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Neuroendocrine response during stress with relation to gender differences

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Abstract. Neuroendocrine activation belongs to the main characteristics of the stress response. This response is not uniform but depends on the stress stimulus involved and on many other factors including the gender of the individual. In rats, corticosterone and ACTH levels as well as functional activity of the hypothalamo-pituitary-adrenocortical axis are higher in females compared to males under both basal and stress conditions. Marked sex differences were observed in stress-induced changes in posterior pituitary hormone release. In male rats, release of vasopressin is not stimulated during stress conditions without an osmotic component while in female rats a rise in plasma vasopressin levels was observed even after short immobilization. Oxytocin release is enhanced in response to the majority of stress stimuli and it was found to be greater in females than in males. Mentioned gender differences are attributed to the effect of sex steroids, particularly those of estrogens. Not enough information is available on gender differences in the neuroendocrine response during stress in humans. We observed a greater neuroendocrine activation in women than in men in response to heat exposure in sauna with pronounced differences in ACTH and prolactin release and partly also after a cold-pressor test. Understanding of gender differences in neuroendocrine response during stress might contribute to the explanation of the development of some emotional and other disorders with higher incidence in women.

Mini-review

Key words: stress, hormones, neuroendocrine response, gender differences

NEUROENDOCRINE RESPONSE DURING STRESS

Neuroendocrine activation belongs to the main characteristics of the stress response in general. Nowadays it is clear that exposure to stress conditions evokes a broad spectrum of changes resulting in stimulation or inhibition of many factors in the brain, endocrine glands and other organs. The number of factors under consideration is still increasing.

In the original stress concept of Hans Selye, stress was considered to be a nonspecific response of the body to any demand (Selye 1936). Later on, development of new techniques made possible to analyze small amounts of biologically active substances in the blood and tissues and the information gathered enabled enlargement and reevaluation of the stress theory. One of the questions repeatedly discussed is the specificity of the stress response.

According to the data of others (Kopin 1995; Pacák et al. 1995) as well as our own research (e.g. Vigaš et al. 1984), neuroendocrine response during stress is not uniform but depends on the stress stimulus involved and on many other conditions. As to the stress stimulus, both its nature and intensity determine resulting changes. There are several other modifying factors, such as species differences, the time of the day or environmental temperature (Ježová et al. 1995a). Neuroendocrine response is significantly determined by the previous stress history and by actual state of the body - the age, health and disease, motivation and other emotional preconditions. Among important factors influencing the stress response we have to consider certain life periods and the gender of the individual.

In general, a stress signal is recognized by neural centers and *via* neurotransmitters and neuropeptides transmitted to neuroendocrine centers located in the hypothalamus. Changes in hypothalamic releasing and inhibiting hormones induce pituitary response with consequent changes in peripheral stress hormone secretion. All this results in cardiovascular, metabolic and immune changes aimed to overcome increasing demands of the body. The dominant components of the complex stress system (Fig. 1) in the central nervous system (CNS) are the corticotropin-releasing factor (CRF) neurons of the hypothalamic paraventricular nucleus and the cate-

MOST IMPORTANT COMPONENTS OF THE STRESS SYSTEM

Central components (coordinators)	* **paraventricular nucleus of the hypothalamus** (CRF, vasopressin)
	* **locus coeruleus of the brainstem** (norepinephrine)
Peripheral components (effectors)	* **pituitary-adrenocortical axis**
	* **sympathetic adrenomedullary system**

Fig. 1. A simplified representation of the central and peripheral components of the stress system.

cholaminergic neurons of the locus coeruleus (LC) and other cell groups of the medulla and the pons. Further, hypothalamo-pituitary-adrenocortical (HPA) axis and sympathetic adrenomedullary system are considered to be peripheral limbs of the stress system (Stratakis and Chrousos 1995).

Though hormones of the hypothalamo-pituitary-gonadal axis are not considered to be typical stress hormones, the reproductive function is very directly linked to the stress system. It is well known that the reproductive axis is inhibited at several levels by the components of the stress system and this interaction is thought to be bidirectional (Rabin et al. 1988). On the other hand, the hormones prolactin and oxytocin, which both exert specific roles in the course of reproduction, are rapidly activated in response to many stress stimuli. Plasma testosterone levels are for a long time known to be markedly reduced during chronic and intensive stress situations both in men and animals (Carstensen et al. 1973, Nakashima et al. 1975, Dessypris et al. 1976, Repčeková and Mikulaj 1977). However, several physical stress stimuli, such as exercise on a bicycle ergometer are inducing an elevation of plasma testosterone (Brisson et al. 1977, Ježová-Repčeková et al. 1982, Ježová et al. 1985).

GENDER DIFFERENCES IN THE STRESS RESPONSE IN RATS

Sex steroids have been shown to influence several components of the stress system. In particular, a number of reports have demonstrated an interaction between the HPA and reproductive endocrine systems. It is well established that corticosterone and ACTH levels as well as functional activity of the HPA axis are higher in females compared to males under both basal and stress conditions (Kitay 1961, Macho et al. 1975, Le Mevel et al. 1979, Lesniewska et al. 1990). An augmented plasma corticosterone response in females was noted following stress stimuli of different nature including an acute activation of the immune system by lipopolysacharide administration used in our laboratory (unpublished data). The higher plasma corticosterone concentrations observed after stress exposure in female rats were not explained by the gender differences in the clearance or metabolism of the steroid (Kitay 1961). The involvement of more central components of the stress system, namely the pituitary and the hypothalamus are now well established (Vamvakopoulos and Chrousos 1994).

Gonadectomy and sex hormone replacements were used to evaluate regulatory pathways underlying this dimorphism (e.g. Lesniewska et al. 1990). Gender differences in the HPA axis can be attributed to the effect of sex steroids, particularly those of estrogens. Increased basal and stress-stimulated HPA axis function in the female gender was noted already in young animals (Hary et al. 1981) and the sexual dimorphism seems to be preformed prenatally and in early postnatal life (Dupouy et al. 1987).

Of particular importance is the finding of Vamvakopoulos and Chrousos (1993) of the presence of estrogen-responsive elements in the promoter area of the CRF gene and direct stimulatory estrogen effects on CRF gene expression. Thus, CRF neurons and the whole HPA axis may be considered a significant target of ovarian steroids and a potential mediator of gender-related differences in the stress response (Vamvakopoulos and Chrousos 1994, Stratakis and Chrousos 1995).

In the rat, marked sex differences were observed in stress-induced changes in posterior pituitary hormone release. Intristingly, the two neuropeptides, vasopressin and oxytocin, are produced in the same magnocellular hypothalamic nuclei and they are similar in structure but differ in their physiological roles as well as in the pattern of stress-induced changes (Ježová et al. 1995b).

Vasopressin is of little importance for reproduction and its dominant role is the control of water-electrolyte balance and blood volume. Release of vasopressin is stimulated during stress conditions involving an osmotic component but not in response to other stressors activating the typical hormones of the stress response, such as immobilization (Fig. 2). However, in female rats a small but significant rise in plasma vasopressin levels was observed even

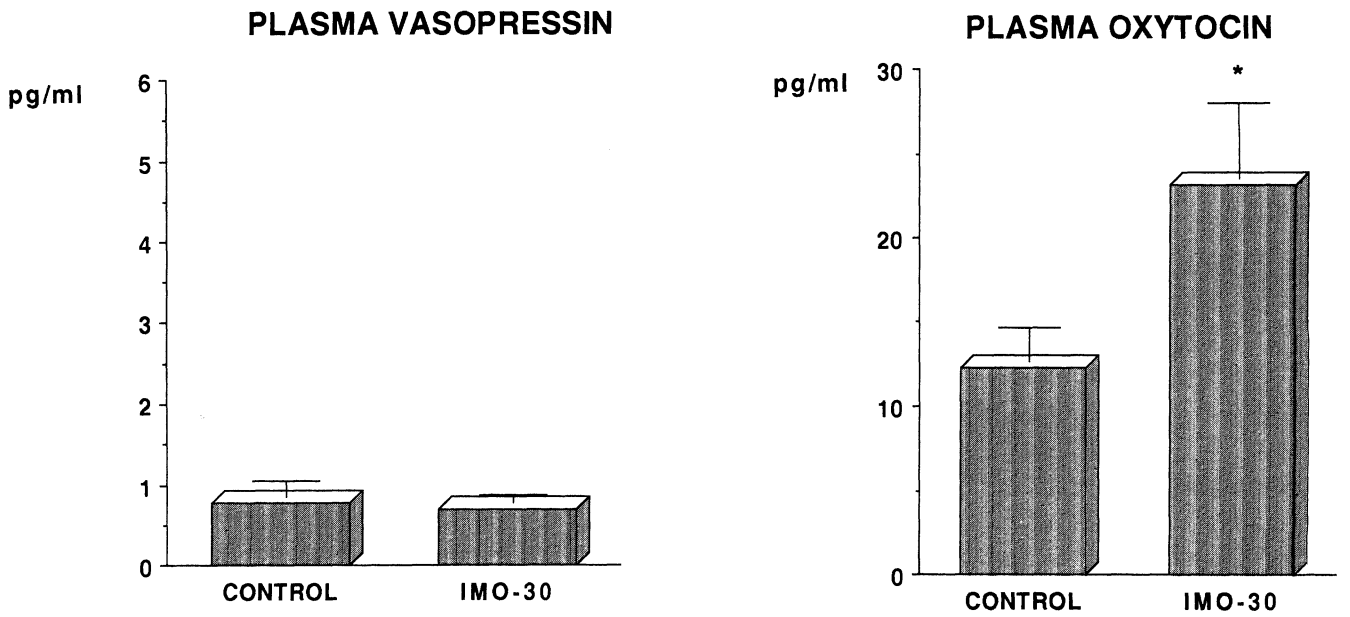

Fig. 2. Plasma oxytocin and vasopressin levels in response to immobilization stress. Adult male Sprague-Dawley rats ($n = 7$) were decapitated under resting conditions (control) and 30 min after immobilization (IMO-30). Vasopressin and oxytocin levels were measured by radioimmunoassays (Ježová and Michajlovskij 1992, Ježová et al. 1993). Means ± SEM. Statistical significance vs. control values: $*P<0.05$.

after short immobilization (Williams et al. 1985). Moreover, gender differences in hypothalamic vasopressin mRNA levels and plasma osmolality were described indicating higher values in females (Dai and Yao 1995).

Unlike vasopressin, oxytocin is important in reproduction-related events, such as parturition and lactation (Neumann et al. 1996). Though a specific role for oxytocin in adaptation to adverse stimuli has not yet been revealed, oxytocin release is known to be a component of the neuroendocrine response to the majority of stress stimuli (Ježová et al. 1995b). Similarly as in the case of HPA axis activity, stress-induced oxytocin release in females was found to be greater than that found in males (Williams et al. 1985). The mentioned authors reported that ovariectomy did not modify the female response, but orchidectomy resulted in an enhanced response of the peptide to immobilization stress in the male, indicating an inhibitory action of androgens. On the other hand, androgenization of female rats in the early postnatal period reduced oxytocin stress responses of adults to a level not different to males (Carter et al. 1988). A modulatory role of gonadal steroids on posterior pituitary hormone production is underlined by the studies on intact and gonadectomized female and male rats subjected to a chronic osmotic challenge. After osmotic activation, only intact animals had enhanced cytoplasmic oxytocin and vasopressin mRNA concentrations in the magnocellular hypothalamic nuclei (Crowley and Amico 1993). Even the regulatory mechanisms controlling oxytocin release might involve gender dependent events. Carter and Lightman (1987) provided functional evidence of a sexually dimorphic inhibitory role of certain central noradrenergic pathways in the control of oxytocin secretion.

In spite of the observed changes in hormone production, no sexual dimorphism in the inhibitory action of oxytocin on feeding and drinking behavior was observed (Benelli et al. 1991). On the other hand, *in vitro* studies in 8-day-old newborn rats showed that oxytocin potentiated ACTH release in response to CRF by the anterior pituitary lobes of females but not by those of males (Hary et al. 1993). Further studies might verify a possible relation of these findings to the enhanced activity of the HPA axis in females.

GENDER DIFFERENCES IN THE STRESS RESPONSE IN HUMANS

Gender differences in the stress response in humans are less understood than those in experimental animals though some differences in hormonal responses to certain (Johansson et al. 1989), but not all (Ng et al. 1994) stress stimuli in female and male subjects were occasionally reported . Our attention was given to the evaluation of possible gender differences in neuroendocrine response during heat exposure with the use of sauna bath as a model of heat stress (Ježová et al. 1994).

In our studies, healthy young men and women were subjected to a sauna bath of relatively low intensity (80°C) for 20 min. Endocrine and cardiovascular responses were evaluated at the end of the exposure in the sauna room and until 30 min thereafter. Rise in body temperature was the same in both sexes. A significant rise in plasma ACTH was observed only in the female subjects while cortisol levels were similar in both groups. Similarly, hyperthermia-induced elevation in prolactin release was significantly more pronounced in women than in men. No gender differences in the responses of plasma catecholamines were noticed but the increase in heart rate during the sauna bath was higher in women. Thus, the neuroendocrine response to sauna exposure of mild intensity is significantly more marked in women that in men (Ježová et al. 1994).

We, as well as others have demonstrated that heat stress is accompanied by a complex neuroendocrine response while the exposure of humans to cold is inducing predominantly an activation of the sympathetic nervous system (Vigaš et al. 1984, Ježová et al. 1985, Kauppinen and Vuori 1986, Vigaš et al. 1988, Wittert et al. 1992, Ježová et al. 1994). In a study on a large group of medical students subjected to a short cold-pressor test, we observed higher increases in the heart rate (Fig. 3) but not other cardiovascular parameter in women. This is in good agreement with the data of Allen et al. (1993) who found that women exhibited larger increases in heart rate in response to psychological stress stimuli while men responded with greater

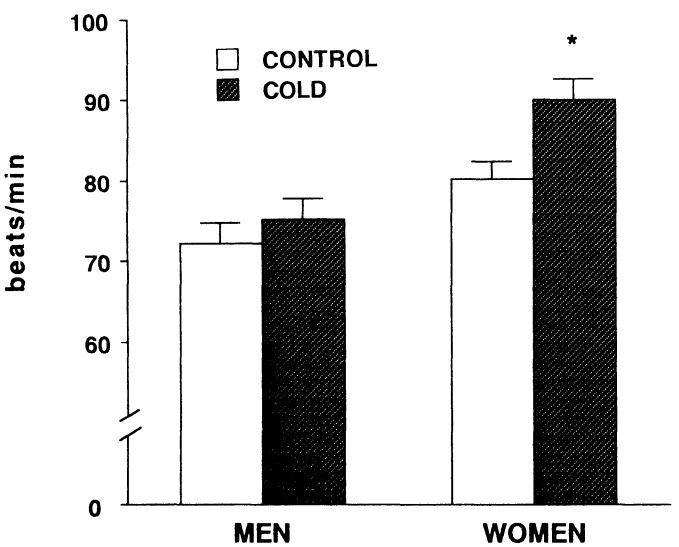

Fig. 3. Gender differences in heart rate response during a short cold pressor test. Healthy students were investigated in the sitting position at room temperature (control) and during placing an arm ankle deep into a pan of ice water for 1 min (cold). Heart rate was measured using an automatic recorder (Dinamap, Criticon, Stampa, USA). Means ± SEM. Statistical significance vs. control values: *$P<0.05$.

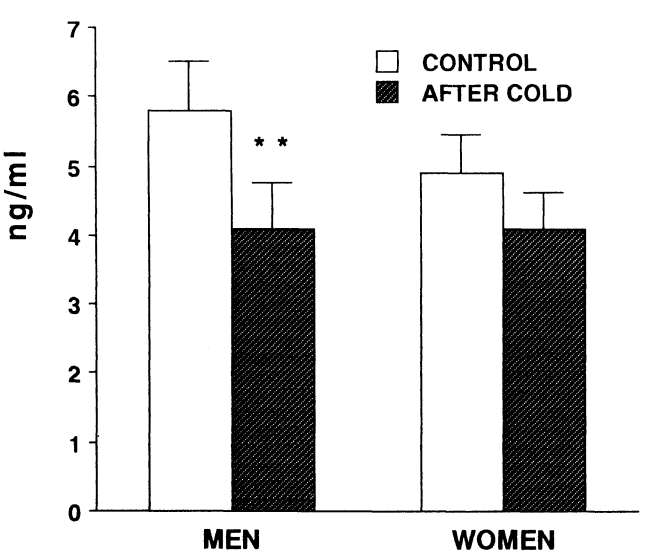

Fig. 4. Salivary cortisol levels in men and women subjected to a cold pressor test. Samples of saliva were obtained before and after a time period of about 60 min involving a short cold exposure. The second sample was taken 20 min after the end of the cold test. Salivary levels of cortisol were measured by a radioimmunoassay (Ježová et al. 1992). Means ± SEM. Statistical significance vs. control values: **$P<0.01$.

blood pressure changes. Consistently, cortisol release in response to this mild stress exposure seemed to be higher in women as indicated by the absence of a significant fall of its concentration during the observation period (about 60 min) found in men (Fig. 4). As an indicator of cortisol release and HPA axis activity, the non-invasive sampling of saliva and measurement of salivary cortisol was used. In accordance with the daily rhythm of cortisol secretion, we have repeatedly observed a significant decrease in its concentrations both in plasma and saliva within a relatively short time period in the morning hours (Ježová and Vigaš 1988). Our results suggest an enhanced pituitary-adrenocortical response to cold exposure in women. This is in agreement with the data of Gerra et al. (1992) who found significant increments in ACTH and cortisol levels in women but not in men exposed to cold for 30 min.

Differences in the stress response between men and women could have several clinical implications. Knowledge of sympathetic-adrenomedullary activation in both sexes is important because of recognized gender differences in incidence of cardiovascular disease. According to Galluci et al. (1993), enhanced activation of CRF neurons by estrogens may be considered an explanation as to why various emotional disorders characterized by elevated CRF secretion, such as depression and anxiety, have a higher incidence in woman than in men.

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REFERENCES

Allen N.T., Stoney C.M., Owens J.F., Matthews. K.A. (1993) Hemodynamic adjustments to laboratory stress: the influence of gender and personality. Psychosom. Med. 55: 505-517.

Benelli A., Bertolini A., Arletti R. (1991) Oxytocin-induced inhibition of feeding and drinking: no sexual dimorphism in rats. Neuropeptides 20: 57-62.

Brisson G.R., Volle M.A., Desharnais M., Dion M., Tanaka M. (1977) Pituitary gonadal axis in exercising men. Med. Sci. Sp. 9: 47-52.

Carstensen H., Amer I. Wide L., Amer B. (1973) Plasma testosterone, LH and FSH during the first 24 hours after surgical operations. J. Steroid Biochem. 4: 605-611.

Carter D.A., Lightman S.L. (1987) Modulation of oxytocin secretion by ascending noradrenergic pathways: sexual dimorphism in rats. Brain Res. 406: 313 316.

Carter D.A., Saridaki E., Lightman S.L. (1988) Sexual differentiation of oxytocin stress responsiveness: effect of neonatal androgenization, castration and a luteinizing hormone-releasing hormone antagonist. Acta Endocrinol. (Copenh.) 117: 525-530.

Crowley R.S., Amico JA. (1993) Gonadal steroid modulation of oxytocin and vasopressin gene expression in the hypothalamus of the osmotically stimulated rat. Endocrinology 133: 2711-2718.

Dai W.J., Yao T. (1995) Effects of dehydration and salt-loading on hypothalamic vasopressin mRNA level in male and female rats. Brain Res. 676: 178-182.

Dessypris A., Kuoppasalmi K., Adlercreutz H. (1976) Plasma cortisol, testosterone, androstenedione and luteinizing hormone (LH) in a non-competitivge marathon run. J. Steroid Biochem. 7: 33-37.

Dupouy J.P., Hary L., Lalau J.D., Grégoire I., Chatelain A. (1987) Influence périnatale des hormones sexuelles sur l'activation différentielle de la fonction corticotrope au cours d'un stress chez le male et la femelle. Annales d'Endocrinologie 48: 385-392.

Galluci W.T., Baum A., Laue L., Rabin D.S., Chrousos G.P., Gold P.W. Kling M.A. (1993) Sex differences in sensitivity of the hypothalamic-pituitary-adrenal axis. Health Psychol. 12: 420-425.

Gerra G., Volpi R., Delsignire R., Maninetti L., Caccavari R., Vourna S., Maestri D., Chiodera P., Ugolotti G., Coiro V. (1992) Sex-related responses of beta-endorphin, ACTH, GH and PRL to cold exposure in humans. Acta Endocrinol. (Copenh.) 126: 24-28.

Hary L., Dupouy J.P., Chatelain A. (1981) Pituitary response to bilateral adrenalectomy, metyrapone treatment and ether stress in the newborn rat. Biol. Neonate 39: 28-36.

Hary L., Dupoy J.P., Chatelain A. (1993) ACTH secretion from isolated hypophysial anterior lobes of male and female newborn rats: effects of corticotrophin-releasing factor, arginine vasopressin and oxytocin alone or in combination. J. Endocrinol. 137: 123-132.

Ježová D., Juránková E., Kvetňanský R., Kaciuba-Uscilko H., Nazar K., Vigaš M. (1995a) Low ambient temperature and neuroendocrine response to hypoglycemia in men. Obes. Res. 3: 713S-719S.

Ježová D., Kvetňanský R., Vigaš M. (1994) Sex differences in endocrine response to hyperthermia in sauna. Acta Physiol. Scand. 150: 293-298.

Ježová D., Michajlovskij N. (1992) N-methyl-D-aspartic acid injected peripherally stimulates oxytocin and vasopressin release. Endocrinol. Reg. 26: 73-75.

Ježová D., Michajlovskij N., Kvetňanský R., Makara G.B. (1993) Paraventricular and supraoptic nuclei of the hypothalamus are not equally important for oxytocin release duringstress. Neuroendocrinology 57: 776-781.

Ježová D., Škultétyová I., Tokarev D.I., Bakoš P., Vigaš M. (1995b) Vasopressin and oxytocin in stress. Ann. N.Y. Acad. Sci. 771: 192-203.

Ježová D., Slezák V., Alexandrová M., Motovská Z., Juránková E., Vigaš M., Černý J. (1992) Professional stress in surgeons and artists as assessed by salivary cortisol. In: Stress: neuroendocrine and molecular approaches (Eds. R. Kvetňanský, R. McCarty and J. Axelrod). Gordon and Breach, New York, p. 953-962.

Ježová D., Vigaš M. (1988) Apomorphine injection stimulates ß-endorphin, adrenocorticotropin, and cortisol release in healthy man. Psychoneuroendocrinology 13:479-485.

Ježová D., Vigaš M., Tatár P., Jurčovičová J., Palát M. (1985) Rise in plasma beta-endophin and ACTH in response to hyperthermia in sauna. Horm. Metab. Res. 17: 693-694, 1985.

Ježová-Repčeková D., Vigaš M., Mikulaj L., Jurčovičová J. (1982) Plasma testosterone during bicycle ergometer exercise without and after L-dopa pretreatment. Endocrinol. Exp. 16: 3-8.

Johansson G.G., Laakso M., Peder M., Karonen S.L. (1989) Endocrine patterns before and after examination stress in males and females. Activ. Nerv. Super. 31: 81-88.

Kauppinen K., Vuori I. (1986) Man in the sauna. Ann. Clin. Res. 18: 173-185.

Kitay J.I. (1961) Sex differences in adrenal cortical secretion in the rat. Endocrinology 68: 818-824.

Kopin I.J. (1995) Definitions of stress and sympathetic neuronal responses. Ann. N.Y. Acad. Sci. 771: 19-30.

Le Mevel J.C., Abitbol S., Beraud G., Maniey J. (1979) Temporal changes in plasma adrenocorticotropin concentration after repeated neurotropic stress in male and female rats. Endocrinology 105: 812-817.

Lesniewska B., Nowak M., Malendowicz L.K. (1990) Sex differences in adrenocortical structure and function. XXVIII. ACTH and corticosterone in intact, gonadectomised and gonadal hormone replaced rats. Horm. Metab. Res. 22: 378-381.

Macho L., Alexandrová M., Hromadová M., Štrbák V., Kvetňanský R. (1975) Response of the adrenals to stress in animals from litters with different numbers of young. Physiol. Bohemoslov. 24: 501-508.

Nakashima A., Koshiyama K., Uozumi T., Monden Y., Hamanaka Y., Kurachi K., Aono T., Mizutani S., Matsumoto K. (1975) Effects of general anesthesia and severity of surgical stress on serum LH and testosterone in males. Acta Endocrinol. (Copenh.) 78: 258-269.

Neumann I., Douglas A.J., Pittman Q.J., Russell J.A., Landgraf R. (1996) Oxytocin released within the supraoptic nucleus of the rat brain by positive feedback action is involved in parturition-related events. J. Neuroendocrinol. 8: 227-233.

Ng A.V., Callister R., Johnson D.G., Seals D.R. (1994) Sympathetic neural reactivity to stress does not increase with age in healthy humans. Am. J. Physiol. 267: H344-H353.

Pacák K., Palkovits M., Kvetňanský R., Zadid G., Kopin I.J., Goldstein D.S. (1995) Effects of various stressors on in vivo norepinephrine release in the hypothalamic paraventricular nucleus and on the pituitary-adrenocortical axis. Ann. N.Y. Acad. Sci. 771: 115-130.

Rabin D., Gold P.W., Margioris A., Chrousos G.P. (1988) Stress and reproduction: interaction between the stress and reproductive axis. In: Mechanisms of physical and emotional stress (Eds. P.W. Gold, D.L. Loriaux and G.P. Chrousos). Plenum Press, New York, p. 377-387.

Repčeková D., Mikulaj L. (1977) Plasma testosterone of rats subjected to immobilization stress and/or HCG administration. Horm. Res. 8: 51-57.

Selye H. (1936) A syndrome produced by diverse noxious agents. Nature 32: 138.

Stratakis C.A., Chrousos G.P. (1995) Neuroendocrinology and pathophysiology of the stress system. Ann. N.Y. Acad. Sci. 771: 1-18.

Vamvakopoulos N.C., Chrousos G.P. (1993) Evidence of direct estrogenic regulation of human corticotropin releasing hormone gene expression: potential implications for the sexual dimorphism of the stress response and immune/inflammatory reaction. J. Clin. Invest. 92: 1896-1902.

Vamvakopoulos N.C., Chrousos G.P. (1994) Hormonal regulation of human corticotropin-releasing hormone gene expression: Implications for the stress response and immune/inflammatory reaction. Endocrinol. Rev. 15: 409-420.

Vigaš M., Kvetňanský R., Jurčovičová J., Ježová D., Tatár P. (1984) Comparison of catecholamine and adenopituitary hormone responses to various stress stimuli in man. In: Stress: the role of catecholamines and other neurotransmitters (Eds. E. Usdin, R. Kvetňanský and J. Axelrod). Gordon and Breach Science Publishers, New York, p. 865-882.

Vigaš M., Martino E., Bukovská M., Langer P. (1988) Effect of acute cold exposure and insulin hypoglycemia on plasma thyrotropin levels by IRMA in healthy young males. Endocrinol. Exp. 22: 229-234.

Williams T.D.M., Carter D.A., Lightman S.L. (1985) Sexual dimorphism in the posterior pituitary response to stress in the rat. Endocrinology 116: 738-740.

Wittert G.A., Or H.K., Livesey J.H., Ricards AM., Donald R.A., Espiner E.A. (1992) Vasopressin, corticotrophin-releasing factor, and pituitary adrenal responses to acute cold stress in normal humans. J. Endocrinol. Metab. 75: 750-755.

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