impairment (Hinkelman *et al.* 2009). Moreover, glucocorticoids play a role in the regulation of several systemic processes, such as the immune system, and may be partially accountable for some of the metabolic disturbances frequently found in patients with mood disorders (Vogelzangs *et al.* 2009).

The present study focused on the cortisol levels of patients with major depressive disorder (MDD) and healthy controls (HC). In light of the available literature, which points to gender

differences between male and female subjects in regard to steroid metabolism and HPA axis activation under stress (Deuschle *et al.* 1998; Shamim *et al.* 2000), we hypothesized that male and female patients would exhibit distinct patterns of HPA axis involvement, with consequent gender-related differences in plasmatic cortisol levels of patients and controls.

## Material and methods

### Participants

The study was carried out at the Division of Mood and Anxiety Disorders of the University of Texas Health Science Center at San Antonio. The sample consisted of 71 outpatients with MDD (21 males, 50 females; mean  $\pm$  SD age = 38.85  $\pm$  13.66 years; BMI = 28.08  $\pm$  7.15) and 60 healthy controls (19 males, 41 females; age = 33.25  $\pm$  11.79 years; BMI = 25.34  $\pm$  4.63). Patients and controls were recruited through local media advertisements and flyers posted in public areas, and were residents of the San Antonio metropolitan area.

All patients met the DSM-IV-R criteria for MDD (single or recurrent), and were unmedicated for a minimum of two weeks at the time of blood sampling. The diagnosis of MDD among patients and the absence of mental disorders among controls were confirmed through the Structured Clinical Interview for DSM Disorders (SCID), which was administered to all participants by trained evaluators. Subjects with any significant medical conditions, such as diabetes or hypertension, were excluded, as were those with neurological disorders or current use of illegal drugs.

For patients, the Hamilton Depression Scale (HAM-D) score was 11.66  $\pm$  9.1 (mean value  $\pm$  SD). Fifty-six patients were acutely depressed at the time of study participation (mean score of the HAM-D  $\pm$  SD = 19.76  $\pm$  4.25), whereas 15 were euthymic. The presence of any oth-

er axis I disorders was adopted as an exclusion criterion, except for anxiety disorders or past substance abuse/dependence, in remission for at least six months. Since our hypothesis emphasized possible gender differences in HPA axis activity, we compared male and female patients with regard to age, number of depressive episodes or other clinical features, and found no significant differences (Table 1).

### Cortisol measurements

Serum cortisol measurements were obtained from all participants. A single blood sample was drawn from the cubital vein and the cortisol levels were estimated through solid phase radioimmunoassay (Riad-Fahmy *et al.* 1979). All subjects were ambulating at the time of the blood draw, but were allowed to rest for at least 10 minutes before the blood samples were obtained. Most of the cortisol samples were obtained in the morning, before breakfast or any kind of strenuous physical activity.

### Statistical analysis

Comparisons of the mean cortisol levels between groups were carried out using factorial analysis of covariance, with age and body weight as covariates. All statistical analysis was carried out using the software Statistical Package for Social Sciences (SPSS) version 14.0. We performed a complementary analysis including time of blood collection as a covariate, in order to account for possible cortisol circadian variations.

### Ethical issues

The study was approved by the University of Texas Health Science Center at the San Antonio institutional review board. Informed consent was obtained from all subjects prior to enrollment in the study.

**Table 1.** Clinical features of male and female patients with major depressive disorder

Feature	Males (n = 21)	Females (n = 50)	p
age in years (mean $\pm$ SD)	38.26 $\pm$ 14.14	38.96 $\pm$ 13.80	NS*
HAM-D scores (mean $\pm$ SD)	12.79 $\pm$ 9.61	11.21 $\pm$ 9.12	NS*
length of illness in months (mean $\pm$ SD)	200.63 $\pm$ 160.13	207.10 $\pm$ 147.34	NS*
age of onset in years (mean $\pm$ SD)	21.63 $\pm$ 6.98	20.83 $\pm$ 11.00	NS*
number of depressive episodes (median)	2	3.5	NS**
BMI	27.36 $\pm$ 5.49	28.39 $\pm$ 7.77	NS*

\*t test

\*\*Mann-Whitney test

## Results

The main effect test of the age and body weight adjusted mean difference between patients and controls with respect to the serum cortisol levels was not statistically significant (patients =  $10.61 \pm 3.38 \mu\text{g/dl}$ , controls =  $11.27 \pm 5.12 \mu\text{g/dl}$ ;  $F = 0.12$ , d.f. = 1/122,  $p > 0.05$ ). The difference remained non-significant even after the non-depressed patients were excluded from the analysis. Moreover, no statistically significant differences were found between depressed and non-depressed patients in terms of cortisol levels (depressed =  $10.97 \pm 3.81 \mu\text{g/dl}$ , non-depressed =  $10.14 \pm 2.72 \mu\text{g/dl}$ ;  $F = 1.04$ , d.f. = 1/64,  $p > 0.05$ ).

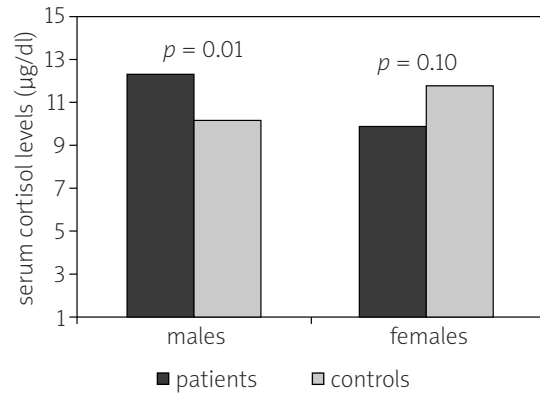
However, there was a statistically significant interaction between diagnosis and gender ( $F = 6.72$ , d.f. = 1/109,  $p = 0.01$ ). Male patients had significantly higher cortisol levels compared to male controls (patients =  $12.33 \pm 3.63 \mu\text{g/dl}$ , controls =  $10.16 \pm 3.44 \mu\text{g/dl}$ ;  $F = 6.25$ , d.f. = 1/35,  $p = 0.01$ ). On the other hand, female patients had nonsignificantly lower cortisol levels compared to female controls (patients =  $9.88 \pm 3.03 \mu\text{g/dl}$ , controls =  $11.79 \pm 5.70 \mu\text{g/dl}$ ;  $F = 2.68$ , d.f. = 1/83,  $p = 0.10$ ) (Fig. 1).

Furthermore, the exclusion of euthymic patients from the analysis elicited similar results (males: patients =  $12.77 \pm 4.65 \mu\text{g/dl}$ , controls =  $10.16 \pm 3.44 \mu\text{g/dl}$ ;  $F = 5.50$ , d.f. = 1/26,  $p \leq 0.05$ ; females: patients =  $10.29 \pm 3.28 \mu\text{g/dl}$ , controls =  $11.79 \pm 5.70 \mu\text{g/dl}$ ;  $F = 0.69$ , d.f. = 1/63,  $p = 0.41$ ). The results also remained similar after we included time of collection as a covariate.

## Discussion

Although sexual dimorphism in the regulation of HPA axis activity has been previously described, this is the first study, to our knowledge, to describe gender-related differences in the serum cortisol levels of patients with MDD. These findings suggest that the role of HPA dysfunction in MDD is more prominent among male patients and may have important implications for a better understanding of that illness and its pathophysiological mechanisms.

Under physiological conditions, synthesis and release of cortisol is down-regulated through binding of endogenous cortisol to specific receptors located at the pituitary, the hypothalamus, and the hippocampus. Thus, decreased number and/or function of glucocorticoid receptors have been



**Fig. 1.** ANCOVA (with age, and body weight as covariates) demonstrated statistically higher ( $p = 0.01$ ) serum cortisol levels among male patients ( $12.33 \pm 3.63 \mu\text{g/dl}$ ) than male healthy controls ( $10.16 \pm 3.44 \mu\text{g/dl}$ ). The difference between female patients ( $9.88 \pm 3.03 \mu\text{g/dl}$ ) and female controls ( $11.79 \pm 5.70 \mu\text{g/dl}$ ) was not statistically significant ( $p = 0.10$ ).

hypothesized as a possible cause of HPA axis hyperactivity in depression. Further, these abnormalities most likely result from the interaction of genetic factors (impaired expression of genes encoding glucocorticoid receptors) and environmental issues (e.g., early-life trauma), which seem to modulate the expression of these genes through epigenetic mechanisms (McGowan *et al.* 2009).

Animal studies suggest a greater HPA axis response to stress, with higher levels of adrenocorticotrophic hormone (ACTH) and cortisol in females than in males (Ogilvie and Rivier 1997). However, research findings among humans are conflicting. Some studies with healthy volunteers display results similar to those described for animals, with higher levels of ACTH and cortisol in males than females under stress. On the other hand, several studies found no gender differences in HPA axis activity under stress exposure (Uhart *et al.* 2006).

The findings are even less consistent among patients with psychiatric disorders. In one study, male patients with panic disorder showed higher salivary cortisol levels than females following a 35% CO<sub>2</sub> challenge (van Duinen *et al.* 2004). Previous findings suggest that baseline cortisol levels may be higher among depressed females than among their healthy counterparts (Young and Korszun 2010). In another study, males and females with MDD displayed a linear correlation between age and baseline cortisol levels, whereas only female healthy subjects were found to show a similar correlation (Halbreich *et al.* 1984). Moreover, smaller hippocampal size in male than female depressed patients has been previously suggested (Frodl *et al.* 2002). Given that decreased hippocampal volume seems to be

a consistent finding in MDD and may be secondary to hypercortisolism, these gender differences would be in consonance with the hypothesis of a greater HPA axis activity among depressed males than among females. However, a meta-analysis failed to demonstrate any gender effects on hippocampal volume among MDD patients (Videbech and Ravnkilde 2004).

Our results suggest that HPA hyperactivity may be more prominent among male than among female MDD patients. Since several clinical factors (such as severity of the illness) seem to influence HPA response patterns in depressed patients (Meador-Woodruff *et al.* 1990), a possible explanation for our findings would be that male patients had a more severe form of MDD. However, no statistically significant differences were found between males and females in regard to the clinical characteristics of depression. Findings similar to ours were obtained by another group in regard to the levels of salivary cortisol, which were higher among depressed males than among male controls, while no differences were found between depressed and non-depressed females (Hinkelmann *et al.* 2012). In regard to urinary free cortisol, a third group reported higher concentrations among male patients with depression than among females (Grant *et al.* 2007). However, the absence of a control group limits the interpretation of those findings. Other studies have not found gender effects regarding higher levels of cortisol among depressed patients (Weber *et al.* 2000).

It has been proposed that gender differences, if present, may result from the involvement of gonadal steroids (particularly estradiol). In rodents, the enhanced HPA axis response to stress seems to be estrogen-mediated, since oophorectomized female rodents display a response pattern similar to males (Ogilvie and Rivier 1997). In a study with healthy volunteers (Kirschbaum *et al.* 1999), women in the luteal phase had responses to stress similar to males, whereas those in the follicular phase had blunted HPA axis responses. On the other hand, abnormalities in gonadal function among female patients with depression have been previously suggested (O'Toole and Rubin 1995). Therefore, we can hypothesize that, in female MDD patients, impairments in the synthesis of gonadal steroids may be present, resulting in attenuated activation of the HPA axis.

Our study has some limitations that are worth discussing. First, we relied solely on baseline cortisol levels as an indicator of HPA activity. Whereas this can be an acceptable approach,

the lack of additional information (such as ACTH levels and pattern of response to the dexamethasone suppression test) restricts the conclusions of the present work.

Moreover, we obtained random samples of serum cortisol, with no control regarding stress exposure. Even though this might represent a methodological issue in our study, the inclusion of time of collection as a covariate did not affect our results. Second, our sample comprised mostly subjects with long-lasting MDD. Although no differences were found between female and male patients in regard to duration of illness, it is not clear if our conclusions could be extended to subjects with a short history of depression. Further, we did not collect data on the use of contraceptives and the phase of the menstrual cycle our female subjects were in at the time of study participation.

In addition, we did not perform any systematic evaluation regarding early-life adverse experiences. Therefore, it is possible (although unlikely) that in our sample male patients had a higher proportion of early trauma than females, which could somehow account for HPA hyperactivity among those patients. On the other hand, no differences were found in regard to the rate of posttraumatic stress disorder among males and females in our sample.

Finally, in our analysis we included not only acutely depressed subjects but also MDD patients in remission. Since previous studies suggest that abnormalities in the HPA axis among depressed patients may be more prominent in the acute phase and normalize after proper treatment, the generality of our findings may be restricted (Holsboer 2000). Nevertheless, there is growing evidence suggesting that elevated cortisol may be a trait-related feature, found not only in acutely depressed patients but also among remitted ones, as well as healthy subjects at elevated genetic risk for MDD (Lok *et al.* 2012). Of note, our results remained similar after non-depressed MDD patients were removed from the analysis.

## Conclusions

Overall, our findings, although preliminary, suggest that gender may in part moderate HPA dysfunction in MDD. Further studies are necessary in order to replicate these findings and investigate their true implications. It has been suggested that HPA axis activity may be implicated in treatment response in MDD (Brouwer *et al.* 2006); therefore putative differences in

HPA axis involvement also may be linked to gender differences in antidepressant response in patients with depression. In addition, dysfunctional HPA activity was recently found to be associated with certain patterns of temperament and character (Hori *et al.* 2013), which hypothetically could impact the course and prognosis of depression.

## Acknowledgments

Partly supported by MH 68766, RR 20571, UTHSCSA GCRC (M01-RR-01346), Veterans Administration (Merit Review), and the Krus Endowed Chair in Psychiatry (UTHSCSA).

## Conflicts of interest

Dr. Soares is a consultant for Organon and Shire Pharmaceuticals, has had research grants from Pfizer, GSK, and Repligen Corporation, currently has grants from Bristol-Myers Squibb and is in the speaker's bureau for Lilly and AstraZeneca. Dr. Sanches has been in the speaker's bureau for AstraZeneca and has received research awards from Janssen Pharmaceuticals.

## References

- Brouwer JP, Appelhof BC, van Rossum EF, et al. Prediction of treatment response by HPA-axis and glucocorticoid receptor polymorphisms in major depression. *Psychoneuroendocrinology* 2006; 31: 1154-1163.
- Deuschle M, Weber B, Colla M, et al. Effects of major depression, aging and gender upon calculated diurnal free plasma cortisol concentrations: a re-evaluation study. *Stress* 1998; 2: 281-287.
- Frodl T, Meisenzahl EM, Zetzsche T, et al. Hippocampal changes in patients with a first episode of major depression. *Am J Psychiatry* 2002; 159: 1112-1118.
- Grant MM, Friedman ES, Haskett RF, et al. Urinary free cortisol levels among depressed men and women: differential relationships to age and symptom severity? *Arch Womens Ment Health* 2007; 10: 73-78.
- Halbreich U, Asnis GM, Zumoff B, et al. Effect of age and sex on cortisol secretion in depressives and normals. *Psychiatry Res* 1984; 13: 221-229.
- Hinkelmann K, Moritz S, Botzenhardt J, et al. Cognitive impairment in major depression: association with salivary cortisol. *Biol Psychiatry* 2009; 66: 879-885.
- Hinkelmann K, Botzenhardt J, Muhtz C, et al. Sex differences of salivary cortisol secretion in patients with major depression. *Stress* 2012; 15: 105-109.
- Hori H, Teraishi T, Sasayama D, et al. Relationship of temperament and character with cortisol reactivity to the combined dexamethasone/CRH test in depressed outpatients. *J Affect Disord* 2013; 147: 128-136.
- Holsboer F. The corticosteroid receptor hypothesis of depression. *Neuropsychopharmacology* 2000; 23: 477-501.
- Kirschbaum C, Kudielka BM, Gaab J, et al. Impact of gender, menstrual cycle phase, and oral contraceptives on the activity of the hypothalamus-pituitary-adrenal axis. *Psychosom Med* 1999; 61: 154-162.
- Lok A, Mocking RJ, Ruhé HG, et al. Longitudinal hypothalamic-pituitary-adrenal axis trait and state effects in recurrent depression. *Psychoneuroendocrinology* 2012; 37: 892-902.
- McGowan PO, Sasaki A, D'Alessio AC, et al. Epigenetic regulation of the glucocorticoid receptor in human brain associates with childhood abuse. *Nat Neurosci* 2009; 12: 342-348.
- Meador-Woodruff JH, Greden JF, Grunhaus L, Haskett RF. Severity of depression and hypothalamic-pituitary-adrenal axis dysregulation: identification of contributing factors. *Acta Psychiatr Scand* 1990; 81: 364-371.
- O'Toole SM, Rubin RT. Neuroendocrine aspects of primary endogenous depression – XIV. Gonadotropin secretion in female patients and their matched controls. *Psychoneuroendocrinology* 1995; 20: 603-612.
- Ogilvie KM, Rivier C. Gender difference in hypothalamic-pituitary-adrenal axis response to alcohol in the rat: activation role of gonadal steroids. *Brain Res* 1997; 766: 19-28.
- Pariante CM, Lightman SL. The HPA axis in major depression: classical theories and new developments. *Trends Neurosci* 2008; 31: 464-468.
- Riad-Fahmy D, Read GF, Gaskell SJ, et al. A simple, direct radioimmunoassay for plasma cortisol, featuring a 125I radioligand and a solid-phase separation technique. *Clin Chem* 1979; 25: 665-668.
- Shamim W, Yousufuddin M, Bakhai A, et al. Gender differences in the urinary excretion rates of cortisol and androgen metabolites. *Ann Clin Biochem* 2000; 37 (Pt 6): 770-774.
- Stokes PE. The potential role of excessive cortisol induced by HPA hyperfunction in the pathogenesis of depression. *Eur Neuropsychopharmacol* 1995; 5 Suppl: 77-82.
- Uhart M, Chong RY, Oswald L, et al. Gender differences in hypothalamic-pituitary-adrenal (HPA) axis reactivity. *Psychoneuroendocrinology* 2006; 31: 642-652.
- van Duinen MA, Schruers KR, Jaegers E, et al. Salivary cortisol in panic: are males more vulnerable? *Neuro Endocrinol Lett* 2004; 25: 386-390.
- Videbech P, Ravnkilde B. Hippocampal volume and depression: a meta-analysis of MRI studies. *Am J Psychiatry* 2004; 161: 1957-1966.
- Vogelzangs N, Beekman AT, Dik MG, et al. Late-life depression, cortisol, and the metabolic syndrome. *Am J Geriatr Psychiatry* 2009; 17: 716-721.
- Weber B, Lewicka S, Deuschle M, et al. Increased diurnal plasma concentrations of cortisone in depressed patients. *J Clin Endocrinol Metab* 2000; 85: 1133-1136.
- Young E, Korszun A. Sex, trauma, stress hormones and depression. *Mol Psychiatry* 2010; 15: 3-28.