Alterations in anterior pituitary function of dogs with pituitary-dependent hyperadrenocorticism

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Abstract
For the purpose of obtaining an integral picture of anterior pituitary function in canine pituitary-dependent hyperadrenocorticism (PDH), 47 dogs with PDH and eight control dogs received combined administration of four hypophysiotropic hormones (CRH, GHRH, GnRH and TRH) and measurements were made of ACTH, cortisol, GH, LH, PRL and TSH.

Basal plasma levels in 47 dogs with PDH were higher for ACTH, cortisol and PRL, lower for GH, and not different for LH (n=25 noncastrated dogs) and TSH compared with controls (n=8). In dogs with PDH the responses to combined hypophysiotropic stimulation, measured as increment and area under the curve (AUC), were not different for ACTH, lower for GH and TSH (increments and AUC) and higher for cortisol (increments), LH (AUC, n=25 noncastrated dogs) and PRL (increments and AUC) than in controls.

We conclude that pituitary function is altered in several respects in dogs with PDH. 1) In spite of persisting hypercortisolemia and the neoplastic transformation of the corticotropic cells, these cells usually remain responsive to combined hypophysiotropic stimulation. 2) Basal plasma GH concentrations and GH responsiveness in the combined stimulation test are decreased, probably as a result of the glucocorticoid-induced increase in somatostatin tone. 3) Plasma PRL concentrations and the PRL response to stimulation are increased, probably as a result of cosecretion with ACTH by the transformed corticotropic cells. 4) Despite the well known effect of glucocorticoids of decreasing circulating concentrations of gonadal steroids and thyroxine, the basal plasma concentrations of LH and TSH remain unchanged and there is a tendency to hyperresponsiveness to stimulation for LH and hyporesponsiveness for TSH. The most likely explanation for these changes is a dual effect of glucocorticoids: a direct effect on the gonads and thyroids and/or the transport and metabolism of their secretory products, and an influence on the sensitivity of the feedback control at the hypothalamic–pituitary level.

Introduction
Pituitary-dependent hyperadrenocorticism (PDH) is known to be associated with several alterations in the release of pituitary hormones. Apart from the principal abnormality, the unrestrained release of adrenocorticotropin (ACTH) and possibly additional peptides derived from pro-opiomelanocortin (POMC) (Ray et al. 1995), in humans the release of the other pituitary hormones is often also affected. These changes are believed to be primarily due to the glucocorticoid effect per se (Cuerda et al. 1991, Marazuela et al. 1993). In addition, there is evidence that the primary lesion itself may give rise to compression of portal vessels, resulting in interruption of the delivery of hypothalamic hormones (Arafah et al. 1994). This seems to apply to the function of the posterior pituitary also. There is general agreement that glucocorticoids inhibit the release of vasopressin in dogs and in humans (Biewenga et al. 1991, Raff 1993). Moreover, it is well known that expansion of corticotropic adenomas may lead to neurohypophysial dysfunction and vasopressin deficiency, as has been established in dogs with hyperadrenocorticism (Biewenga et al. 1989). With regard to the anterior pituitary, there are several reports on separate studies in humans that were concerned with the release of one or more of the non-POMC derived peptides. Taken together, these observations appear to suggest that the glucocorticoid excess causes impaired release of growth hormone (GH), thyrotropin (TSH) and luteinizing hormone (LH) (Marazuela et al. 1993, Giustina et al. 1994, Leal-Cerro et al. 1994, Magiakou et al. 1994). The basal plasma levels of prolactin (PRL) tend to be increased, and there is a blunted response to hypophysiotropic stimulation (Cuerda et al. 1991). Most of the explanations of the results have concentrated on effects of glucocorticoids at the hypothalamic–pituitary level, although some of
the observations seem to conflict with this concept (Cuerda et al. 1991, Marazuela et al. 1993, Leal-Cerro et al. 1994).

In order to obtain an integral picture of anterior pituitary function in dogs in situations of pituitary-dependent hyperadrenocorticism, we used a combined anterior pituitary (CAP) function test, applying stimulation with four hypothalamic releasing hormones and measuring five pituitary hormones (Meij et al. 1996a, b). These studies were performed in dogs, a species in which pituitary-dependent hyperadrenocorticism has many similarities with Cushing’s disease in humans (Kemppainen & Peterson 1994).

Materials and Methods

Animals

The dogs with PDH had a median age of 10 years (range 6–14 years) and comprised 26 females (16 spayed) and 21 males (six castrated). Healthy male dogs (n=8, median age 2 years, range 1–6 years) were used as a control group. The anterior pituitary function test was performed between 0800 and 1200 h after an overnight fast.

The experimental protocols were approved by the Institutional Committee on Animal Care and Use, Faculty of Veterinary Medicine, Utrecht University, The Netherlands.

The diagnosis of hyperadrenocorticism was based upon increased (>10 × 10⁻⁶) corticoid/creatinine (C/C) ratios in two consecutive morning urine samples (Rijnberk et al. 1988a). Differentiation between PDH and hyperadrenocorticism due to an adrenocortical tumour was accomplished by administering, after the collection of the second urine sample, three oral doses of dexamethasone 0·1mg/kg body weight at 8-h intervals. When the C/C ratio in the third urine sample was less than 50% of the mean of the first two samples, the dog was categorized as being responsive to dexamethasone and PDH was diagnosed. In 11 dogs in which there was less than 50% suppression of the C/C ratio (the dexamethasone-resistant animals), the diagnosis of PDH was confirmed by at least five measurements of basal plasma ACTH (Rijnberk et al. 1987), visualization of the adrenals by ultrasonography (Voorhout et al. 1990) and computed tomography (CT) of the pituitary gland (Voorhout et al. 1988, Voorhout 1990).

Forty-five dogs had pituitary glands which were of normal size or moderately enlarged; only two of the 47 dogs had pituitary tumours which measured more than 10 mm in height. In all dogs, the baseline urinary C/C ratios exceeded the upper limit of the reference range (10 × 10⁻⁶). The C/C ratios in 47 dogs with PDH ranged from 10⁻⁶ to 275 × 10⁻⁶ (median 56·0 × 10⁻⁶). In all dogs plasma ACTH values (≥50 ng/l) were consistent with PDH. The basal plasma ACTH concentration in 47 dogs with PDH ranged from 51·0 to 1063·3 ng/l (median 163·3 ng/l). Data from eight healthy dogs, studied previously (Meij et al. 1996a, b), were used as control values.

Anterior pituitary function test

Anterior pituitary function was investigated by rapid sequential intravenous administration of four hypothalamic releasing hormones, as described by Meij et al. (1996a, b). Ovine corticotropin-releasing hormone (CRH) (oCRF, Peninsula Laboratories Inc., Belmont, CA, USA) and human GH-releasing hormone (GHRH) (hGHRF, Peninsula Laboratories Inc.) were both stored frozen at −25 °C and thawed at room temperature immediately before use. The gonadotropin-releasing hormone ( GnRH) analog, gonadorelin (Fertagyl, Intervet, Boxmeer, The Netherlands) and thyrotropin-releasing hormone (TRH; Hoffman-La Roche, Basel, Switzerland) were stored at 4 °C. An intravenous catheter was placed in the cephalic vein of each dog to facilitate rapid sequential injections. In this CAP test, all four releasing hormones were injected intravenously within 30 s, immediately after the collection of the zero blood sample from the jugular vein. The releasing hormones were injected in the following order and doses: CRH 1µg/kg, GHRH 1µg/kg, GnRH 10µg/kg and TRH 10µg/kg. The clock for timing of blood sampling was started immediately after the administration of the last releasing hormone. Blood samples were collected by jugular vein puncture at −30, −15, 0, 5, 10, 20, 30, 45, 60, 90, and 120 min (−15 to 45 min for plasma TSH) after injection and were placed in ice-chilled EDTA-coated plastic tubes. The samples were centrifuged at 4 °C for 10 min and plasma was stored at −25 °C until assayed for cortisol, ACTH, GH, LH, PRL and TSH. Basal plasma total thyroxine and α-MSH were also measured.

Hormone determination

Plasma ACTH was measured by RIA without extraction, according to the procedure described by Arts et al. (1985) and validated for the dog. Antiserum was obtained from IgG Corp. (Nashville, TN, USA). The tracer was purchased from International CIS (St Quentin-Yvelines, France), and the standard was obtained from the NIH (Bethesda, MD, USA). The intra- and interassay coefficients of variation were 8% and 12%, respectively, and the limit of detection was 10 ng/l. Plasma cortisol was measured by RIA (Rijnberk et al. 1988a, b). The limit of detection for cortisol was 1 nm and the intra- and interassay coefficients of variation were 6% and 8%, respectively. Plasma GH was measured in a homologous RIA (Eigennmann & Eigenmann 1981). The intra- and interassay coefficients of variation were 3·8% and 7·2%, respectively, and the sensitivity of the assay was 0·4 µg/l plasma. The degree of cross-reaction of canine prolactin was 2%.

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Plasma LH was measured using a heterologous RIA (Nett et al. 1975). A rabbit antiserum raised against ovine LH (CSU-204), radioiodinated NIAMDD-bLH-4 and canine pituitary standard LER 1685–1 were used in this assay. The limit of detection for LH was 0·31 µg/l and the intra- and interassay coefficients of variation were 2·3% and 3·5%, respectively. In accordance with a previous study (Rijnberk et al. 1988a), the dogs were divided into a group with PDH of presumably pars distalis origin (nonelevated basal plasma α-MSH concentrations, ≤60 ng/l) and a group with PDH of presumably pars intermedia origin (elevated basal plasma α-MSH concentrations, >60 ng/l).

Calculation and statistics

Results are expressed as means ± s.e.m. Mean basal levels were calculated from the −30, −15, and 0 min values in the anterior pituitary function test. The areas under the curve (AUC) of the hormone concentrations in the stimulation tests were calculated by the trapezoidal method after subtraction of the mean basal level. Increments in the plasma concentrations in the stimulation tests were calculated as the difference between peak levels and mean basal levels. Differences between the control dogs (n=8) and the PDH dogs (n=47) were analysed by the Mann–Whitney U–Wilcoxon rank sum test (two-tailed) because of the different group sizes. After division of the dogs into a noncastrated PDH group (n=25) and a castrated PDH group (n=22), differences between noncastrated PDH dogs, castrated PDH dogs and control dogs were first analysed by Kruskal–Wallis one-way ANOVA and, if significant differences were found, these were further analysed by the Mann–Whitney U–Wilcoxon rank sum test. The same procedure was followed after division of the dogs into a group with normal (n=37) and a group with elevated (n=10) basal plasma α-MSH concentrations.

Results

Basal plasma levels of ACTH, cortisol and PRL were significantly higher and basal plasma GH levels significantly lower in PDH dogs than in control dogs (Table 1, Fig. 1a–c and e). As expected, the basal LH values in the 22 castrated animals (21·6 ± 3·5 µg/l) (with no differences for males and females) were significantly higher (P<0·00005) than values in the 25 noncastrated PDH dogs (5·0 ± 0·7 µg/l) and in eight male healthy beagle dogs (5·3 ± 0·8 µg/l) (Table 1, Fig. 1d). Hyperprolactinemia

### Table 1

<table>
<thead>
<tr>
<th></th>
<th>Basal</th>
<th>AUC (0–120 min)</th>
<th>Increment</th>
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<tr>
<td></td>
<td>PDH (ng/l)</td>
<td>Control (ng/l)</td>
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<tr>
<td>ACTH (ng/l)</td>
<td>205·4 ± 26·0b</td>
<td>58·9 ± 5·1</td>
<td>11·00 ± 3·00</td>
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<td>Cortisol (mm)</td>
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<td>104·3 ± 20·1</td>
<td>30·15 ± 5·22</td>
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<td>LH (µg/l)</td>
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<td>1·7 ± 0·3</td>
<td>0·28 ± 0·06a</td>
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<td>1·23 ± 0·18d</td>
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<td>Castrated (n=22)</td>
<td>21·6 ± 3·5c</td>
<td></td>
<td>0·66 ± 0·13c</td>
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<tr>
<td>PRL (µg/l)</td>
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<td>1·4 ± 0·1</td>
<td>3·32 ± 0·61c</td>
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<tr>
<td>TSH (µg/l)</td>
<td>0·21 ± 0·03</td>
<td>0·17 ± 0·03</td>
<td>0·01 ± 0·00d</td>
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</table>

*P<0·00005, bP<0·0005, aP<0·005, dP<0·05 (Mann–Whitney U–Wilcoxon rank sum test) compared with control values. *P<0·00005, fP<0·005 (Mann–Whitney U–Wilcoxon rank sum test) compared with values in noncastrated dogs with PDH. *AUC (0–45 min).
was present in most dogs with PDH (Table 1, Fig. 1e), but there were no significant differences in the basal plasma PRL levels between the sexes, either noncastrated or castrated. The mean plasma thyroxine concentration in the PDH dogs (14·3 ± 0·9 nm) was significantly lower than in the normal dogs (30·3 ± 3·3 nm) (Mann–Whitney, \( P=0·0001 \)), whereas basal plasma TSH levels were not significantly different (Table 1, Fig. 1f). There were 37 PDH dogs with nonelevated (≤ 60 ng/l) basal plasma α-MSH levels (21·7 ± 2·0 ng/l) and 10 PDH dogs with elevated (>60 ng/l) plasma α-MSH levels (415·0 ± 94·2 ng/l). There were no significant differences in basal plasma ACTH, cortisol, GH, LH, PRL or TSH values between the two groups.

No side effects were observed after the combined administration of releasing hormones in the dogs with PDH. Administration of four releasing hormones in 47 dogs with PDH resulted in prompt increases in plasma ACTH, cortisol, GH, LH, PRL and TSH concentrations (compared with values in control dogs), the maximum plasma ACTH, cortisol, GH, LH, PRL and TSH concentrations were (mean ± s.e.m.): ACTH, 469·7 ± 54·8 ng/l at 5 min (277·3 ± 42·7 ng/l at 5 min) (Fig. 1a); cortisol, 803·7 ± 63·9 nm at 20 min (355·0 ± 38·4 nm at 30 min) (Fig. 1b); GH, 5·1 ± 0·9 µg/l at 20 min (12·6 ± 2·5 µg/l at 20 min) (Fig. 1c); LH, 37·0 ± 4·9 µg/l in 25 noncastrated dogs at 20 min (22·0 ± 2·3 µg/l at 10 min) (Fig. 1d); PRL, 113·0 ± 17·8 µg/l at 5 min (27·3 ± 7·9 µg/l at 5 min) (Fig. 1e); and TSH, 0·59 ± 0·05 µg/l at 20 min (0·85 ± 0·17 µg/l at 30 min) (Fig. 1f). In the dogs with PDH, mean plasma ACTH, cortisol, GH and PRL levels had not returned to baseline values at 120 min, whereas the mean plasma LH had reached baseline by 120 min (Fig. 1).

In more general terms, the responses to combined hypophysiotropic stimulation were quite different for the respective pituitary hormones. The plasma ACTH curve in the dogs with PDH was almost an exact replica of the curve in the control dogs, although it had shifted about 150 ng/l higher (Fig. 1a). The increment for cortisol in the dogs with PDH was significantly higher than in the control dogs (Table 1, Fig. 1b). The increments and AUC for plasma GH and TSH were significantly lower, and those for plasma PRL significantly higher, in the dogs with PDH than in the control dogs (Table 1, Fig. 1c, e and f). The data for the increments in plasma LH were not significantly different between the noncastrated PDH dogs and noncastrated controls, but the apparent difference between the mean data suggests that, with larger and more equal group sizes, a real difference might become apparent (Table 1). The AUC for plasma LH in noncastrated PDH dogs was significantly higher than in castrated PDH dogs and noncastrated male control dogs (Table 1, Fig. 1d).

**Discussion**

The CAP test is a dynamic test of pituitary function in humans, and is used as a screening test in suspected pituitary dysfunction and for assessment of pituitary function after pituitary surgery (Thorner et al. 1992). It has been recommended instead of the traditional triple test consisting of insulin, TRH and GnRH, especially in patients predicted to have pituitary–adrenocortical insufficiency (Oki et al. 1993). It has proved to be a rapid, safe and reliable test in normal subjects (Holl et al. 1985a, b, Sheldon et al. 1985, Cohen et al. 1986, Schopohl et al. 1986, Kogure 1987, Bando et al. 1989) and patients with endocrine disorders (Cohen et al. 1986, Kogure 1987, Bando et al. 1989). Compared with separate administration, the combined administration of four hypothalamic releasing hormones in healthy beagle dogs caused no apparent inhibition or synergism with respect to the responses to CRH, GHRH and TRH, whereas a 50% attenuation in the LH response was seen compared with the LH response to single GnRH administration (Meij et al. 1996a, b). In the present study, the CAP test was used to study anterior pituitary function in dogs with PDH. The results demonstrate that pituitary function in dogs with PDH is altered in several respects other than the unrestrained release of ACTH responsible for hypercortisolism. Basal GH levels were decreased and PRL levels were increased, whereas TSH levels and LH levels (noncastrated PDH dogs) were not different from those in control dogs. The pituitary corticotropic response to combined stimulation with four hypothalamic releasing hormones was the same as that in control dogs: the somatotrophic and thyrotropic responses were decreased, whereas the gonadotropic and lactotropic response were increased compared with the responses in control dogs.

The ACTH response to combined hypophysiotropic stimulation in dogs with PDH was not different from that in control dogs, but occurred at higher ACTH levels. In studies of the responsiveness to single CRH administration, it was found that, in dogs with PDH, increments in plasma ACTH were similar to (Rijnberk et al. 1987) or somewhat lower than (Van Wijk et al. 1994) those in control dogs. Although not found in dogs, ACTH hyperresponsiveness to CRH is commonly found in humans with Cushing’s disease caused by pituitary microadenomas (Orth et al. 1982, Pieters et al. 1983, Schrell et al. 1987, Fukata et al. 1993), although testing in individual patients has revealed hyperresponsiveness, normal responsiveness and even unresponsiveness (Pieters et al. 1983). Apparently, the increased mass of corticotrophic cells has a relatively low responsiveness (per unit of cell mass) to hypophysiotropic stimulation, which may be the result of the neoplastic transformation and the persisting hypercortisolism.

Increases in cortisol levels after combined hypophysiotropic stimulation were greater in dogs with PDH than in control dogs. Hyperresponsivity of the adrenals to ACTH or CRH in dogs with PDH has been reported previously and can be explained by an increased mass of adrenocortical tissue (Meijer et al. 1978, Rijnberk et al. 1987). Growth hormone levels in dogs with PDH were slightly (30%) but significantly reduced, whereas GH response to stimulation was more than 100% reduced compared with controls. Hyposcretion of GH in human patients with Cushing’s disease is considered to be a direct result of glucocorticoid excess or an indirect result of the obesity of the patients (Cuerda et al. 1991, Magiakou et al. 1994). There is increasing experimental evidence in rats that glucocorticoids exert their effect on GH release mainly by enhancement of somatostatin secretion (Wehrenberg et al. 1990, Giustina et al. 1995). Alternatively, the obesity in
hypercortisolemic patients might induce IGF-I-mediated GH suppression (Maggiou et al. 1994). In patients with Cushing’s syndrome, blunted GH responses were found to a synthetic hexapeptide (GHRP-6) which releases GH by a direct effect at the pituitary level, through receptors other than GHRH receptors (Leal-Cerro et al. 1994). This suggests that chronic hypercortisolism may also have a direct effect on pituitary somatotropin cells, although the impaired response might also be the result of the persisting inhibitory effect of somatostatin.

In humans, suppression of the hypothalamic–pituitary–gonadal axis, in states of increased stress and Cushing’s disease, is presumed to be mediated by glucocorticoid excess from the activated hypothalamic–pituitary–adrenocortical axis (Odagiri et al. 1988, Saketos et al. 1993, Samuels et al. 1994). Exogenous glucocorticoids also suppress LH plasma levels and the LH response to stimulation, although the results depend on gonadal status and type, dose and duration of administered steroids (Saketos et al. 1993, Samuels et al. 1994). The suppression of LH secretion by GnRH inhibition may be mediated through endogenous opioid peptides in normal women (Barbarino et al. 1989) and in rats (Belhadj et al. 1989). In dogs, short-term prednisone treatment (Kemppainen et al. 1983) or novelty stress in inexperienced laboratory dogs (Knol et al. 1989) suppresses plasma LH levels. There was no significant suppression of basal plasma LH levels in our dogs compared with controls, whereas a tendency to LH hyperresponsiveness after hypophysiotropic stimulation was seen. This may be the result of a dual effect – that is, both on the peripheral gland and at the hypothalamic–pituitary level. Glucocorticoid administration causes low basal testosterone concentrations in dogs (Kemppainen et al. 1983). Negative feedback could be expected to lead to highly elevated LH levels, but this seems to be prohibited by the effect of glucocorticoid excess at the hypothalamic–pituitary level, resulting in normal LH values and a tendency to hyperresponsiveness to stimulation.

Hyperprolactinemia was common in our dogs with PDH, in which basal plasma PRL levels were twice as high as in a previous study (Stolp et al. 1986). Also, the observed PRL increments after administration of four releasing hormones were 60% greater than the reported PRL increments following single TRH administration in dogs with PDH (Stolp et al. 1986). In dogs with an adrenocortical tumour, the PRL response to single TRH was not significantly different from that in control dogs (Stolp et al. 1986). In dogs with hyperadrenocorticism caused by an adrenocortical tumour, basal plasma PRL concentrations have also been found to be significantly elevated, although still significantly lower than the values in dogs with PDH (Stolp et al. 1986). After the administration of bromocriptine (a dopamine agonist), the plasma PRL concentrations in dogs with PDH decreased considerably, but remained higher than those in the control dogs and the dogs with an adrenocortical tumour (Stolp et al. 1986). It was concluded that PDH in the dog is associated with a disturbance in the regulation of PRL secretion that is not secondary to hypercortisolism per se (Stolp et al. 1986).

In humans, mild hyperprolactinemia frequently accompanies the hypopituitarism associated with macroadenomas not secreting ACTH and PRL (Arafah et al. 1994). Because they observed that ACTH increased and PRL levels decreased to baseline levels within a few hours after pituitary surgery, Arafah et al. (1994) concluded that hypopituitarism was largely caused by compression of portal vessels, which resulted in interruption of delivery of CRH and a hypothalamic PRL inhibiting factor. This mechanism is not likely to have caused the increased plasma PRL levels in our dogs with PDH, because only two of 47 dogs had pituitary tumours which measured more than 10 mm in height. Hyperprolactinemia in dogs with PDH may have been the result of cosecretion, resembling the coincident hyperprolactinemia described in pituitary tumours other than prolactinomas in humans (Asa and Kovacs 1983). Such adenomas may be composed of one cell population which produces two or more hormones (monomorphous adenomas), or of multiple cell types, each producing one hormone (plurimorphous adenomas) (Asa and Kovacs 1983).

Plasma thyroxine concentrations were lower in the dogs with PDH, but plasma TSH concentrations were not different from those in controls. After hypophysiotropic stimulation, the plasma TSH response in the dogs with PDH was lower than that in controls. The lowering of circulating thyroxine concentrations in dogs is considered mainly the result of altered transport, distribution and metabolism of thyroxine (Kaptein et al. 1992) due to the glucocorticoid excess per se. There is also evidence in dogs that glucocorticoids may interfere with basal thyroid hormone secretion by inhibiting lysosomal hydrolysis in the thyroid follicular cell (Kemppainen et al. 1983). Thus, for the hypothalamic–pituitary–thyroid axis, a concept may be applicable that is similar to that presented above for the pituitary–gonadal axis: a dual effect, at the hypothalamic–pituitary level and at the peripheral level. The low circulating levels of thyroxine secreted by the thyroid gland would be expected to cause high TSH concentrations, but these seem to be prevented by the effects of glucocorticoids at the hypothalamic–pituitary level, even to the extent that there is hyporesponsiveness of TSH to hypophysiotropic stimulation, which has also been reported for glucocorticoid excess in humans (Cuerda et al. 1991, Samuels et al. 1994).

We conclude that, in dogs with PDH, pituitary function is altered in several respects. 1) In spite of persisting hypercortisolemia and the neoplastic transformation of the corticotropic cells, these cells usually remain responsive to combined hypophysiotropic stimulation. 2) Basal plasma GH concentrations and GH responsiveness in the combined stimulation test are decreased, probably as a result of
the glucocorticoid-induced increase of the somatostatin tone. 3) Plasma PRL concentrations and the PRL response to stimulation are increased, probably as a result of cosecretion with ACTH by the transformed corticotropic cells. 4) Despite the well known effects of glucocorticoids in lowering circulating concentrations of gonadal steroids and thyroid hormone, the basal plasma concentrations of LH and TSH remain unchanged and there is a tendency to hyperresponsiveness to stimulation for LH and hyporesponsiveness for TSH. The most likely explanation for these changes is a dual effect of glucocorticoids: a direct effect on the gonads and thyroids and/or the transport and metabolism of their secretory products, and an influence on the sensitivity of the feedback control at the hypothalamic–pituitary level.

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References


Arts CJM, Koppeschaar HPE, Veeman W & Thijsen JHH 1985 A direct radioimmunoassay for the determination of adrenocorticotropic hormone (ACTH) and a clinical evaluation. Annals of Clinical Biochemistry 22 247–256.


Leal-Cerro A, Pumar A, Garcia–Garcia E, Dieguez C & Casanueva FF 1994 Inhibition of growth hormone release after the combined administration of GHRH and GHRP-6 in patients with Cushing’s syndrome Clinical Endocrinology 41 649–654.


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