The hormonal response to stress is not modified by the dramatic decrease in prolactin plasma concentration during surgery for microprolactinoma

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Abstract

Objectives—To determine the endocrine response to surgical stress in a homogeneous population of 36 women with microprolactinomas, particularly to evaluate the effect of the sharp decrease in plasma prolactin on stress induced hormonal secretion. In addition, the effects of exogenous opiates on prolactin secretion were studied. **Methods**—The plasma kinetics of cortisol, prolactin, ACTH, GH, and β -endorphin like immunoreactivity (β -ELI) were analysed by including patients operated on with strict anaesthetic and surgical protocols, and by sampling blood every 10 minutes, starting at premedication up to 3 hours after induction.

Results and conclusions—(a) Surgical stress or opiate administration did not induce prolactin release in patients with microprolactinoma. (b) The dramatic decrease in prolactin concentrations have apparently no effect on the release of other hormones involved in stress. (c) The existence of an early GH peak, independently of any surgical procedure, strongly suggests that GH is released by surgical stress whereas β -endorphin is secreted in response to pain. Thus GH may be a useful marker of surgical stress.

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The endocrine response to stress has been extensively studied since the early 1950s.¹⁻³ Most studies in humans, however, have been conducted in patients who were heterogeneous for pathology, type of surgery, or anaesthesia protocol.¹⁻⁷ The endocrine response to surgical stress has been studied more recently with more homogeneous patient groups—namely, premature babies,⁸ neonates,⁹ children,¹⁰ or adults,¹¹ but data on pituitary surgery are sparse.¹² Based on the above findings, the present work had a treble aim: (1) to study the hormonal response, in particular that of the anterior pituitary, and surgical stress in a population highly homogeneous in terms of sex and pathology (women with microprolactinomas) and anaesthesia protocol; (2) to evaluate the immediate effect of the decrease in prolactin concentrations on the concentration of other hormones involved in the stress response such as cortisol, ACTH, GH,

and β -endorphin, by taking frequent samples before, during, and after surgery. Although it has been established that endogenous opioids, in particular β -endorphin, stimulate prolactin secretion,¹³ little is known about the effect of prolactin on β -endorphin secretion in humans; (3) because of previous reports that opiates can induce the release of both glycosylated and non-glycosylated prolactin forms by non-adenoma cells¹⁴ we also investigated the in vivo action of exogenous opiates on prolactin secretion by adenoma cells in these patients.

Patients and methods

Thirty six women between 18 and 44 (mean 29) years of age with a microprolactinoma, as well as 10 healthy women (without endocrine abnormalities and without treatment) between 21 and 39 (mean 31) years of age were included in the study. The diagnostic criteria for microprolactinoma were: (1) a syndrome of amenorrhoea or amenorrhoea-galactorrhoea; (2) a pituitary tumour seen on MRI and not larger than 10 mm at its largest diameter; and (3) basal PRL concentrations higher than 20 μ g/l and not exceeding 250 μ g/l, not stimulated by TRH or metoclopramide. All patients and controls were informed of the aims and methods of the study according to the Helsinki convention and we obtained the agreement of the ethics committee of Timone Teaching Hospital. Exclusion criteria were patients with a mixed adenoma, those with chronic pain, and those presenting a contraindication to anaesthesia or surgery. Those with a previous transphenoidal resection for prolactin secreting adenoma were also excluded. Among the 36, 12 had been taking bromocriptine (2.5 to 5 mg/kg) for 1 to 60 (mean 17) months before surgery. Treatments were stopped 1 to 10 months before the surgery. Twenty four were operated on without a delay.

SURGERY

All the patients were operated on by the same surgeon, via the rhinoseptal route. The operation began at 8 30 am $\pm$ 5 minutes, and duration as measured between the skin incision and the end of resection was 50 $\pm$ 5 minutes.

ANAESTHESIA

Anaesthesia was performed by the same anaesthetist. Premedication was given 30 minutes before induction and comprised 10 mg/kg morniflumate (suppository) and 20 μ g/kg lorazepam (intramuscular). Induction was with

Table 1 Means (SD) of basal concentrations of pituitary hormones and of cortisol measured in 36 patients with microprolactinomas and in 10 healthy women (in patients, samples were collected before premedication, in controls, samples were drawn between 8 00 and 8 30 am)

Hormone	Patients (n=36)	Controls (n=10)	Mann-Whitney test
ACTH (pg/ml)	17/9	20/11	p > 0.05
Cortisol (µg/100 ml)	16.7/4.7	15.3/3.5	p > 0.05
β-Endorphin-like immunoreactivity (pg/ml)	22/5.9	22.6/7	p > 0.05
GH (µg/l)	3.8/0.47	3.2/0.6	p > 0.05
Prolactin (µg/l)	93/44	8/1.5	p < 0.0001

Except for prolactin, there is no significant difference in basal concentrations of stress hormones between patients and controls.

propofol (2.5 mg/kg) and pancuronium bromide (8 mg/kg). After intubation, analgesia was obtained with 20 µg/kg phenoperidine and maintained with N₂O/O₂ (60%/40%). When the transpalatine retractors were installed, 20 µg/kg alfentanil was given.

SAMPLES

A total of 23 samples (each of 3 ml) of venous blood were drawn into EDTA from each patient, using a small catheter. The first sample

was taken before premedication (30 minutes before induction), then every 10 minutes, starting 20 minutes before anaesthesia induction up to 80 minutes after extubation. A final sample was taken 24 hours after extubation. In controls, only one sample was taken, between 8 00 and 8 30 am. Samples were immediately centrifuged at 2500 g for 10 minutes and supernatant liquids were frozen at -80°C before analysis.

ASSAY METHODS

Prolactin was assayed by immunoenzymofluorescence with a Stratus II apparatus (DADE®). Sensitivity was 0.4 ng/ml and the intra-assay coefficient of variation was 4%. Cortisol and GH were assayed by chemiluminescence using an Immulite® (Behring) instrument. Sensitivity and intra-assay coefficient of variation were 4 pg/ml and 6% for GH, and 2 ng/ml and 9% for cortisol. ACTH and β-endorphin-like immunoreactivity (referred to as β-endorphin in the rest of the paper) were assayed by radioimmunoassay as previously

Figure 1 Means (SD) of preoperative, perioperative, and postoperative concentrations of some hormones involved in the response to stress during rhinopalatine route resection of microprolactinomas in 32 women. Samples were taken every 10 minutes starting just before premedication (30 minutes before induction) and up to 80 minutes after extubation. Time 1 is induction, time 2 skin incision, time 3 the installation of transpalatine retractors, time 4 the end of adenoma resection, and time 5 extubation. Note that between time 1 and 2 phenoperidine (20 µg/kg) and at time 3, alfentanil (20 µg/kg) were administered.

Figure 2 Means (SD) of preoperative, perioperative, and postoperative concentrations of GH and prolactin during surgical resection of microprolactinomas in three women in whom the operation was not successful. Samples were taken as in fig 1. Note the absence of a rapid drop after the end of the resection.

described.¹⁵⁻¹⁷ The limit of detection and the intra-assay coefficient of variation were 2 pg/ml and 7% for ACTH, and 3 pg/ml and 9% for β -endorphin. The cross reactions for β -endorphin were 50% with β -lipotropin, <1% with leu-enkephalin and met-enkephalin, and <0.5% with dynorphin 1-17. Other opioid peptides did not interfere with the assay.

STATISTICAL ANALYSIS

The Mann-Whitney *U* test was used to compare basal hormone concentrations in patients and controls, analysis of variance (ANOVA) to compare intraoperative changes in hormone concentrations, and Spearman's rank coefficient to analyse the correlation between β -endorphin, ACTH, and cortisol concentrations. Spearman's correlation coefficient was calculated on the means of β -endorphin, cortisol, and ACTH concentrations.

Figure 3 Preoperative, perioperative, and postoperative changes in the concentrations of cortisol, ACTH, β -endorphin, GH, and prolactin in a case of microprolactinoma surgery. In this patient, for technical reasons, the cutaneous incision was made 30 minutes (not 10 minutes) after induction, by contrast with the other patients. Note that there is an early peak of GH before the surgical procedure.

Figure 4 Regression curve of β -endorphin versus ACTH and cortisol concentrations in 32 patients operated on for microprolactinoma. The curve was plotted after taking into account mean plasma concentrations of ACTH, cortisol, and β -endorphin, measured at nine time points: 30 minutes before induction, at the time of induction, then every 30 minutes up to 180 minutes after anaesthesia induction. The last sample was taken 24 hours after extubation.

Results

Among the 36 patients included in the study, 32 were without recurrence at 186 months, three were not cured. In one patient, for technical reasons, the skin incision was made 30 minutes after induction.

BASAL CONCENTRATIONS

There was no difference in the basal concentrations of GH, ACTH, β -endorphin, and cortisol between patients and controls (table). In patients, the basal concentrations of prolactin were significantly higher (Mann-Whitney, $p < 0.0001$).

KINETICS

For all patients, no increase in prolactin concentrations occurred after giving the two treatments with morphine-like drugs (between time 1 and 2 and at time 3, figs 1, 2, and 3).

In the 32 potentially cured women (fig 1), there was a dramatic decrease in prolactin concentration which was significant as far as 10 minutes after the end of the resection (time 4+10 minutes; $p < 0.001$).

Growth hormone

There was a significant increase in GH concentration that started 10 minutes after the moment of anaesthesia induction (time 2; $p > 0.01$), becoming maximal 30 minutes after induction (fig 1). There was a second and significant increase in GH concentration starting at extubation (time 5; $p < 0.01$), becoming maximal 1 hour after incubation.

β -Endorphin

There was a significant decrease in β -endorphin, 30 minutes after induction (time 3; $p < 0.01$). Inversely, an increase became significant 60 minutes after induction ($p < 0.001$), culminating 15 minutes after extubation (fig 1). Plasma concentrations returned to basal 24 hours after extubation.

Cortisol

Cortisol was significantly lower than basal 20 minutes after the induction (time 1+20 minutes, $p < 0.01$), and the decrease continued until the end of resection. Inversely, there was a significant high cortisol concentration, 30 minutes after the end of the resection (at time 4+30 minutes; $p < 0.01$), culminating 1 hour after extubation and reaching about 1.5 times the basal level. Cortisol returned to basal concentrations 24 hours after extubation.

ACTH

There was a significant low ACTH plasma concentration, 20 and 30 minutes after induction time ($p < 0.01$). Inversely there was a significant increase in plasma ACTH concentration 20 minutes after the end of the resection ($p < 0.001$) with a peak at extubation time (time 5).

In the three women in whom the operation was not successful no fall in prolactin concentration was found until time 4+40 minutes (fig 2). Except for prolactin, hormonal responses to stress were in the same range as the 32 potentially cured patients.

Finally, in the case of the patient whose operation was delayed, the same hormonal response to surgical stress was obtained (fig 3).

Correlations

There were two β -endorphin correlations, one with ACTH (Spearman's $R = 0.95$, $p < 0.001$), the other with cortisol (Spearman's $R = 0.88$, $p < 0.001$, fig 4). Cortisol and ACTH were also correlated ($R = 0.87$; $p < 0.0001$).

Discussion

Prolactin secretion is under dual control—by inhibiting factors, the most important of which is dopamine^{18–20}—and by stimulatory factors. Among the second, it is established that morphine-like compounds^{14–21} such as endogenous opioids,^{18–22} in particular β -endorphin,¹³ stimulate not only prolactin secretion, but also that of GH.²³ Nevertheless, β -endorphin in vitro has no direct effect on prolactin secretion, but antagonises the inhibitory control exerted by dopamine.²² This inhibitory effect on dopaminergic neurons^{24–25} is thought to be exerted via the activation of μ and κ receptors.²⁶

Plasma β -endorphin is secreted by the pituitary stem, whereas the β -endorphin of the CSF arises from the CNS.²⁷ In our study, we have measured the plasma concentration of β -endorphin only every 30 minutes because its half life is longer than the other anterior pituitary hormones.²⁷ At the beginning of the operation, β -endorphin concentrations remained low, undoubtedly because of the administration of exogenous morphine-like drugs that inhibited its secretion from the pituitary.^{9–28} On the other hand, there was an increase in ACTH and β -endorphin concentrations, significant 30 minutes after the transpalatine retractors were installed and continuing in the recovery period. This suggests that either the intensity of pain was sufficient to overcome the negative effect of morphine on CRF secretion or that corticotrophin cells were stimulated by a releasing factor different from CRF. It is known that arginin

vasopressin (AVP) stimulates β -endorphin and ACTH secretion. Interestingly, it has been shown that in sheep hypophyseal portal blood, a moderate stress induces an equal release of CRF and AVP whereas during an intense stress, AVP secretion was much more stimulated than that of CRF.²⁹ As the plasma concentration of β -endorphin increased about 30 minutes after a painful stimulus^{30,31} the pain caused by placing the retractor, despite the concomitant administration of alfentanil, as well as pain stress accompanying awakening, could explain the kinetics obtained in our study. Comparable kinetic results have been published for heart surgery⁹ and pituitary surgery.¹²

The perioperative increases in prolactin and GH in response to stress have been known for a long time.⁷ In the case of the existence of a prolactin adenoma, however, the administration of opiates does not cause release of prolactin, as reported in the case of non-adenoma cells.¹⁴ In addition, the dramatic decrease in prolactin concentrations during the operation had apparently no effect on the kinetics of other hormones involved in stress. Indeed the same type of curves were obtained in the three women in whom the operation was not successful, as shown by the absence of a rapid fall in plasma prolactin concentrations.

Concerning GH, the first peak was considered to be due to induction of anaesthesia, which generates intense stress,¹² and not by pain for the following reasons: (1) in the general case, GH concentrations increased twofold before any surgical procedure; (2) in the case of the patient whose operation was delayed after induction, there was a GH peak before surgery, the amplitude of which was comparable with that seen in the other patients. These findings suggest that the first GH peak was related to induction stress alone. The second GH peak occurred at the same time as consciousness was regained, about 40 minutes after the β -endorphin peak. As it is impossible to dissociate pain from the stress of awakening at that time, it is not possible to determine if the second peak was caused by pain, by stress, or by both at the same time. Because ACTH and β -endorphin are cosecreted, the differential timing of secretion in GH and β -endorphin is also found between GH and ACTH. This is seen not only during surgical stress but also during dynamic tests of pituitary function. Thus it is not possible to conclude that the underlying stress is different for the two groups of hormones. However, as GH secretion is not modified by exogenous opioids during anaesthesia, measurement of GH may be a useful marker of stress during surgery for microprolactinoma.

The response of the adrenal cortex to surgical stress is attenuated by the administration of morphine-like drugs,^{8,9} perhaps via the inhibition of release of CRF^{32,33} and of ACTH.³⁴ By contrast, morphine antagonists stimulate ACTH secretion in animals^{35,36} and in humans³⁷ suggesting the existence of an inhibitory tonic control exerted by endogenous opioids on the anterior pituitary.^{32,37,38} These effects could be

secondary to central or peripheral mechanisms, but also to a direct action of opioids on endocrine glands.³⁹ The increase in circulating cortisol concentrations at the time of awakening in the present study, although moderate, was significant and confirms prior findings.^{11,40} This postoperative increase may be attributed to the disinhibition of the corticotrophic axis during drug withdrawal. However, the increase in cortisol during surgical stress is probably an epiphenomenon because the increase is too small to have strong biological effects.⁴¹ It was not possible to include an adequate control group in this study. As we have not included a surgical control group, the responses measured may or may not be specific for pituitary surgery or for the patients with hyperprolactinaemia, but this was not the aim of our study.

Hormonal responses to surgical stress have been studied in both general anaesthesia¹⁴ and epidural or peridural anaesthesia.^{10,42} The recent appearance of endoscopic surgical techniques had raised hopes for reduced surgical stress in comparison with conventional surgery, but this hope has been dashed by the comparative study of the main markers of stress during different types of anaesthesia.^{42,43}

Finally, the existence of a correlation between β -endorphin and ACTH concentrations has been known for almost 20 years.⁴⁴ This correlation is apparently unchanged by the presence of a prolactin tumour.

Our conclusions are summarised thus:

(1) During prolactinoma surgery, the existence of an early GH peak at the anaesthesia induction time, independently of any surgical procedure, strongly suggests that GH is a preferential marker of surgical stress, whereas β -endorphin is secreted in response to pain.

The fact that GH is a marker of surgical stress has been established in many surgical states including pelvic surgery.⁴⁵ Our results strongly suggest that this is also true during surgery for microprolactinoma. As it was reported in children that the increase in stress hormone concentrations during surgical procedures is associated with postoperative metabolic complications,⁸ it will be interesting to study the relation between high GH concentrations and postoperative complications.

(2) The rapid fall in prolactin concentrations has no effect on the release of other hormones involved in stress.

(3) The administration of opiates does not cause the release of prolactin, as previously was reported in vitro, from non-adenoma cells.

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