1. Use of Tandem Mass Spectrometry (LC-MS/MS) for the Measurement of Thyroid Hormones in Dogs with Spontaneous Hypothyroidism

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In human medicine, liquid chromatography tandem mass spectrometry (LC-MS/MS) is actually considered the "gold standard" for measurement of many hormone concentrations, and it is widely used in clinical practice; its diagnostic performance has never been investigated in dogs with hypothyroidism (DWH).

The aim of this study was to determine whether serum concentrations of fT₄, fT₃, rT₃, 3.3-T₂, 3.5-T₂, measured with LC-MS/MS, were able to differentiate DWH (n=13) from dogs with non-thyroidal illness (DNTI) (n=12), septic dogs (SD) (n=12), and healthy dogs (HD) (n=12).

Hypothyroidism was diagnosed based on consistent clinical signs, laboratory findings, total T₄ (TT₄) and cTSH concentrations below and above the reference interval (RI), respectively; in dogs with normal cTSH, a rhTSH stimulation test was performed to confirm the diagnosis. In DNTI, hypothyroidism was excluded upon a negative result of a rhTSH stimulation test. SD were diagnosed based on alteration of temperature, cardiac and respiratory frequency, differential leukocyte count and C-reactive protein concentration above RI. HD were considered healthy upon history and physical examination. Hormone evaluations were performed with LC-MS/MS on surplus serum stored at -80°C. TT₄ and cTSH were measured using a validated immunoassay (Immulite®).

Non-significant differences considering signalment, age and body weight were found between groups. Median TT₄ and fT₄ serum concentrations were significantly higher (p<0.001) in HD compared to DNTI, DWH and SD. Median fT₃ serum concentration was significantly lower in DWH and DNTI compared to SD (p<0.001 and p=0.0091, respectively) and HD (p<0.001 and p=0.0024, respectively). Median rT₃ serum concentration was significantly lower in DWH compared to SD (p=0.0141) and HD (p=0.0128). Median 3.3-T₂ serum concentration was significantly higher in DWH compared to DNTI (p=0.0038) and HD (p=0.0447). There were non-significant differences regarding median 3.5-T₂ serum concentrations among the dogs of the four groups.

Using the ROC curve analysis to differentiate DWH from DNTI+SD, an AUC of 0.86 (p=0.003), 0.76 (p=0.009) and 0.75 (p=0.012) was obtained for fT₃, fT₄ and TT₄, respectively. Values of fT₃ <0.61 pmol/L better discriminated hypothyroidism with 69% sensitivity (95%CI: 39–91%), 83% specificity (95%CI: 63–95%) and accuracy of 0.86 (95%CI: 0.74–0.98).

Although serum fT₃ and fT₄ (LC-MS/MS) have shown better performances than the serum TT₄ (Immulite®) in identifying DWH, the overlap between DWH and DNTI+SD was unfortunately relevant also for the thyroid hormone measurements with LC-MS/MS. Despite the introduction of new analytical methods, the use of dynamic tests (e.g., rhTSH stimulation test) remains the better method to discriminate DWH from DNTI.

DISCLOSURES

No disclosures to report