IDEXX

Catalyst™ Cortisol Test: An accurate and reliable in-house tool for canine cortisol evaluation.

Introduction

Hypoadrenocorticism and Cushing's syndrome (hypercortisolism) are relatively uncommon endocrine disorders in dogs, but accurate diagnosis and effective management are critical.[1–4] In the case of hypoadrenocorticism, timely intervention can be lifesaving, while appropriate treatment for Cushing's syndrome can significantly improve a patient's quality of life and alleviate carer burden.

While point-of-care (POC) cortisol tests have been available for some time, most clinicians rely on commercial veterinary laboratories for cortisol measurement due to the need for high analytical accuracy and precision.[5] However, a POC assay that delivers reference laboratory-level performance would offer several clinical advantages. For example, a single resting cortisol result of ≥ 2.00 µg/dL (55.2 nmol/L) provides a practical and efficient means to help rule out hypoadrenocorticism as a diagnosis in dogs with compatible clinical signs or clinicopathologic changes, such as chronic gastrointestinal issues, acute vomiting or diarrhoea, hypoalbuminaemia, or electrolyte imbalances.[6–9]

Having reliable cortisol results available during the patient visit enables timely, in-person communication with pet owners. This not only supports shared decision-making but may also enhance client understanding and adherence to diagnostic and treatment recommendations.

This study evaluates the analytical performance of a novel POC immunoassay, the Catalyst™ Cortisol Test, for quantifying cortisol concentrations in canine serum.

Materials and methods

Method comparison

A method comparison study was conducted to evaluate the accuracy of the Catalyst Cortisol Test within a clinical setting using 705 canine serum or plasma samples originally collected for clinical purposes. These samples were analysed on Catalyst chemistry analysers located in 18 veterinary practices across the United States. Residual serum from each patient was submitted to IDEXX Laboratories, where cortisol concentrations were measured using the IMMULITE™ Veterinary Cortisol assay* performed on the IMMULITE™ 2000 Immunoassay System. The mean of two IMMULITE Veterinary Cortisol replicates served as the reference standard for comparison.

Correlation (R) and bias between the Catalyst Cortisol Test and the reference method were assessed using a Passing–Bablok regression. All method comparison analyses were done as per CLSI EP09c guidelines.[10]

Precision

Analytical precision was assessed using pooled canine serum samples at three cortisol concentrations as outlined in table 1. Testing was performed over 10 consecutive days on two Catalyst Dx™ and two Catalyst One™ chemistry analysers. On each day, four replicate measurements were obtained from each analyser during both morning and afternoon sessions to assess intra- and interday variability. All precision analyses were done as per CLSI EP05-A3 guidelines.[11]

Cross-reactivity

Understanding antibody cross-reactivity with other steroid hormones is essential when evaluating cortisol assays, as cross-reactivity can impact the clinical utility of the assay. To assess this, pooled canine serum samples at two cortisol concentrations (2.10 µg/dL (57.9 nmol/L) and 25.00 µg/dL (689.7 nmol/L)) were aliquoted and spiked with 13 naturally occurring steroid hormones and commonly administered corticosteroid medications (table 2). Each spiked sample was analysed in 12 replicates using Catalyst chemistry analysers, and the mean values were used to calculate percent cross-reactivity according to the following formula:

Percent cross-reactivity = [(spiked result – actual result) / steroid concentration] x 100

Interfering substances

Pooled canine serum samples with high (31.2 µg/dl (860.7 nmol/L)) and low (2.1 µg/dl (57.9 nmol/L)) cortisol concentrations and visually free of interfering substances were prepared for interference testing. To assess the potential impact of common interferents – haemolysis, lipaemia and icterus – canine red blood cell haemolysate†, Intralipid™‡ and ditaurobilirubin§ were used, respectively. Aliquots of the pooled serum were spiked with varying concentrations of each interferent, as detailed in table 3. All samples were then analysed on both a Catalyst One and a Catalyst Dx analyser to evaluate the assay's robustness to these substances. Per cent mean bias was calculated using the following formula:

Per cent mean bias = (spiked result – actual result) / actual result x 100

All interference analyses were done as per CLSI EP07 guidelines.[12]

Results

Method comparison

A regression plot evaluating correlation across the assay dynamic range is shown in Figure 1. The Catalyst Cortisol Test has excellent correlation (R = 0.95) with the reference method, with minimal to no bias (slope 1.06).

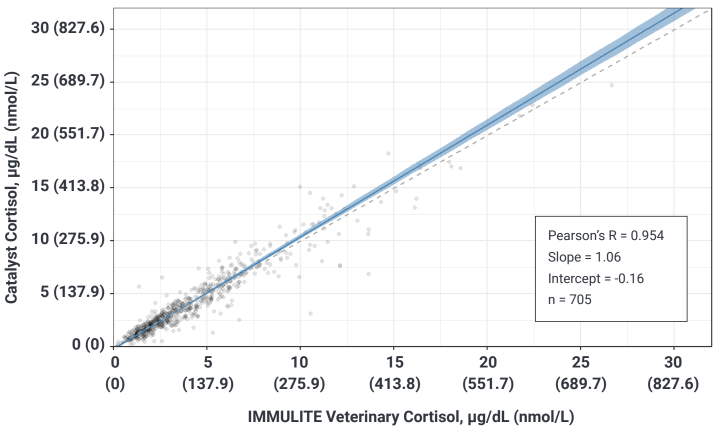

Figure 1. Correlation graph (Passing–Bablok regression) of pairwise comparisons of the Catalyst™ Cortisol Test and the IMMULITE™ Veterinary Cortisol assay in canine samples across the reportable range. The line of best fit (linear regression) is shown on the graph (solid blue), with 95% CI (shaded area) and X = Y (grey-dashed line).

Precision

The precision study results are summarised in Table 1. The assay demonstrated a total coefficient of variation (%CV) below 10% across clinically relevant cortisol concentrations (2.1mg/dl (57.9 nmol/L)–20.4 µg/dl (562.8 nmol/L)), indicating excellent analytical precision for veterinary use.

Cross-reactivity

The cross-reactivity profile for the Catalyst Cortisol Test is shown in Table 2. Cross-reactivity with naturally occurring steroid hormones is not expected to affect the clinical interpretation of results. The assay's cross-reactivity with commonly used glucocorticoid medications is comparable to that of other commercially available cortisol assays. For example, samples from patients receiving prednisone or prednisolone may show falsely elevated cortisol concentrations, while dexamethasone has minimal effect.

Interfering substances

The interfering substances results are summarised in Table 3. No interference was observed in lipaemic samples. However, icterus and moderate to marked haemolysis affected the results. Samples with these interferents should be avoided for use with this assay.

Conclusion

The Catalyst Cortisol Test demonstrates minimal bias, excellent precision, and strong correlation with the IMMULITE Veterinary Cortisol assay, supporting its accuracy and reliability for point-of-care cortisol measurement in dogs.

Icteric or moderately to severely haemolysed samples should be avoided, as these substances may impact assay performance.

Corticosteroid medications such as prednisone and prednisolone cross-react with the assay and may result in falsely elevated cortisol concentrations. Testing should be delayed in a patient receiving corticosteroid medications until after an appropriate withdrawal period, which depends on the medication administered, dosage and duration of use.

While dexamethasone does not cross-react with the Catalyst Cortisol Test, its administration alters pituitary-adrenal function. Therefore, performing cortisol testing prior to dexamethasone administration is recommended for patients suspected of having hypoadrenocorticism.

Mean concentration, µg/dL (nmol/L)	Standard deviation, µg/dL (nmol/L)	Coefficient of variation (%)	Number of replicates
2.10 (57.9)	0.14 (3.9)	7.75	320
6.30 (173.8)	0.29 (8)	5.39	320
20.40 (562.8)	1.11 (20.3)	6.81	320

Table 1. Summary of results from precision study.

Compound type	Compound	Compound concentration, µg/dL (nmol/L)	Catalyst™ Cortisol Test % cross-reactivity (base cortisol concentration 2.10 µg/dL)	Catalyst Cortisol Test % cross-reactivity (base cortisol concentration 25.00 µg/dL)
Naturally occurring hormone	Corticosterone	400 (11034.5)	7.12	5.18
	Cortisone	400 (11034.5)	11.24	8.56
	11-deoxycortisol	100 (2758.6)	10.27	2.93
	17-alpha-hydroxyprogesterone	400 (11034.5)	0.05	0.11
	Aldosterone	1,000 (27586.2)	0.13	0.15
	Progesterone	400 (11034.5)	0.03	0.23
Medication	Methylprednisolone	200 (5517.2)	0.10	0.57
	Desoxycorticosterone pivalate (DOCP)	400 (11034.5)	0.03	0.28
	Dexamethasone (1)	400 (11034.5)	0.02	0.51
	Dexamethasone (2)	4,000 (110344)	0.01	0.04
	Fludrocortisone	1,000 (27586.2)	4.09	2.75
	Prednisolone	8 (220.7)	23.87	15.56
	Prednisone	16 (441.4)	1.51	1.51
	Triamcinolone	5,000 (137931)	< 0.01	0.02

Table 2. Summary of cross-reactivity study with calculated cross-reactivities.

Interfering substance	Interfering level	Catalyst Cortisol Test concentration, µg/dL (nmol/L)		% Mean bias	
		Low	High	Low	High
Haemolysis	Control/not spiked	2.15 (59.3)	30.29 (835.6)	–	–
	25	2.28 (62.9)	31.08 (857.4)	6.0	2.6
	150	2.55 (70.3)	31.02 (855.7)	18.6	2.4
	250	2.53 (69.8)	30.55 (842.8)	17.7	0.9
	500	2.37 (65.4)	28.29 (780.4)	10.2	-6.6
Lipaemia	Control/not spiked	2.18 (60.1)	31.49 (868.7)	–	–
	125	2.12 (58.5)	31.05 (856.6)	-2.8	-1.4
	250	2.12 (58.5)	31.05 (856.6)	-3.0	-1.4
	500	2.12 (58.5)	30.67 (846.1)	-2.7	-2.6
Icterus	Control/not spiked	2.07 (57.1)	31.77 (876.4)	–	–
	0.5	2.14 (59)	29.88 (824.3)	3.3	-5.4
	1.0	2.24 (61.8)	28.36 (782.3)	8.3	-10.7
	2.0	2.40 (66.2)	25.42 (701.2)	15.8	-20.0

Table 3. Summary of results from interfering substances study with calculated bias.

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*Siemens Medical Solutions Diagnostics, Los Angeles, California, USA.
†Lysate from canine red blood cells washed in saline and lysed in water with no surfactant.
‡Intralipid™ (Sigma-Aldrich, Inc., St. Louis, Missouri, USA), a phospholipid-stabilised soya bean oil.
§Bilirubin conjugate (Scripps Laboratories, San Diego, California, USA; catalogue number: B0114), a synthesised ditaurobilirubin.