Effects of Bromocriptine and Cyproheptadine on Basal and Corticotropin-Releasing Factor (CRF)-Induced ACTH Release in a Patient with Nelson's Syndrome

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Abstract

The effects of bromocriptine, a dopamine agonist, and cyproheptadine, a serotonin antagonist, on basal and corticotropin-releasing factor (CRF)-stimulated ACTH release were investigated in a 40-year-old female patient with Nelson's syndrome.

Oral administration of either bromocriptine (2.5 mg) or cyproheptadine (8 mg) caused a marked drop in plasma ACTH levels. Intravenous administration of synthetic ovine (o) CRF (50 μg) produced an exaggerated response of plasma ACTH. Short-term (3-week) treatment with either bromocriptine (7.5 mg/day) or cyproheptadine (12 mg/day) resulted in a marked suppression of basal ACTH release. Furthermore, a blunted response of plasma ACTH to oCRF was observed after short-treatment with either drug. However, after a longer period of treatment with cyproheptadine (18-week), plasma ACTH levels rose again and hyperresponsiveness to oCRF was restored to the pretreatment levels.

These data indicate that synthetic oCRF is a potent secretagogue for ACTH release in a patient with Nelson's syndrome. It is suggested that bromocriptine and cyproheptadine are effective drugs in reducing basal and CRF-stimulated ACTH release, possibly acting at the pituitary level in this case. However, the apparent refractoriness after chronic treatment with cyproheptadine may limit its therapeutic use in the present case.

Progressive hyperpigmentation of the skin, increased concentrations of plasma ACTH, and occurrence of a pituitary tumor after bilateral adrenalectomy in patients with Cushing's disease were first described by Nelson et al. (1958) and by Salassa et al. (1959). Nelson's syndrome occurs in 10–35% of patients who have undergone total adrenalectomy for Cushing's disease (Moore et al., 1976; Cohen et al., 1978). The mechanism of the occurrence of the pituitary tumor in these patients remains undetermined. Most patients with Cushing's disease appear to have an ACTH-secreting microadenoma in the pituitary (Tyrrell et al., 1978; Bigos et al., 1980; Kuwayama et al., 1981) which may lead to an increase in tumor growth following total adrenalectomy.

The primary structure of corticotropin-
releasing factor (CRF) from ovine (o) hypothalami has been determined by Vale et al. (1981). The native oCRF and its synthetic counterpart have been shown to be fully active in vitro as well as in vivo in stimulating ACTH release from the pituitary of animals (Vale et al., 1981; Rivier et al., 1982; Vale et al., 1983; Donald et al., 1983; Schulte et al., 1982) and humans (Grossman et al., 1982; Orth et al., 1983; Müller et al., 1982; Nakahara et al., 1983).

The present study was designed to study the effect of synthetic oCRF on release of ACTH in a patient with Nelson’s syndrome, and to investigate the effects of bromocriptine and cyproheptadine on basal and CRF-induced ACTH release.

**Patient and Methods**

A 40-year-old female was diagnosed as having Cushing’s disease in 1972. Radiographs of the sella turcica were normal. Bilateral adrenalectomy was performed in 1972 with subsequent maintenance on hydrocortisone (20 mg/day). Progressive hyperpigmentation of the skin was noted in 1979, associated with increased plasma concentrations of ACTH which ranged from 190–625 pg/ml. In 1980, plasma ACTH levels exceeded 1000 pg/ml. Visual field examination and X-ray films of the sella turcica were normal. In July, 1982, she was admitted for further evaluation of endocrine function. CT scanning of the brain revealed neither deformity of the sella turcica nor abnormalities in the pituitary. Results of radiographical examination of the chest and the abdomen were normal.

The following tests were performed at 0900 after bed rest for at least 30 min and the hydrocortisone replacement (20 mg) was withheld one day before each testing; lysine vasopressin (LVP, 10 U im., Sandoz), LHRH (100 µg, iv., Daiichi), TRH (500 µg, iv., Tanabe), regular insulin (0.1 U/kg body weight, iv., Shimizu), cyproheptadine (8 mg, p.o.) and bromocriptine (2.5 mg, p.o.). Synthetic oCRF (50 µg, Protein Research Foundation, Osaka, Japan) sterilized and dissolved in 5 ml saline, was administered iv. as one bolus. Similar CRF tests were performed after 3 weeks treatment with bromocriptine (7.5 mg/day) and 3 and 18 weeks treatment with cyproheptadine (12 mg per day).
mg/day). No side-effects were observed during or after oCRF administration. Heparinized plasma was immediately separated by centrifugation at 4°C and stored at −20°C until assayed.

Plasma ACTH and β-endorphin (EP) were extracted with silicic acid and measured by specific RIAs as previously reported (Hirata et al., 1975; Yoshimi et al., 1978). The antigenic determinant of the anti-ACTH serum was the 18–24 amino-acid sequence of the ACTH molecule, and that of the anti-β-EP serum was the 17–31 amino-acid sequence of the β-EP molecule, which reacted equimolarly with β-lipotropic hormone (LPH).

For molecular sieving, aliquots of the plasma extracts were applied on a Sephadex G-50 fine (Pharmacia) column (0.9 × 56 cm) equilibrated and eluted at 4°C with 50 mM phosphate buffer, pH 7.4, containing 500 KIU/ml aprotinin (Bayer) and 0.5% human serum albumin. 1 ml-fractions were collected.

Results

Basal plasma concentrations of ACTH and β-EP were extremely elevated, ranging from 1000 to 1500 pg/ml (normal 20–110 pg/ml) and 300 to 600 pg/ml (normal 10–50 pg/ml), respectively. High-dose (8 mg) dexamethasone only reduced plasma ACTH levels from 967 pg/ml to 147 pg/ml. Administration of LVP caused a four-fold increase in plasma levels of both ACTH and β-EP over the basal levels, whereas insulin-induced hypoglycemia showed no changes despite more than a 50% fall of blood glucose from the control level. Administration of LHRH, but not TRH, paradoxically caused a three-fold rise in plasma ACTH and β-EP levels above the control. Administration of cyproheptadine caused an acute reduction in plasma ACTH at 60 min (16% of basal value), while administration of bromocriptine caused a slow and modest reduction (36% of basal value) in plasma ACTH (Fig. 1). Both basal and provocation-stimulated secretion of gonadotropins, thyrotropin, growth hormone and prolactin was normal.

Administration of synthetic oCRF caused an exaggerated response (about a sixfold increase in plasma ACTH over the basal levels) compared to the responses to oCRF in normal subjects (Fig. 2). No significant

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**Fig. 2. Effect of ovine (o) corticotropin-releasing factor (CRF) on ACTH release in Nelson’s syndrome and in normal subjects.**

Synthetic oCRF (50 μg) was administered intravenously. (Top) Plasma ACTH responses to oCRF (●-●) and saline (○-○) in Nelson’s syndrome are shown. (Bottom) The mean concentrations of plasma ACTH with the standard errors by shaded areas by oCRF (50 μg) in 7 normal subjects are shown for comparison.
fluctuation in plasma ACTH was observed during saline infusion on a control day. After treatment for 3 weeks with bromocriptine, the basal plasma ACTH levels (149–213 pg/ml) were significantly lowered. Furthermore, oCRF-stimulated ACTH release was markedly diminished after bromocriptine treatment (Fig. 3). Treatment with cyproheptadine for 3 weeks also reduced both the basal plasma ACTH levels (390–620 pg/ml) and the response to oCRF (Fig. 3). However, the basal plasma ACTH levels (877–980 pg/ml) again rose after 18 weeks of cyproheptadine treatment. Administration of oCRF induced an exaggerated response of plasma ACTH similar to that before treatment (Fig. 3).

Gel filtration of the plasma extracts revealed that the major component of ACTH and β-EP coeluted with standard ACTH and β-LPH standard, respectively, while a minor component with both ACTH and β-EP immunoreactivities eluted in the void volume (Fig. 4).

Discussion

Although the patient had progressive skin pigmentation and increased plasma levels of ACTH after bilateral adrenalectomy for Cushing's disease, there was no evidence of an overt pituitary tumor. Differential diagnosis between Nelson's syndrome and the ectopic ACTH syndrome is sometimes difficult; some patients with an occult ectopic ACTH-producing tumor may present clinical and endocrine features indistinguishable from Nelson's syndrome (Flint and Jacobs, 1974). However, it seems unlikely that this patient had the ectopic ACTH-producing tumor. Radiological studies revealed no abnormal mass in the lung or the abdomen. Furthermore, gel filtration of the patient’s plasma revealed a predominance of ACTH- and β-LPH-sized component, whereas the ectopic ACTH-producing tumors secrete principally high molecular weight ACTH (“big ACTH”) and ACTH fragments (Orth et al., 1973; Gewirtz and Yalow, 1974). Thus, hypersecretion of ACTH in this case probably originates from the pituitary. Selective venous sampling of ACTH from the inferior petrosal sinus should provide a more reliable aid to differentiate these two entities (Foulding et al., 1981; Tanaka et al., 1984).

The endocrine features in this case also appear to be consistent with those of Nelson's syndrome; the plasma ACTH response to LVP, the paradoxical response to LHRH and the suppressive effect of both bromocriptine and cyproheptadine are often found in some patients with Cushing's disease or Nelson's syndrome (Krieger et al., 1975; Krieger et al., 1976; Krieger et al., 1977; Lamberts et al., 1977; Lamberts et al., 1980; Pieters et al., 1982). The failure of plasma ACTH response to insulin-induced hypoglycemia in this case would not be of any help in the diagnosis because various responses to hypoglycemic stress have been reported in Nelson's syndrome (Krieger and Luria, 1977; Imura et al., 1980).

oCRF is a potent secretagogue for ACTH release in human, and is a useful diagnostic tool in patients with hypothalamic-pituitary-adrenal disorders (Grossman et al., 1982; Müller et al., 1982; Nakahara et al., 1983). The results of hyperresponsiveness of plasma ACTH to oCRF in our case (Fig. 2) are consistent with those reported by others (Müller et al., 1982; Nakahara et al., 1983). Therefore, it is suggested that ACTH release from the pituitary tumor in Nelson's syndrome may not be autonomous but still regulated by hypothalamic CRF.

It has been reported that bromocriptine, a dopamine agonist, and cyproheptadine, a serotonin antagonist, are effective in reducing ACTH release, and thus could be used for treatment in some patients with Cushing's disease or Nelson's syndrome (Krieger et al., 1975; Krieger et al., 1976; Benker et al., 1976; Lamberts et al., 1977; Lamberts et al.,
Fig. 3. Effects of bromocriptine and cyproheptadine on oCRF-induced ACTH release in Nelson's syndrome.

After 3 weeks treatment with bromocriptine (7.5 mg/day, ●-●) or cyproheptadine (12 mg/day, ○-○), and 18 weeks treatment with cyproheptadine (Δ-Δ), plasma ACTH responses to oCRF (50 μg) are shown.

Fig. 4. Sephadex G-50 gel exclusion chromatography of plasma extracts from Nelson's syndrome.

Concentrations of immunoreactive ACTH (●-●) and β-EP (○-○) in the eluates are shown. Recovery of ACTH and β-EP from the column was 80% and 95%, respectively. Calibration standards are indicated by arrows (Vo: void volume, Vt: total volume).
The present study clearly shows a marked suppressive effect of both drugs on the basal ACTH release (Fig. 1). Furthermore, short-term (3-week) treatment with either drug resulted in a marked reduction in basal and CRF-induced ACTH release (Fig. 3). It seems unlikely that this effect may be due to spontaneous fluctuation in ACTH release or exogenous hydrocortisone because no significant variation in plasma ACTH was observed on the control day (Fig. 2).

The site(s) on which these drugs act remains undetermined. Changes in neurotransmitters in the central nervous system, such as an increase in serotonin content and/or depletion of dopamine content may be involved in the pathogenesis of Cushing's disease (Krieger et al., 1975; Lamberts et al., 1977); modulation of the hypothalamic neurotransmitters by cyproheptadine or bromocriptine may cause a reduction in ACTH release, and thus lead to clinical remission in some patients with Cushing's disease and Nelson's syndrome. Indeed, serotonin increases CRF secretion from rat hypothalamic fragments incubated in vitro (Jones & Hillhouse, 1977), whereas serotonin itself has no effect on ACTH release from the pituitary (Lamberts et al., 1980; Gillies et al., 1980; Mashiter et al., 1980, Ishibashi et al., 1981). Cyproheptadine and bromocriptine both reduce ACTH release from pituitary adenomas causing Cushing's disease in vitro (Lamberts et al., 1980; Adams et al., 1981; Ishibashi et al., 1981; Suda et al., 1983), suggesting a direct action of either drug at the pituitary level. The suppressive effect on both basal and CRF-induced ACTH release by cyproheptadine and bromocriptine in this case suggests that both drugs exert their effect directly on the pituitary rather than by reducing hypothalamic endogenous CRF.

The failure of the suppressive effect on basal and CRF-induced ACTH release after a longer period of treatment with cyproheptadine (18-week) may be in accordance with the fact that relapse occurs in patients with Cushing's disease even after a successful long-term treatment with this drug (Krieger, 1979). This phenomenon may be accounted for by the refractory responsiveness of the drug after chronic treatment. However, the results of this study of a single patient do not make it possible to conclude that both cyproheptadine and bromocriptine are effective in treating Nelson's syndrome. Further study should be done on a wide range of patients with Nelson's syndrome.

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References


