Pharmacology Biochemistry & Behavior, Vol. 29, pp. 699–703. © Pergamon Press plc, 1988. Printed in the U.S.A. 0091-3057/88 $3.00 + .00

Influence of Exogenously Administered Oxytocin on the Corticosterone and Prolactin Response to Psychological Stress

JANICE L. MUIR AND H. PETER PFISTER

Department of Psychology, The University of Newcastle N.S.W. 2308, Australia

Received 1 December 1986

MUIR, J. L. AND H. P. PFISTER. *Influence of exogenously administered oxytocin on the corticosterone and prolactin response to psychological stress.* PHARMACOL BIOCHEM BEHAV **29**(4) 699–703, 1988.—Prolactin along with corticosterone is a stress responsive hormone. Evidence suggests that oxytocin (OXT) modulates not only ACTH secretion but also prolactin release. The present study was therefore designed to examine the possible role of oxytocin in the corticosterone and prolactin response to predictable and unpredictable novelty stress. Repeated stress and oxytocin treatment produced a substantial increase in corticosterone. A greater increase was obtained for the larger OXT dose (11.6 IU/kg) than for the smaller dose (5.8 IU/kg). In addition, for the smaller oxytocin dose only, unpredictable exposure to the novelty apparatus produced a more substantial increase in corticosterone than predictable exposure to the same stressor. In contrast, oxytocin produced a significant suppression of the prolactin response in all OXT treated animals. No significant interaction between stress and oxytocin was obtained. It was concluded that an important role exists for oxytocin in the modulation of both corticosterone and prolactin secretion.

Oxytocin Corticosterone Prolactin Novelty Predictable stress Unpredictable stress

WE have recently examined the possibility that oxytocin (OXT) has a role in the corticosterone response to a psychological stressor [20]. Although corticotropin-releasing factor (CRF) appears to be the primary hypothalamic hormone involved in the control of ACTH secretion, it has been suggested that additional neuropeptides such as oxytocin may also contribute to the regulation of ACTH release by potentiating CRF-induced ACTH secretion [1, 3, 8]. The results of our study [20] clearly demonstrated that oxytocin produces a significant increase in corticosterone in all OXT treated animals. However, potentiation of the corticosterone response to a psychological stressor was not observed following administration of OXT. This result confirms previous reports [5,14] which have stated that the participation of OXT in the control of ACTH secretion may be stress specific.

To further elucidate a role for OXT in the response to a stressor, Gibbs [6] examined the effect of injections of anti-oxytocin antiserum on stress-induced ACTH secretion. Gibbs [6] reported that in vivo immunoneutralization of oxytocin attenuated the ACTH response to tail-hang stress, suggesting that, at least for this stressor, OXT was involved in the physiological regulation of ACTH secretion. However, since OXT appears to be selectively released in response to some but not all types of stress, a further study [7] examined the ability of anti-oxytocin antiserum to attenuate ACTH secretion following tail-hang stress, ether stress and exposure to a novel environment. Gibbs [7] found that the ability of the antiserum to attenuate ACTH secretion was dependent on whether OXT was itself stimulated by that particular stressor. While oxytocin secretion was stimulated by tail-hang and ether stress it was unchanged by exposure to the novel environment despite a 3-fold increase in ACTH concentration. Subsequently, although antiserum significantly attenuated the ACTH response to tail-hang and ether stress, it had no effect on the ACTH response to a novel environment. Gibbs suggests that basal levels of OXT have no direct effect on ACTH secretion and that until OXT levels rise in response to stress, the potentiating effects are not expressed.

However, the ability of OXT to modulate anterior pituitary function is not limited to a role in ACTH secretion but also includes a possible involvement in prolactin (PRL) release. Numerous examples exist of the concomitant release of oxytocin and prolactin. For instance, both hormones are released after suckling in the human [35] and in the rat [27]. Oxytocin is in fact present in the appropriate anatomical location to alter anterior pituitary function. Neural fibres stained immunocytochemically with OXT-specific antisera have been found in the zona externa of the median eminence [30,33] with some of these OXT fibres in contact with the

[1]Requests for reprints should be addressed to Dr. H. Peter Pfister.

portal capillaries [30]. Consequently, the issue of whether OXT possesses an ability to release PRL from the anterior pituitary gland has received some attention and has also been the subject of controversy. Petersen [22] postulated that OXT could stimulate PRL release, based on the observation that suckling releases both OXT from the neurohypophysis and PRL from the adenohypophysis. However, while Gala and Reece [4] found that incubation of OXT with quartered pituitary glands would release bioassayable PRL, other investigators have found no effect of OXT on PRL release in vitro [21,32].

In vivo studies using lactating rats have also yielded conflicting results. Benson and Folley [2] and McCann, Mack and Gale [17] provided evidence that OXT may stimulate PRL secretion by showing a delay in mammary gland involution in lactating rats following injections of OXT. On the other hand, OXT administration has been claimed to inhibit lactation through inhibition of milk ejection and to decrease lactose content of the mammary gland [11]. However, recently Samson, Lumpkin and McCann [28] have demonstrated a physiological role for oxytocin in the control of PRL release. Infusion of anti-oxytocin serum into lactating female rats and into estrogen-primed ovariectomized rats 2 hr before the expected release of endogenous oxytocin, delayed and significantly reduced subsequent PRL surges.

Using cycling female rats, Shani, Urbach, Terkel and Goldhaber [29] observed no stimulation of the release of PRL after a single intra-arterial injection of a physiological dose of oxytocin (0.1 IU). They concluded that since no changes were observed following oxytocin administration, then the possible galactopoietic and mammotropic effects of oxytocin in the rat were not necessarily mediated via prolactin. However, using a higher dose of OXT (4 IU), Lumpkin, Samson and McCann [16] observed a brief surge of PRL release 5 min following OXT administration in male rats. In contrast, a large dose of OXT (10 IU) used by Kuhn, Krulich and McCann [12] appeared to inhibit PRL secretion in lactating females. Lumpkin *et al.* [16], on re-evaluating the controversy surrounding OXT's effect on PRL release, concluded that the dose of OXT administered may be a critical factor in determining its effect on PRL secretion. Lumpkin *et al.* [16] propose that lower doses of OXT administered systemically, such as the 0.1 IU dose used by Shani *et al.* [29], are insufficient to alter PRL secretion. An intermediate dose such as the 4 IU used by Lumpkin *et al.* [16], is sufficient to produce a small degree of stimulation of PRL, while higher doses of OXT, such as the large 10 IU dose used by Kuhn *et al.* [12], appear to inhibit PRL secretion. Lumpkin *et al.* suggest that the reduction in PRL levels following a very large dose of OXT may be attributed to its penetration into the central nervous system in amounts sufficient to exert a "negative ultrashort loop feedback effect" at the level of the hypothalamus. Exogenous OXT thus may suppress the release of endogenous OXT, depriving the anterior lobe of an OXT stimulus for PRL release hence resulting in a decrease in plasma levels of PRL.

Although the dose of OXT administered is obviously a critical factor in PRL release, little information is available regarding the effect of repeated administration of this hormone. This procedure would allow for relatively large amounts of OXT to be administered over a period of time. In the few studies which have considered this issue, an increase in PRL levels has been observed [26,34]. Salisbury *et al.* [26], for example, administered 0.4 IU OXT each hour for 4 hours to ovariectomized rats and found a significant augmentation of the PRL surge 1 hour after the final OXT injection. Administering a much larger dose (1.0 IU every 3 hours for 48 hours), Vaughan *et al.* [34] found a significant rise in PRL in castrated male rats.

Since OXT is implicated in the release of both corticosterone and prolactin, the present experiment was designed to investigate a possible role for OXT in the release of these hormones in a stress situation. A psychological stressor (novelty) was used as we have previously established a substantial release of both hormones following repeated exposure to this stressor [19]. In addition, because previous studies have indicated that the severity of the stressor may be important when examining 11-hydroxycorticosterone (11-OHCS) output from the adrenal cortex, the predictable/unpredictable paradigm used previously by Muir and Pfister [19] was again included. The 11-OHCS response provides a measure of activation of the hypothalamic-pituitary-adrenocortical axis in the animal. Consequently, Muir and Pfister [19] have recently observed a more substantial 11-OHCS output when animals were exposed to a novel apparatus on an unpredictable schedule compared to animals who received predictable exposure to the same novel apparatus.

METHOD

Animals

Ninety-six nulliparous female Wistar rats 90–100 days of age at the start of the experiment were used. Three weeks prior to testing, all animals were individually housed in a fully air conditioned holding room at 22±1°C. A 12:12 hr light/dark cycle was instituted. Food and water were available ad lib.

Glucocorticoid and Prolactin Assays

The glucocorticoid stress response measured was the plasma level of free 11-OHCS, the predominant glucocorticoid response in the rat [15,23]. At the times indicated in the procedure, animals were sacrificed by decapitation. The blood was collected in heparinized tubes and centrifuged to obtain cell-free plasma which was then frozen. One-half of each sample was used for determination of corticosterone levels. Corticosterone levels in plasma were obtained by the fluorometric method of Mattingly [18] which is specific for free plasma 11-OHCS. Plasma prolactin levels were determined by radioimmunoassay [13].

Apparatus

The novelty cage was made of 1 cm wire mesh of 2 mm thick wire with a hinged lid and external dimensions 14.5×20×26 cm. The novelty cage was placed inside the rat holding room described above.

Procedure

The animals were randomly allocated to three groups of 32 rats each: a control group (CON), a predictable stress group (NOV1) and an unpredictable stress group (NOV2). Each of these groups were further subdivided into four treatment groups (n=8): a group which received no injections (NI), a group which received 1 ml/kg injections of the vehicle solution (0.9% saline) (VS), a group which received injections of 5.8 IU OXT/kg (OXT1) and a group which received 11.6 IU OXT/kg (OXT2). All injections were given via the IP route.

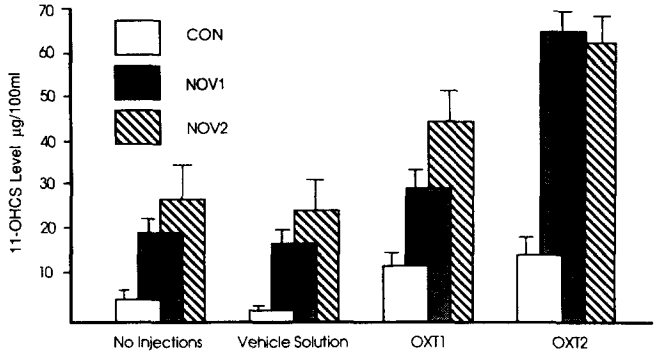

FIG. 1. Effect of various doses of oxytocin on plasma 11-OHCS levels (in µg/100 ml) for CON, NOV1 and NOV2 groups. Values are the mean+S.E.

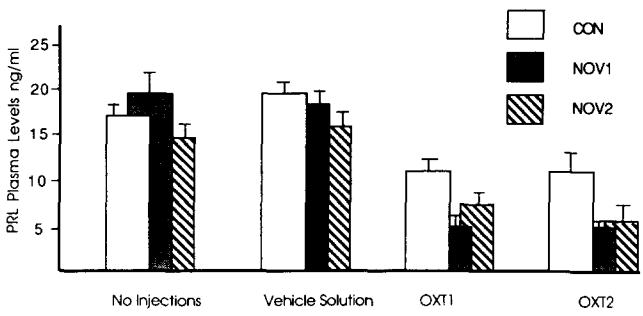

FIG. 2. Effect of various doses of oxytocin on plasma PRL levels (in ng/ml) for CON, NOV1 and NOV2 groups. Values are the mean+S.E.

The two doses of oxytocin were chosen for two reasons. Firstly, King, Brown and Kusnecov [10] have shown that 5.8 IU OXT/kg and 11.6 IU OXT/kg are effective in potentiating the startle response of rats to auditory stimulation. Secondly, Rivier and Vale [25] have shown that doses of 8–24 nmoles OXT/kg are effective in elevating ACTH secretion in freely moving rats. Our doses of 5.8 IU OXT/kg and 11.6 IU OXT/kg equate to approximately 12–14 and 24–28 nmoles OXT respectively.

Animals of the four CON groups were used for determination of plasma 11-OHCS base levels. Other than drug treatment (CON/VS and CON/OXT), all control animals were left undisturbed until plasma collection. As described previously [20] animals allocated to the four NOV1 groups were subjected daily, for five successive days, to a single 30 min exposure to the novel apparatus. For these groups, novelty treatment began each day 2 hr prior to light onset, and thus at the trough of the circadian rhythm in relation to 11-OHCS levels [24]. Injections for NOV1/VS, NOV1/OXT1- and NOV1/OXT2 animals were given immediately prior to each exposure to the novel apparatus. At this time (i.e., 2 hr prior to light onset) injections were also given to animals of the CON/VS, CON/OXT1 and CON/OXT2 groups.

Those animals allocated to the NOV2 condition received four exposures to the novel apparatus on a random schedule (one 30 min exposure to the novel apparatus each day at a randomly selected time within each 24 hr period). However, on the fifth day, novelty treatment for NOV2 animals began 2 hr prior to light onset and therefore at the trough of the circadian rhythm. Once again injections for animals of the NOV2/VS, NOV2/OXT1 and NOV2/OXT2 groups were administered immediately prior to each exposure to the novel apparatus.

Novelty treatment for animals of the NOV1 and NOV2 groups was administered by picking up each rat by the base of the tail and gently placing it in the novel apparatus. Thirty minutes later, the rat was again picked up by the base of the tail and returned to its holding box. After their final manipulation, animals of the NOV1 and NOV2 groups were removed in their novelty cage from the holding room and taken to the preparation room. Blood plasma was collected within 60 sec of removal from the holding room. Similarly, at this time, (2 hr before light onset), the undisturbed control animals (CON) were removed in their boxes to the preparation room where they were sacrificed.

RESULTS

Corticosterone

The mean changes in plasma 11-OHCS levels as a function of oxytocin and stress treatment are shown in Fig. 1. Two-way analysis of variance revealed a significant OXT effect, $F(3,84)=35.0$, $p<0.001$; a significant stress treatment effect, $F(2,84)=57.6$, $p<0.001$; and a significant OXT × stress treatment interaction, $F(6,84)=3.6$, $p<0.01$.

Newman-Keuls post-hoc comparisons on the OXT effect revealed that the OXT injected animals (OXT1 and OXT2) had significantly higher plasma 11-OHCS levels than animals which received no injections and those which received saline injections ($p<0.01$). In addition, OXT2 injected animals responded with significantly higher 11-OHCS levels than animals of the OXT1 group ($p<0.01$). No significant change was observed in the plasma 11-OHCS response between animals which received saline injections compared to those animals which received no injections. Newman-Keuls post-hoc analysis of the stress treatment effect showed that animals of the NOV1 and NOV2 stress groups responded with a significant elevation in 11-OHCS compared to CON animals ($p<0.01$). There was also a significantly larger increase in 11-OHCS levels of NOV2 animals as compared to animals of the NOV1 group ($p<0.05$).

Newman-Keuls multiple comparisons carried out on the OXT × stress treatment interaction revealed that animals of the NOV1/OXT2 and NOV2/OXT2 groups responded with significantly higher 11-OHCS levels than animals of any of the other groups examined ($p<0.01$). Although no significant difference was obtained between the 11-OHCS response of NOV1/OXT2 and NOV2/OXT2 animals, NOV2/OXT1 animals responded with a significantly larger increase in 11-OHCS-than animals of the NOV1/OXT1 group ($p<0.05$).

Prolactin

The mean changes in plasma PRL levels as a function of oxytocin and stress treatment are shown in Fig. 2. Inter-assay reliability for PRL levels was 9.4% while intra-assay reliability was 12%. Two-way analysis of variance revealed a significant OXT effect, $F(3,84)=33.0$, $p<0.001$, and a significant stress treatment effect, $F(2,84)=5.3$, $p<0.01$. The OXT × stress treatment interaction failed to reach significance.

Newman-Keuls post-hoc comparisons on the OXT effect revealed that animals of the OXT1 and OXT2 groups had

significantly lower plasma PRL levels than both animals which received no injection and those which were saline injected ($p<0.01$). There was no significant difference in PRL levels between OXT1 and OXT2 treated animals. In addition, no significant change was observed in plasma PRL levels of animals which received saline injections compared to animals which received no injections. Newman-Keuls post-hoc analysis of the stress treatment effect revealed that both NOV1 ($p<0.05$) and NOV2 ($p<0.01$) animals had significantly lower PRL levels than CON animals. No significant difference in PRL levels were obtained between animals of the NOV1 group and animals of the NOV2 group.

DISCUSSION

Consistent with our previous findings [20], administration of oxytocin produced a marked increase in plasma 11-OHCS levels. This result was observed for both the smaller (5.8 IU/kg) and larger (11.6 IU/kg) dose of oxytocin used in the present study. In contrast, administration of oxytocin produced a marked suppression of the prolactin response in all OXT treated animals.

With respect to corticosterone, a significant interaction between oxytocin and stress treatment was obtained. Animals which received either dose of OXT and were exposed to the novelty apparatus on a predictable schedule (NOV1/OXT1, NOV1/OXT2) responded with a substantially larger 11-OHCS output than animals on the same stress schedule but not receiving OXT treatment. Similarly, administration of oxytocin to animals on an unpredictable stress schedule produced a marked increase in 11-OHCS compared to animals who did not receive oxytocin. This supports the results of previous studies which indicate that OXT potentiates CRF-induced release of ACTH secretion [1,3].

However, this result does not support the findings of Gibbs [7] who reports that anti-OXT antiserum does not attenuate ACTH release to novel environment stress. Gibbs suggests that until OXT levels rise in response to stress, the effects of OXT on ACTH secretion are not expressed. It must be noted, however, that in contrast to the Gibbs study which used a single 3 min exposure to the novel environment, our study employed repeated and substantially longer exposure to the novelty stressor as well as repeated OXT injection. In addition, the present study used female rather than male rats. As pointed out recently [36], sex differences in the OXT response to stress are apparent. Williams *et al.* [36] report that 1 min immobilization produces a marked stimulation of OXT in females whereas in males only a slight OXT response is observed.

Although support for the greater aversiveness of an unpredictable stressor was obtained for the lower dose of oxytocin (OXT1) as measured by the 11-OHCS index, NOV1 and NOV2 animals administered the higher dose of oxytocin (OXT2) responded with equally high 11-OHCS levels. It is therefore apparent that for this larger dose of OXT, a ceiling effect may have been reached in the 11-OHCS response.

In contrast to the increase in 11-OHCS output following OXT and stress treatment, oxytocin produced a pronounced decrease in PRL levels. Regardless which dose of oxytocin was administered, OXT treated animals of all stress groups exhibited a substantial reduction in PRL release. Although this result does not support the previous findings regarding repeated OXT treatment [26,34], inhibition of PRL has previously been reported in lactating females [12] following a single large dose of oxytocin. Lumpkin *et al.* [16] suggest that this reduction in PRL may be attributed to the exogenous OXT suppressing the release of endogenous OXT and thus depriving the anterior pituitary of an OXT stimulus for PRL release. The results of the present study suggest that repeated administration of OXT allows for a relatively large dose of OXT (11.6 IU/kg), comparable to that used by Kuhn *et al.* [12], to be administered over a period of 5 days with a similar effect on PRL release to a single OXT treatment. It is also possible to obtain the same level of suppression of PRL using a much lower dose of oxytocin (5.8 IU/kg).

In addition, there was a trend towards a greater suppression of PRL when the animal had been exposed to the novelty stressor on either stress schedule compared to CON animals. This result is in accordance with earlier findings [9,31] which report a decrease in PRL concentration following intermittent and repeated restraint stress. In the present study, the suppression of PRL following OXT treatment may have been accentuated when exposure to the novelty stressor was also included.

It is therefore apparent from the results of this study that oxytocin has a differential effect on the release of corticosterone and prolactin following psychological stress. While administration of OXT potentiates the 11-OHCS response to stress, it suppresses the PRL response. Although both NOV1 and NOV2 animals responded with equally high 11-OHCS levels following administration of the larger dose, a more substantial increase was obtained for the smaller dose when the stressor was presented on an unpredictable schedule. In contrast, the PRL response was suppressed in all OXT treated animals. However, there was a trend towards a greater suppression of PRL when the animal also received novelty stress. The results of this experiment are not conclusive. However, an important relationship between oxytocin, corticosterone and prolactin release is indicated. Further detailed investigation of this relationship in nulliparous as well as lactating females is required.

REFERENCES

1. Antoni, F. A., M. C. Holmes and M. T. Jones. Oxytocin as well as vasopressin potentiate ovine CRF in vitro. *Peptides* **4:** 411–415, 1983.
2. Benson, G. K. and S. J. Folley. The effect of oxytocin on mammary gland involution in the rat. *J Endocrinol* **16:** 189–201, 1957.
3. Beny, J. L. and A. J. Baertschi. Oxytocin: Major corticotropin-releasing factor secreted from diabetes insipidus rat posterior pituitary in vitro. *Neuroendocrinology* **31:** 261–264, 1980.
4. Gala, R. R. and R. P. Reece. Influence of neurohumors on anterior pituitary lactogen production in vitro. *Proc Soc Exp Biol Med* **120:** 220, 1965.
5. Gibbs, D. M. Dissociation of oxytocin, vasopressin and corticotropin secretion during different types of stress. *Life Sci* **35:** 487–491, 1984.
6. Gibbs, D. M. Immunoneutralization of oxytocin attenuates stress-induced corticotropin secretion in the rat. *Regul Pept* **12:** 273–277, 1985.
7. Gibbs, D. M. Stress-specific modulation of ACTH secretion by oxytocin. *Neuroendocrinology* **42:** 456–458, 1986.
8. Gibbs, D. M., W. Vale, J. Rivier and S. S. C. Yen. Oxytocin potentiates the ACTH-releasing activity of CRF[41] but not vasopressin. *Life Sci* **34:** 2245–2249, 1984.

9. Kawakami, M., T. Higuchi and M. Matsuura. Immobilization stress and prolactin secretion in male rats. *Neuroendocrinology* **29:** 262–269, 1979.
10. King, M. G., R. Brown and A. Kusnecov. An increase in startle response in rats administered oxytocin. *Peptides* **6:** 567–568, 1985.
11. Kuhn, E. R. and S. M. McCann. An inhibitory action of large doses of oxytocin on milk yield in the lactating rat. *Endocrinology* **87:** 1266–1273, 1970.
12. Kuhn, E. R., L. Krulich and S. M. McCann. Influence of exogenously administered oxytocin on prolactin release in the lactating rat. *Neuroendocrinology* **11:** 11–21, 1973.
13. Kwa, H. G. and F. Vorhfstad. Radioimmunoassay of rat prolactin. *Biochim Biophys Acta* **133:** 186–188, 1967.
14. Lang, R. E., J. W. E. Heil, D. Ganten, K. Hermann, T. Unger and W. Rascher. Oxytocin unlike vasopressin is a stress hormone in the rat. *Neuroendocrinology* **37:** 314–316, 1983.
15. LaPlanta, C. C., J. Geroud and J. Stachenko. Lack of appreciable 17-hydroxylase activity in the normal and regenerated rat adrenal tissues. *Endocrinology* **75:** 825–827, 1964.
16. Lumpkin, M. D., W. K. Samson and S. M. McCann. Hypothalamic and pituitary sites of action of oxytocin to alter prolactin secretion in the rat. *Endocrinology* **112:** 1711–1717, 1983.
17. McCann, S. M., R. Mack and C. Gale. The possible role of oxytocin in stimulating the release of prolactin. *Endocrinology* **64:** 870–889, 1959.
18. Mattingly, D. A. A simple fluorometric method for estimation of free 11-hydroxycorticosterone in human plasma. *J Clin Pathol* **15:** 374–379, 1962.
19. Muir, J. L. and H. P. Pfister. Corticosterone and prolactin responses to predictable and unpredictable novelty stress in rats. *Physiol Behav* **37:** 285–288, 1986.
20. Muir, J. L., R. Brown and H. P. Pfister. A possible role for oxytocin in the response to a psychological stressor. *Pharmacol Biochem Behav* **25:** 107–110, 1986.
21. Nicoll, C. S. and J. Meites. Failure of neurohypophyseal hormones to influence prolactin secretion in vitro. *Endocrinology* **70:** 927–929, 1962.
22. Petersen, W. E. Lactation. *Physiol Rev* **24:** 340, 1944.
23. Peron, F. G. The isolation and identification of some adrenocorticosteroids released by the rat adrenal tissue incubated in vitro. *Endocrinology* **66:** 458–469, 1960.
24. Pfister, H. P. and M. G. King. Adaptation of the glucocorticosterone response to novelty. *Physiol Behav* **17:** 43–46, 1976.
25. Rivier, C. and W. Vale. Effects of corticotropin-releasing factor, neurohypophyseal peptides, and catecholamines on pituitary function. *Fed Proc* **44:** 189–195, 1985.
26. Salisbury, R. L., R. J. Kreig and H. R. Seibel. Effects of arginine vasotocin, oxytocin and arginine vasopressin on steroid-induced surges of luteinizing hormone and prolactin in ovariectomized rats. *Acta Endocrinol* **94:** 166–173, 1980.
27. Samson, W. K., M. D. Lumpkin and S. M. McCann. Further correlation of oxytocin and prolactin releases: lactation. *Soc Neurosci Abstr* **9:** 208.5, 1983.
28. Samson, W. K., M. D. Lumpkin and S. M. McCann. Evidence for a physiological role for oxytocin in the control of prolactin secretion. *Endocrinology* **119:** 554–560, 1986.
29. Shani, J., L. Urbach, J. Terkel and G. Goldhaber. Serum prolactin and LH in chronically-cannulated cycling rats after intra-arterial administration of oxytocin. *Arch Int Pharmacodyn* **221:** 323–327, 1976.
30. Silverman, A. J. Ultrastructural studies on the localization of neurohypophyseal hormones and their carrier proteins. *J Histochem Cytochem* **24:** 807–810, 1976.
31. Tache, Y., P. Du Ruisseau, J. R. Ducharme and R. Collu. Pattern of adenohypophyseal hormone changes in male rats following chronic stress. *Neuroendocrinology* **26:** 208–219, 1978.
32. Talwalker, P. K., A. Ratner and J. Meites. In vitro inhibition of pituitary prolactin synthesis and release by hypothalamic extracts. *Am J Physiol* **205:** 213–218, 1963.
33. Vandesande, F., K. Dierick and J. DeMey. The origin of the vasopressinergic and oxytocinergic fibres of the external region of the median eminence of the rat hypophysis. *Cell Tissue Res* **180:** 443, 1977.
34. Vaughan, M. K., D. E. Blask, L. Y. Johnson and R. J. Reiter. The effect of subcutaneous injections of melatonin, arginine vasotocin, and related peptides on pituitary and plasma levels of luteinizing hormone, follicle-stimulating hormone, and prolactin in castrated adult male rats. *Endocrinology* **104:** 212–217, 1979.
35. Weitzman, R. E., R. D. Leake, R. T. Rubin and D. A. Fisher. The effect of nursing on neurohypophyseal hormone and prolactin secretion in human subjects. *J Clin Endocrinol Metab* **51:** 836–839, 1980.
36. Williams, T. D. M., D. A. Carter and S. L. Lightman. Sexual dimorphism in the posterior pituitary response to stress in the rat. *Endocrinology* **116:** 738–740, 1985.