Diagnosis of Pituitary Adenoma by Using a Combined Dexamethasone Suppression and TRH Stimulation Test

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The measurement of plasma cortisol after a combined dexamethasone suppression–thyroid-releasing hormone (TRH) stimulation test consistently separated normal horses from horses with pituitary adenoma. Combined testing appears to be more powerful in diagnosing pituitary adenomas in horses than either the dexamethasone suppression or TRH stimulation test used alone. Authors' addresses: Depts. of Large Animal Clinical Sciences (Andrews and Green), Animal Science (Eiler and Fecteau), Comparative Medicine (Oliver), and Pathology (McCracken), College of Veterinary Medicine, The University of Tennessee, P.O. Box 1071, Knoxville, TN 37901-1071. © 1997 AAEP.

1. Introduction

Pars intermedia pituitary adenoma (PIPA) affects all breeds of horses 7 years of age or older and may lead to clinical signs of hirsutism, polyuria, polydipsia, a pot-bellied appearance, and chronic laminitis. A diagnosis of PIPA can be made by the presence of clinical signs in advanced cases. However, a diagnosis of PIPA in horses with absent or mild clinical signs requires laboratory support. The measurement of plasma cortisol concentration after a thyroid-releasing hormone (TRH) stimulation test or dexamethasone (DX) suppression test is currently being used to diagnose PIPA in horses. However, with current testing methods, the separation of horses with PIPA from normal horses may not be possible by using a single test. For example, a previous report showed a significant increase in plasma cortisol concentration 30 min after the administration of TRH in horses with PIPA. In that study, after TRH stimulation, plasma cortisol concentrations in normal horses overlapped plasma cortisol concentrations seen in horses with PIPA. Thus, an interpretation of a single TRH stimulation test may prove inconclusive.

Another report showed a significant suppression of plasma cortisol concentrations 16–24 h after the administration of DX. In that study, the plasma cortisol concentration was less than 1 µg/dl in normal horses 16, 20, and 24 h after administration of DX, whereas horses with PIPA had plasma cortisol concentrations above 1.0 µg/dl at the same times. However, in our experience, plasma cortisol concentrations may not be suppressed completely 20–24 h after DX administration in some horses with early PIPA, especially when the resting plasma cortisol concentrations are high normal. The lack of sup-
pression in some normal horses may result in an overlap of plasma cortisol concentrations between normal horses and horses with PIPA 20–24 h after DX administration. Furthermore, normal horses with higher resting baseline plasma cortisol concentrations may not show suppression below 1.0 µg/dl and thus may be interpreted as a false positive (i.e., normal horses misdiagnosed as a horse with PIPA).

The objective of this present report was to increase the accuracy of diagnosing PIPA in horses by using a combined DX suppression and TRH stimulation (CDS–TRH) test. We hypothesize that the administration of DX prior to the administration of TRH will result in a similar baseline plasma cortisol concentration in both healthy horses and horses with PIPA and may provide selective blockade of TRH pituitary receptors in normal horses. We propose that a CDS–TRH test will be a more powerful test in diagnosing PIPA in horses than either test used alone.

2. Materials and Methods

Seven normal Thoroughbred mares and five mares with PIPA, 8–26 years old and weighing 361–498 kg, were used in this study. A diagnosis of PIPA was based on clinical signs of an excessively long hair coat (hirsutism), loss of the normal shedding pattern, weight loss, polydipsia, polyphagia, hyperglycemia, suborbital fat pad enlargement (determined by ultrasound and digital palpation), and a history of chronic laminitis. One of the five affected horses had PIPA at gross necropsy, which was confirmed on histopathologic examination of the pituitary gland. Jugular catheters were inserted at 8:00 am and the test procedure began at 8:30 am. Blood was withdrawn from the jugular catheter and placed in vacutainers containing ethylenediamine tetra-acetic acid anticoagulant, prior to and 3 h after the administration of DX (40 µg/kg IV). TRH (1.0 mg of acetate salt extracted weight (diluted in physiologic saline), IV) was then given and blood samples were withdrawn at 15, 30, 45, 60, and 90 min and 24 h after DX administration. Blood samples were centrifuged and plasma was collected and stored for a week at −20°C until a cortisol analysis was conducted by using a validated radioimmunoassay procedure.

An analysis of variance was done to determine the effect of treatment on cortisol response. When a significant difference among treatment means was identified by an F test, a mean separation was accomplished by using Duncan's multiple-range test. Significance was reported at p < 0.05.

3. Results

Baseline plasma cortisol concentrations were significantly lower (p < 0.05) in horses with PIPA than in normal horses in this study (Fig. 1). Plasma cortisol concentrations were similarly suppressed in normal horses and horses with PIPA 3 h after the administration of DX (Fig. 1). TRH administration did not cause an increase in plasma cortisol concentration in 7/7 normal horses pretreated with DX in this study (Fig. 1). However, plasma cortisol concentrations were significantly (p < 0.05) increased from 66% to 294% in the 5/5 horses with PIPA (Fig. 1). In addition, there was no overlap in plasma cortisol concentrations between normal horses and horses with PIPA, 30 min after TRH administration (Fig. 1).

4. Discussion

Resting plasma cortisol concentrations were lower in horses with PIPA than in normal healthy horses. The results in this study are similar to previous reports, but they are contrary to another report in which horses with PIPA had higher plasma cortisol concentrations. Thus, finding a low to normal (rather than increased) plasma cortisol concentration does not rule out PIPA. Furthermore, variable results have been found in horses with PIPA and may be due to assay procedures (protein binding, radioimmunoassay, and enzyme immunoassay). For example, a protein-binding technique has been reported to overestimate the resting plasma cortisol concentration in horses by 220%, and adrenocorticotropic hormone-stimulated values by 107%.

Plasma cortisol concentrations were suppressed equally in healthy horses and horses with PIPA syndrome 3 h after dexamethasone administration. The results in this study are consistent with previous reports of horses with PIPA that used a combined dexamethasone and adrenocorticotropic hormone stimulation. Also, plasma cortisol concentrations in horses with PIPA returned to baseline values sooner than those in normal healthy horses in this study. This (cortisol escape) is consistent with previous dexamethasone suppression results. The claimed resistance of DX suppression in horses with PIPA syndrome may be explained by infrequent sampling after DX injection rather than actual resistance of suppression. In a previous report, blood
samples were withdrawn between 8 and 15 h after DX injection, and it is likely that the suppression phase was missed, with plasma cortisol concentrations returning to baseline prior to 8–15 h. The latter statement is supported by the fact that in the same publication a different group of horses with PIPA did show suppression at 3 h after DX injection. Therefore, a final sample at 24 h after DX injection is of diagnostic value because horses with PIPA syndrome will have returned to baseline cortisol concentrations, whereas cortisol in healthy horses remains suppressed. However, in some horses with PIPA syndrome, the 24-h cortisol values may be inconclusive as a result of either a low baseline cortisol level or cortisol assay variability.

A significant finding in this study was the fact that DX completely blocked the discrete elevation in mean concentration of plasma cortisol that is observed after the administration of TRH in healthy horses, but not in horses with PIPA syndrome. Previous studies using TRH stimulation alone showed that PIPA horses responded differently than healthy horses. This is debatable because maximal plasma cortisol concentrations after TRH stimulation did not differ in the two groups of horses. Again, it is suspected that the increase in plasma cortisol concentration becomes significant in horses with PIPA syndrome because the pretest plasma cortisol value is lower. However, if the pretest concentration of plasma cortisol is normal or high normal in horses with PIPA syndrome, a single TRH stimulation test may not discriminate and may actually provide a false negative result. The critical procedure that provided diagnostic value in our study was the combination of DX and the ability of the TRH to override the DX suppression in horses with PIPA and not in healthy horses. Furthermore, in this study, the plasma cortisol concentration increased from 66% to 294% in horses with PIPA, whereas there was no increase in plasma cortisol concentration in normal horses after TRH administration. Thus, CDS–TRH testing is a promising diagnostic tool for equine PIPA syndrome.

5. Recommended Test Procedure for Clinical Use

1. Withdraw blood for baseline plasma cortisol concentrations.
2. Inject dexamethasone (40 µg/kg IV).
3. Withdraw blood for plasma cortisol concentrations 3 h after the administration of dexamethasone.
4. Administer TRH (1.0 mg IV).
5. Withdraw blood for plasma cortisol concentrations 30 min after the administration of TRH.
6. Withdraw blood for plasma cortisol concentrations 24 h after the administration of dexamethasone.

6. Test Interpretation

Horses with PIPA are characterized by (1) low or low-normal baseline plasma cortisol concentrations; (2) suppression of plasma cortisol concentrations 3 h after DX administration; (3) a return to baseline plasma cortisol concentrations 30 min after TRH stimulation; (4) a return to baseline plasma cortisol concentrations 24 h after DX administration; and (5) a test that provides a double check (plasma cortisol 30 min after TRH stimulation and plasma cortisol 24 h after DX administration) for the diagnosis of PIPA.

References and Footnotes


*Sigma Chemical Co., St. Louis, MO 63178.