Evaluation of serum 17-hydroxyprogesterone concentration after administration of ACTH in dogs with hyperadrenocorticism

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Objective—To evaluate serum 17-hydroxyprogesterone (17-OHP) concentration measurement after administration of ACTH for use in the diagnosis of hyperadrenocorticism in dogs.

Design—Prospective study.

Animals—110 dogs.

Procedure—Serum 17-OHP concentrations were measured before and after ACTH stimulation in 53 healthy dogs to establish reference values for this study. Affected dogs had pituitary-dependent (n = 40) or adrenal tumor–associated (12) hyperadrenocorticism or potentially had atypical hyperadrenocorticism (5; diagnosis confirmed in 1 dog). In affected dogs, frequency interval and borderline and abnormal serum 17-OHP concentrations after ACTH stimulation were determined. Serum cortisol concentrations were assessed via low-dose dexamethasone suppression and ACTH stimulation tests.

Results—In healthy dogs, serum 17-OHP concentration frequency intervals were grouped by sex and reproductive status (defined as < 95th percentile). Frequency intervals of serum 17-OHP concentrations after ACTH stimulation were < 7.7, < 2.0, < 3.2, and < 3.4 ng/mL (< 23.3, < 6.1, < 9.7, and < 10.3 nmol/L) for sexually intact and neutered females and sexually intact and neutered males, respectively. In 53 dogs with confirmed hyperadrenocorticism, serum cortisol concentrations after ACTH stimulation and 8 hours after administration of dexamethasone and serum 17-OHP concentrations after ACTH stimulation were considered borderline or abnormal in 79%, 93%, and 69% of dogs, respectively. Two of 5 dogs considered to have atypical hyperadrenocorticism had abnormal serum 17-OHP concentrations after ACTH stimulation.

Conclusions and Clinical Relevance—Serum 17-OHP concentration measurement after ACTH stimulation may be useful in the diagnosis of hyperadrenocorticism in dogs when other test results are equivocal. (*J Am Vet Med Assoc* 2005;227:1095–1101)

Naturally occurring hyperadrenocorticism is a well-recognized endocrine disorder in dogs, the clinical signs of which are results of a chronic excess in circulating cortisol concentration. Approximately 85% of these dogs have **pituitary-dependent hyperadrenocorticism (PDH)**, which is caused by excessive secretion of ACTH by a pituitary gland tumor that results in bilateral adrenocortical hyperplasia and excessive secretion of glucocorticoids by the adrenal glands.[1] Approximately 15% of dogs with naturally occurring hyperadrenocorticism have a functioning adrenocortical adenoma or carcinoma that autonomously secretes excessive quantities of cortisol.[2]

A diagnosis of hyperadrenocorticism in a dog should be suspected initially from review of the history and physical examination findings. Results of a CBC, serum biochemical analyses, and urinalysis typically indicate abnormalities consistent with the diagnosis. Confirmation of hyperadrenocorticism is traditionally accomplished by use of an ACTH stimulation test or **low-dose dexamethasone suppression (LDDS)** test.[3-5] However, in some dogs, the results of these tests may be within the reference intervals despite clinical signs, physical examination findings, and clinicopathologic abnormalities suggestive of hyperadrenocorticism.[6] It has been suggested that some of these dogs may have an atypical form of hyperadrenocorticism resulting from 1 or several derangements in the biosynthetic pathway for production of cortisol.

17-Hydroxyprogesterone (17-OHP) is one of the intermediary steroids produced when cholesterol is metabolized to cortisol (**Figure 1**). It has been hypothesized that some dogs with naturally occurring hyperadrenocorticism may have abnormalities solely in serum 17-OHP concentration. This biochemical abnormality could result from a relative deficiency in enzymes required for synthesis of cortisol (such as 21β hydroxylase or 11β hydroxylase) subsequent to the synthesis of 17-OHP, causing accumulation of precursor

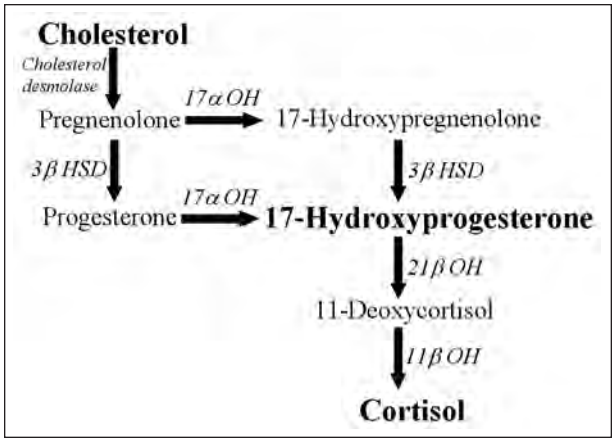

Figure 1—Main pathways of adrenocortical steroidogenesis. HSD = Hydroxysteroid dehydrogenase. OH = Hydroxylase.

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steroids proximal to the blockade in the synthetic pathway. Alternatively, 17-OHP may be the primary steroid synthesized by an adrenocortical tumor.[6] High concentrations of 1 or more steroid precursors may cause clinical signs or may be shunted into alternative metabolic pathways and cause excesses in other steroid hormones such as androstenedione.[7] 17-Hydroxyprogesterone is a progestin with intrinsic glucocorticoid activity that could also increase the bioavailability of cortisol by displacing it from cortisol-binding protein.[8,9] It has been suggested that measuring serum 17-OHP concentrations after ACTH administration may aid in the diagnosis of hyperadrenocorticism in dogs.[6,10,11,a] Chronic excesses in systemic concentrations of sex hormones (such as 17-OHP) have been reported[12-19] in humans, dogs, and cats with adrenocortical tumors. The purpose of the study reported here was to evaluate the usefulness of measuring serum 17-OHP concentration after ACTH administration in the diagnosis of hyperadrenocorticism in dogs.

Materials and Methods

Dogs—The study included dogs that were evaluated by at least one of the authors at the University of California, Davis, Veterinary Medical Teaching Hospital from March 1, 2002, to December 1, 2003. All dogs were enrolled with the informed consent of their owners. Healthy dogs (selected on the basis of not having a history of illness and no abnormal findings on physical examination) were used to establish reference values for serum 17-OHP concentration for this study. None of these dogs had a recent history of glucocorticoid administration. All sexually intact females were in anestrus; this was determined on the basis of serum progesterone concentration < 1 ng/mL (3.2 nmol/L) and lack of clinical signs of proestrus.[20] In all healthy dogs, an ACTH stimulation test was performed to determine baseline serum 17-OHP concentration and serum 17-OHP concentration 1 hour after ACTH administration.

For inclusion in the group of dogs with naturally occurring hyperadrenocorticism, each dog had both historical data and physical examination results consistent with this diagnosis.[21] Furthermore, each dog had clinicopathologic abnormalities suggestive of hyperadrenocorticism detected via serum biochemical analyses (eg, high alkaline phosphatase and alanine aminotransferase activities, hypercholesterolemia, low BUN concentration, and high blood glucose concentration after food had been withheld) and urinalysis or microbial culture of urine (eg, urine specific gravity < 1.020 and microbial growth on culture of urine).

Dogs with naturally occurring hyperadrenocorticism were allocated to 1 of 3 groups as follows: group 1 included dogs with PDH, group 2 included dogs with **adrenal tumor–associated hyperadrenocorticism (ATH)**, and group 3 included dogs that potentially had so-called atypical hyperadrenocorticism. Group 1 dogs had to have results of an LDDS test that were indicative of PDH or high serum cortisol concentration following ACTH stimulation. The diagnosis of PDH in these dogs had to be supported by identification of 2 relatively equal-sized adrenal glands via abdominal ultrasonography and good clinical response to the adrenocorticolytic drug o,p'-DDD or pituitary gland radiation therapy or evidence of bilateral adrenocortical hyperplasia at necropsy.[3,4,22-26] Group 2 dogs had to have an abnormal LDDS test result, an undetectable plasma concentration of endogenous ACTH, an adrenal mass detected via abdominal ultrasonography, an adrenocortical tumor (adenoma or carcinoma) identified via histologic examination of the adrenal mass (removed during celiotomy or at necropsy), and adrenal mass measurements available for determination of its approximate volume.[22] Volume was estimated from adrenal tumor measurements obtained via gross examination. Group 3 dogs had to have clinical signs as reported by the owners of these dogs and clinicopathologic abnormalities consistent with hyperadrenocorticism. Each dog in group 3 had serum cortisol concentration < 1.4 µg/dL (38.6 nmol/L) 8 hours after administration of a low dose of dexamethasone and serum cortisol concentration < 22 µg/dL (607 nmol/L) 1 hour after ACTH stimulation.[3,b]

Hormone tests—Each dog in groups 2 and 3 underwent an ACTH stimulation test and an LDDS test. All dogs in group 1 underwent an ACTH stimulation test and 35 of 40 dogs in this group underwent an LDDS test. For the ACTH stimulation test, blood samples were collected before and 1 hour after IM administration of synthetic ACTH[c] (0.25 mg/dog) for the measurement of serum cortisol and 17-OHP concentrations. Serum cortisol concentration > 22 µg/dL (607 nmol/L) 1 hour after administration of ACTH was considered consistent with naturally occurring hyperadrenocorticism.[b] Serum cortisol concentrations in the range of > 17 to 22 µg/dL (469 to 607 nmol/L) were categorized as borderline, and those in the range of 6 to 17 µg/dL (165.5 to 469 nmol/L) were considered to be within the reference interval.[b] For the LDDS test, blood samples were collected before and 4 and 8 hours after IV administration of dexamethasone (0.01 mg/kg [0.005 mg/lb]) for determination of serum cortisol concentration. Serum cortisol concentration > 1.4 µg/dL (38.6 nmol/L) 8 hours after dexamethasone administration was considered consistent with naturally occurring hyperadrenocorticism.[22] Serum cortisol concentrations from 0.9 to 1.4 µg/dL (24.8 to 38.6 nmol/L) were categorized as borderline, and serum cortisol concentrations < 0.9 µg/dL were considered to be within the reference interval.[b]

Hormone assays—Blood samples obtained for determination of plasma concentrations of endogenous ACTH were collected, stored, and assayed as previously described.[4] Serum cortisol concentrations were measured by use of a commercial cortisol radioimmunoassay[d] that has been validated for use in dogs.[1] The analytical sensitivity of this radioimmunoassay is 0.3 µg/dL (8.3 nmol/L). Serum progesterone concentration was measured by use of a radioimmunoassay that has been validated for use in dogs.[27]

For serum 17-OHP assessments, sera were obtained and frozen at –70°C for no more than 1 month prior to assay. Serum 17-OHP concentration was measured by use of a commercial solid-phase radioimmunoassay.[e] The assay was validated for precision, recovery, sensitivity, and stability of frozen samples. Multiple aliquots of pooled serum samples containing low (< 0.5 ng/mL [1.5 nmol/L]), midrange (1.0 to 3.0 ng/mL [3.0 to 9.1 nmol/L]), and high (> 5.0 ng/mL [15.2 nmol/L]) concentrations of 17-OHP were frozen to avoid thawing and refreezing of samples. Intra-assay precision was determined from 8 repeated measurements of aliquots from each of the 3 pooled serum samples within 1 assay. Interassay precision was determined from 6 repeated measurements of sample aliquots provided in the assay kit (human serum-based assay) performed on 6 separate days. Interassay precision calculations were determined on replicates of human serum-based samples provided in the kit because assayed commercial canine serum-based control samples for 17-OHP were not available. The intra-assay coefficients of variation were 5.7%, 1.4%, and 2.9%, and the interassay coefficients of variation were 11.6%, 11.7%, and 12.5% for samples containing low, midrange, and high 17-OHP concentrations, respectively. Recovery was determined by adding standard solutions of 17-OHP provided in the assay kit to canine sera

and measuring completeness of recovery of 17-OHP in the samples. Recovery was carried out over a concentration interval of 0.8 to 6.3 ng/mL (2.4 to 19.1 nmol/L). Recovery ranged from 80% to 106%, corresponding to the lowest and highest concentrations used for the recovery studies. Sensitivity was determined by identifying the intercept of the lower limit of the 95% confidence interval of B/Bo × 100 (ratio of the slope [B] to the y-intercept [Bo]) on the standard curve. Analytical sensitivity was 0.06 ng/mL (0.2 nmol/L). Stability of frozen samples (–70°C) was determined by repeated assay of the same sample aliquots over a 4-week period. Frozen samples were stable for at least 4 weeks.

Statistical analyses—The exact Kruskal-Wallis 1-way ANOVA was used to detect differences in baseline serum 17-OHP concentrations and serum 17-OHP concentrations after ACTH stimulation among categories based on sex and reproductive (neuter) status. Once significance was detected, an exact Mann-Whitney *U* test was used to evaluate differences in serum 17-OHP concentrations between sex groups, between age groups, and between baseline serum 17-OHP concentrations in groups of healthy dogs and dogs with hyperadrenocorticism. The exact Wilcoxon signed rank test for paired data was used to evaluate baseline serum 17-OHP concentrations and serum 17-OHP concentrations after ACTH stimulation in healthy dogs. The results of tests on the distribution of data for serum 17-OHP concentration after ACTH stimulation indicated that more than half of the observations were non-normally distributed. Therefore, to maintain consistency, all data are presented as 90% frequency intervals (5th to 95th percentile). We arbitrarily established the reference interval as serum 17-OHP concentration < the 95th percentile, borderline results as serum 17-OHP concentration from the 95th to 100th percentile, and abnormal results as serum 17-OHP concentration > the 100th percentile of the frequency interval. Statistical analyses were performed by use of a statistical software package.[f] A value of $P < 0.05$ was considered significant.

Results

Fifty-three healthy dogs were enrolled in the study. These dogs included 16 neutered males (designated MN dogs), 15 neutered females (designated FN dogs), 12 sexually intact males (designated MI dogs), and 10 sexually intact females (designated FI dogs). Median age was 4 years (range, 0.4 to 15 years). The dogs' weights ranged from 4 to 50 kg (8.8 to 110 lb), and 20 different breeds were represented. With regard to serum 17-OHP concentrations after ACTH stimulation, reference values and borderline and abnormal results for our study were determined in the 53 healthy dogs (**Table 1**). In healthy dogs, there was a significant ($P < 0.001$) difference between baseline serum 17-OHP concentration and serum 17-OHP concentration after ACTH stimulation. There was no significant ($P = 0.6$) difference between baseline serum 17-OHP concentrations of healthy dogs ≤ 4 years of age (n = 25) and those > 4 years of age (28). After ACTH stimulation, serum 17-OHP concentrations were significantly ($P = 0.03$) increased in FI dogs, compared with FN dogs, and were significantly ($P < 0.001$) decreased in FN dogs, compared with MI dogs (**Figure 2**). Because of these differences, reference intervals and borderline and abnormal results of serum 17-OHP concentrations after ACTH stimulation for this study were grouped by sex and neuter status.

Fifty-two dogs met the criteria for inclusion in groups 1 and 2. These included 40 dogs with PDH (group 1) and 12 dogs with ATH (group 2). Five dogs were tentatively assigned to group 3 (dogs with atypical hyperadrenocorticism). Compared with findings in healthy dogs, baseline serum 17-OHP concentration in dogs with hyperadrenocorticism was significantly ($P = 0.009$) different; the median concentration was 0.1 ng/mL (0.3 nmol/L; range, 0.04 to 2.1 ng/mL [0.1 to 6.4 nmol/L]) in healthy dogs and 0.2 ng/mL (0.6 nmol/L; range, 0.04 to 3.9 ng/mL [0.1 to 11.8 nmol/L])

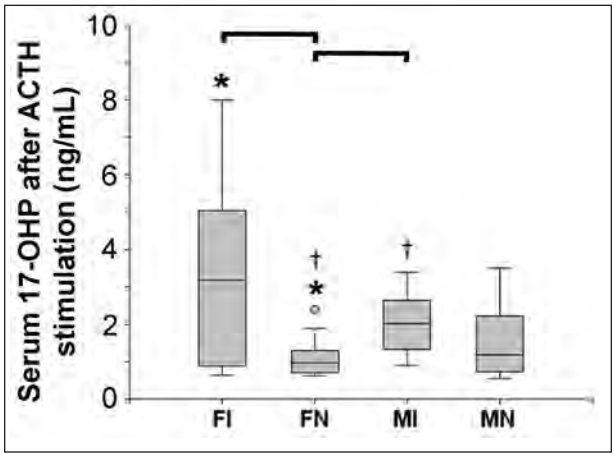

Figure 2—Box and whisker plot of serum 17-hydroxyprogesterone (17-OHP) concentrations (ng/mL) after ACTH stimulation in 10 sexually intact female (FI) dogs, 15 neutered female (FN) dogs, 12 sexually intact male (MI) dogs, and 16 neutered male (MN) dogs. Boxes represent the 25th to 75th percentile of dogs, and whisker bars represent the 5th and 95th percentiles of dogs. Circular datum point represents outliers in a category. The line in the middle of each box represents the median serum 17-OHP concentration after ACTH stimulation. *,†Significant ($P < 0.05$) difference between values in the bracketed groups.

Table 1—Reference intervals and borderline and abnormal serum 17-hydroxyprogesterone (17-OHP) concentrations after ACTH stimulation determined in 53 healthy dogs.

	Serum 17-OHP concentrations (ng/mL [nmol/L])		
Dogs	**Reference interval***	**Borderline results***	**Abnormal results***
Sexually intact females (n = 10)	< 7.7 (23.3)[a]	7.7–8.0 (23.3–24.2)[a]	> 8.0 (24.2)[a]
Neutered females (15)	< 2.0 (6.1)[a,b]	2.0–2.4 (6.1–7.3)[a,b]	> 2.4 (7.3)[a,b]
Sexually intact males (12)	< 3.2 (9.7)[b]	3.2–3.4 (9.7–10.3)[b]	> 3.4 (10.3)[b]
Neutered males (16)	< 3.4 (10.3)	3.4–3.5 (10.3–10.6)	> 3.5 (10.6)

*Reference interval was defined as < the 95th percentile, borderline serum 17-OHP concentration was defined as the 95th to 100th percentile, and abnormal serum 17-OHP concentration was defined as > the 100th percentile of the frequency interval.

[a,b]Within a column, values with the same superscript letters are significantly ($P < 0.05$) different from each other.

in dogs with hyperadrenocorticism. After ACTH stimulation, serum 17-OHP concentration in dogs with hyperadrenocorticism was significantly ($P < 0.001$) different, compared with that in healthy dogs; the median concentration was 1.4 ng/mL (4.2 nmol/L; range, 0.6 to 8.0 ng/mL [1.8 to 24.2 nmol/L]) in healthy dogs and 4.0 ng/mL (12.1 nmol/L; range, 0.5 to 27.4 ng/mL [1.5 to 83.0 nmol/L]) in dogs with hyperadrenocorticism. Results of serum cortisol assessment after ACTH stimulation and 8 hours after dexamethasone administration and results of serum 17-OHP assessment after ACTH stimulation were borderline or abnormal in 79%, 93%, and 69% of 52 dogs (groups 1 and 2 combined) with confirmed hyperadrenocorticism, respectively.

Group 1 included 26 FN, 11 MN, and 3 MI dogs. Median age was 11 years (range, 6 to 15 years), and 23 breeds were represented. For all 40 dogs with PDH, results of an ACTH stimulation or LDDS test (or both) were consistent with the diagnosis of hyperadrenocorticism. Twenty-seven of the 40 (68%) dogs had abnormal ACTH stimulation test results (results were within the reference interval for 4 dogs and considered borderline for 9 dogs); 31 of 35 (89%) dogs had abnormal LDDS test results (results were within the reference interval for 3 dogs and considered borderline for 1 dog; 5 dogs did not undergo LDDS testing). All 5 dogs that did not undergo the LDDS test had abnormal ACTH stimulation test results. Twenty-seven of 40 (68%) dogs with PDH (16/26 FN dogs; 8/11 MN dogs; and 3/3 MI dogs) had abnormal serum 17-OHP concentrations after ACTH stimulation (results were within the reference interval for 11 dogs and considered borderline for 2 dogs). Of those 27 dogs with abnormal serum 17-OHP concentrations after ACTH stimulation, 16 also had abnormal ACTH stimulation test and LDDS test results, 3 had abnormal LDDS test results and borderline ACTH stimulation test results, 1 had an abnormal LDDS test result and an ACTH stimulation test result within the reference interval, and 2 had an abnormal ACTH stimulation test result and an LDDS test result within the reference interval. Five of the 5 dogs that did not undergo LDDS testing had abnormal serum 17-OHP concentrations after ACTH stimulation. One of 5 dogs with PDH for which ACTH stimulation test results were within the reference interval had an abnormal serum 17-OHP concentration after ACTH stimulation, and 2 of 3 dogs with LDDS test results within the reference interval had an abnormal serum 17-OHP concentration after ACTH stimulation. Overall, serum 17-OHP and cortisol concentrations after ACTH stimulation were abnormal in 68% of dogs in group 1.

Group 2 included 8 FN and 4 MN dogs. Median age was 10 years (range, 8 to 13 years), and 8 breeds were represented. The range of sizes of the adrenal masses determined via gross examination was 0.22 to 405 cm^3. Histologically, all adrenal masses were classified as adrenocortical in origin (9 carcinomas and 3 adenomas). All 12 dogs had abnormal LDDS test results. Four dogs had abnormal serum cortisol concentrations after ACTH stimulation (results were within the reference interval for 7 dogs and considered borderline for 1 dog), and 7 dogs (5/8 FN dogs and 2/4 MN dogs) had abnormal serum 17-OHP concentrations after ACTH stimulation (results were within the reference interval for the other 5 dogs). Four of 7 dogs with abnormal serum 17-OHP concentrations after ACTH stimulation had abnormal results of ACTH stimulation and LDDS tests, 1 had an abnormal LDDS test result and a borderline ACTH stimulation test result, and 2 had abnormal LDDS test results and ACTH stimulation test results that were within the reference interval. Two of 7 dogs with an ACTH stimulation test result that was within the reference interval had abnormal serum 17-OHP concentrations after ACTH stimulation. After ACTH stimulation, serum 17-OHP and cortisol concentrations were abnormal in 7 and 4 of the dogs in group 2, respectively. The median serum 17-OHP concentrations after ACTH stimulation were 4.5 ng/mL (13.6 nmol/L; range, 3.8 to 27.4 ng/mL [11.5 to 83.0 nmol/L]) and 2.0 ng/mL (6.1 nmol/L; range, 0.5 to 25.2 ng/mL [1.5 to 76.4 nmol/L]) for adenomas and carcinomas, respectively. These results were not significantly ($P = 0.3$) different. There was no correlation between volume of adrenal mass and serum 17-OHP or cortisol concentrations after ACTH administration.

Group 3 included 5 dogs with clinical signs and routine laboratory test results suggestive of hyperadrenocorticism; in these dogs, serum cortisol concentration after ACTH stimulation was < 22 µg/dL and serum cortisol concentration 8 hours after administration of a low dose of dexamethasone was < 1.4 µg/dL. In 3 dogs, serum cortisol concentration after ACTH stimulation was considered borderline (17 to 22 µg/dL); 8 hours after administration of a low dose of dexamethasone, serum cortisol concentration was < 0.9 µg/dL in all group 3 dogs. There were 4 MN dogs and 1 MI dog in group 3; median age was 12 years (range, 11 to 15 years), and 4 breeds were represented. Clinical signs of hyperadrenocorticism (as reported by the owners of these dogs) included polyuria (n = 4), polydipsia (4), polyphagia (3), lethargy (2), and excessive panting (1). Physical examination findings included thin skin (n = 2), hepatomegaly (2), alopecia (1), and pendulous abdomen (1). Typical serum biochemical and urine abnormalities included high liver enzyme activities (alkaline phosphatase [4 dogs], alanine aminotransferase [4], and γ-glutamyltransferase [4]), hypercholesterolemia (1), and urine specific gravity < 1.020 (4). After ACTH stimulation, the median serum 17-OHP concentration was 2.0 ng/mL (range, 1.4 to 6.4 ng/mL [4.2 to 19.4 nmol/L]). Two of the 5 dogs with suspected hyperadrenocorticism had abnormal serum 17-OHP concentrations after ACTH stimulation (6.4 and 4.0 ng/mL). One of these 2 dogs was treated with o,p'-DDD; following treatment, complete remission of all clinical abnormalities related to hyperadrenocorticism was reported. In this dog, serum cortisol concentrations after ACTH stimulation and 8 hours after administration of dexamethasone were 13.8 µg/dL (380.7 nmol/L) and 0.3 µg/dL, respectively. After 1 year of o,p'-DDD treatment, the dog had no clinical signs and serum 17-OHP concentration after ACTH stimulation was within the reference interval. In the other dog, serum cortisol concentrations after ACTH stimulation and 8 hours after administration of dex-

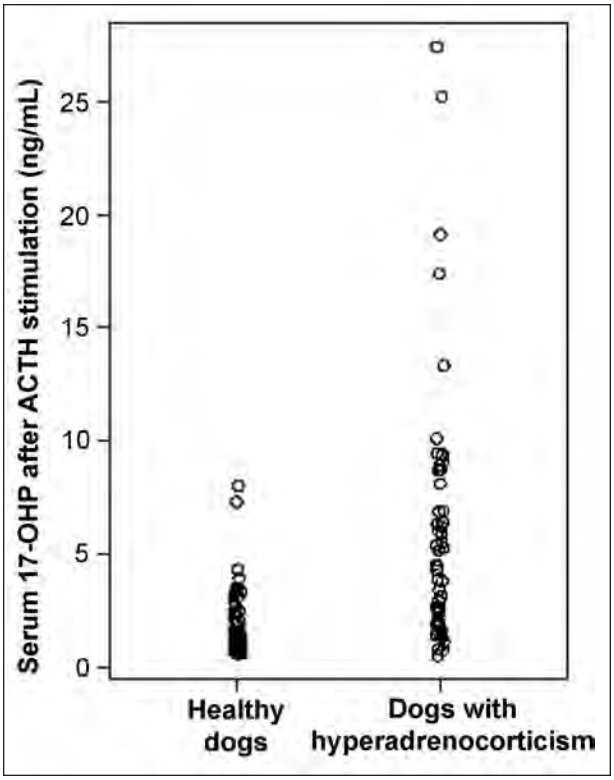

Figure 3—Distribution of serum 17-OHP concentrations (ng/mL) in 53 healthy dogs and 53 dogs with confirmed hyperadrenocorticism.

amethasone were 17.7 µg/dL (488.3 nmol/L) and 0.2 µg/dL (5.5 nmol/L), respectively. This latter dog and the other 3 dogs were not treated, and no further diagnosis was made. Therefore, in 1 of the 5 dogs for which a tentative diagnosis of atypical hyperadrenocorticism had been made, a final diagnosis of atypical hyperadrenocorticism was made. Overall, 53 dogs had confirmed hyperadrenocorticism. The serum 17-OHP concentrations after ACTH stimulation in the 53 healthy dogs and 53 dogs with confirmed hyperadrenocorticism were compared (**Figure 3**).

Discussion

In the study reported here, serum 17-OHP concentrations in sexually intact and neutered healthy dogs of both sexes were determined before and after stimulation with ACTH. Baseline serum 17-OHP concentrations and concentrations after ACTH stimulation were highest in FI dogs in anestrus. The finding that sex and neuter status affected serum 17-OHP concentration in dogs differs with that of a recent study[28] in which there were no significant differences in baseline serum 17-OHP concentrations or concentrations after ACTH stimulation between male and female dogs or between neutered and sexually intact dogs, although the specific stage of estrus cycle was not taken into account. In another study,[29] sexually intact dogs (both male and female) had significantly greater serum concentrations of 17-OHP before and after ACTH stimulation than neutered dogs and there was no significant difference in those concentrations between neutered males and females or between sexually intact males and females. Results from the healthy dogs indicated that independent reference intervals be established for dogs in each of the 4 sex-neuter categories (ie, sexually intact and neutered females and sexually intact and neutered males). The data obtained from the large number of healthy dogs included in the present study are of use, but larger groups of dogs of both sexes (sexually intact and neutered dogs) would be necessary to establish appropriate reference intervals. The serum 17-OHP concentration measurements in our study were similar to recently reported values.[28,29]

In dogs with PDH, serum 17-OHP and cortisol concentrations after ACTH stimulation were abnormal in 68% (27/40) of dogs. Synthesis and secretion of intermediary steroid hormones, such as 17-OHP, are possible in dogs with PDH because ACTH stimulates cortisol synthesis from cholesterol. It seems likely that increases in any or all of the intermediary products could occur. However, it seems unlikely that a dog would simultaneously acquire both PDH and a deficiency solely in the enzymes involved in the synthesis of cortisol subsequent to the synthesis of 17-OHP in that biochemical pathway and thereby have abnormalities only in serum 17-OHP concentration. In the 12 dogs with ATH, serum 17-OHP and cortisol concentrations after ACTH stimulation were abnormal in 7 and 4 dogs, respectively. It is possible that serum concentrations of intermediary products involved in the synthesis of cortisol could vary. In humans, dogs, and cats, an adrenocortical tumor can be associated with secretion of any of a large number of steroid products, including 17-OHP alone.[12-19]

The use of serum 17-OHP concentration measurement in the diagnosis of atypical hyperadrenocorticism has been recently reported.[6] In that study, the LDDS test cortisol response was suppressed in 4 dogs and 3 dogs had low serum cortisol concentration throughout the LDDS test, but all 7 dogs had marked increases in serum 17-OHP concentration and no response with regard to cortisol concentration following ACTH administration. Four dogs had a good clinical response to treatment, and in 2 other dogs, an adrenal tumor was detected postmortem. In the study of this report, no dog had a similar pattern of test results. Five dogs with clinical signs and routine clinicopathologic abnormalities (detected via CBC, serum biochemical analyses, and urinalysis) consistent with hyperadrenocorticism had serum cortisol concentration after ACTH stimulation < 22 µg/dL and suppression of the cortisol response in an LDDS test. Only 2 of the 5 dogs had an abnormal serum 17-OHP concentration after ACTH stimulation; 1 of these 2 dogs was treated (administration of o,p'-DDD) with resultant decreases in serum cortisol and 17-OHP concentrations. The test results in these 2 dogs are consistent with the suggestion that the diagnosis of hyperadrenocorticism can be supported via assessment of serum 17-OHP concentration after administration of exogenous ACTH when the diagnosis cannot be confirmed via assessment of serum cortisol concentration in ACTH stimulation or LDDS tests. However, this scenario would appear to be rare. We are confident that 53 dogs evaluated over a 21-month period in our study (groups 1 and 2 and 1 dog in group 3) had hyperadrenocorticism.

To our knowledge, assessment of serum 17-OHP concentration has not been evaluated in humans with PDH. Serum 17-OHP concentration following ACTH administration was evaluated in humans with various adrenal tumors, including nonhyperfunctioning adenomas, aldosterone-producing adenomas, adrenal cysts, and pheochromocytomas before and after unilateral adrenalectomy.[14,30] After unilateral adrenalectomy, serum 17-OHP concentration following ACTH stimulation substantially decreased in some of these patients. After stimulation with ACTH, both serum 17-OHP and cortisol concentrations were significantly correlated with the size of tumors in humans, indicating that tumoral volume itself may be an important determinant of this ACTH-induced response.[14,30] Such an association was not detected in the present study, possibly because of the small number of dogs with adrenal tumors. In our study, there was no significant difference in the median serum 17-OHP concentration after ACTH stimulation between dogs with ATH as a result of an adenoma and those with ATH as a result of a carcinoma; this is consistent with reported values of serum cortisol concentration after ACTH stimulation in dogs with ATH.[4]

Although serum cortisol concentrations after ACTH stimulation tests were abnormal in only 31 of 53 (58%) dogs with hyperadrenocorticism in the present study, this assessment is the only means by which iatrogenic hyperadrenocorticism can be identified and information regarding the outcome of treatment can be provided. Therefore, it is understood that this test remains useful in certain situations. In addition, after evaluating serum cortisol concentration, one could request that serum 17-OHP concentration be evaluated in the same sample. Results of the study reported here suggest that the measurement of serum 17-OHP concentration after ACTH administration could be useful in the diagnosis of hyperadrenocorticism in dogs only if other test results are equivocal. These conclusions are similar to those drawn from a recent study[29] in which serum 17-OHP concentrations before and after ACTH stimulation were evaluated for use in the diagnosis of hyperadrenocorticism in dogs. However, in the present study, serum 17-OHP concentration after ACTH stimulation as the sole abnormality was identified in only 1 of 53 dogs with confirmed hyperadrenocorticism and in only 2 of 5 dogs suspected of having atypical hyperadrenocorticism. These findings are not consistent with those of a similar study involving a larger number of dogs with atypical hyperadrenocorticism in which increases in serum 17-OHP concentration after ACTH stimulation were detected.[6] Results of serum 17-OHP assessment do not distinguish between PDH and ATH; measurements can be performed on serum samples following determination of serum cortisol concentration after ACTH administration, which is convenient and noninvasive. Serum samples can be stored at –20°C or –70°C for at least 1 month. As with serum cortisol determinations, results of serum 17-OHP assessments must be interpreted in relation to clinical findings, the most valuable diagnostic tool for clinicians.

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