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ORAL RESEARCH COMMUNICATIONS

ESCG-O-1

The fecal microbiota and unconjugated fecal bile acids in dogs with diabetes mellitus

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Alteration of intestinal and fecal microbiota (dysbiosis), together with changes in fecal bile acid (BA) concentrations have been associated with type 1 diabetes mellitus (DM) in people. DM in dogs resembles human type 1 DM. The aim of this study was to evaluate changes in fecal microbiota and fecal unconjugated BAs profile in dogs with naturally-occurring DM during insulin therapy and compare them to healthy control dogs (HC).

To this aim, naturally-passed fecal samples and left over serum samples obtained for diagnostic proposals from 17 adult dogs with DM were collected. Fecal samples from 16 clinically healthy dogs were used as a control population. In addition, serum folate, cobalamin and cTLI were assessed in the DM group to exclude the presence of other causes of dysbiosis; for each DM dog, the fed diet was known and the use of antibiotic within the previous 12 months was excluded. DNA was extracted from each fecal sample prior to quantitative PCR (qPCR) analysis. Data for 8 bacterial groups was compiled to calculate a microbiota dysbiosis index (DI). Concentrations and proportions of fecal unconjugated primary (cholic/chenodeoxycholic) and secondary (litho/deoxy/ursodeoxycholic) BAs were measured using a gas chromatography mass spectrometry platform. Unpaired *t* test and Mann-Whitney *U* test compared median values between HC and DM dogs with significance set at $P < 0.05$.

Median DI was statistically different between HC and DM dogs (HD −5.3 vs DM -1,5; $P = 0.0019$;). A negative dysbiosis index (DI), indicative of normobiosis, was present in 82% of DM dogs. Four dogs in the DM were dysbiotic (DI index >0). TLI, folate and cobalamin concentrations were normal in all dogs with DM, though two dogs had cobalamin levels <350 ng/L. There was no significant difference between groups for total BAs and proportion of secondary BAs (respectively $P = 0.1124$ and $P = 0.5814$).

The results of this preliminary study did not confirm the association between intestinal dysbiosis, fecal bile acids dysmetabolism and DM in dogs under insulin therapy. Fecal unconjugated BA profiles of DM dogs mimicked those seen in healthy dogs. Further studies are needed to evaluate the possible role of intestinal microbiota in the pathogenesis of canine DM.

Disclosures

No disclosures to report.

ESCG-O-2

Impact of antibiotic administration on fecal bacterial groups potentially associated with dysbiosis in kittens

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In humans, antibiotic use results in long-lasting changes of the bacterial constituents of the gastrointestinal (GI) tract. Antibiotic-induced microbial shifts are associated with predisposition to certain diseases such as chronic GI diseases, obesity, and allergies. No studies exist in kittens investigating the effect of antibiotic administration on the GI microbiota and the duration of this effect. The study aim was to determine the effect of antibiotic administration in kittens on certain bacterial groups that are potentially associated with dysbiosis.

Naturally passed feces were collected from 17 healthy kittens that did not receive antibiotics (Group 1), 14 kittens that received amoxicillin/clavulanic acid for 20 days (Group 2), and 13 kittens that received doxycycline for 28 days (Group 3) as part for standard treatment of upper respiratory tract infection. Kittens were approximately 2 months of age, on the same diet and the same antiparasitic treatment prior to sample collection. Fecal samples were collected on days 0 (before antibiotic treatment), 20 or 28 (Group 2 and Group 3, respectively; after the end of antibiotic administration), and 60. DNA was extracted from each sample and qPCRs were performed for total bacteria, *Turicibacter* spp., *Faecalibacterium* spp., *Streptococcus* spp., *Escherichia coli (E. coli)*, *Blautia* spp., *Fusobacterium* spp., *Clostridium hiranonis (C. hiranonis), and Bifidobacterium* spp. The data were tested for normal distribution and appropriate statistical analyses were used for either repeated or independent measurements. Statistical significance was set at $p < 0.05$ and correction for multiple comparisons was used where appropriate.

On day 0, no significant differences were identified among the 3 groups. On day 20, there were significant increases in *E. coli* ($p = 0.003$) and decreases in total bacteria ($p = 0.002$), *Blautia* ($P = 0.0038$), *C. hiranonis* ($P = 0.026$), and *Faecalibacterium* ($p = 0.031$) in group 2 compared to group 1. On day 60, *E.coli* ($P = 0.012$) was significantly increased in group 3 compared to group 1. In group 1 *E. coli* significantly decreased ($P < 0.0001$) and *Faecalibacterium* significantly increased ($P = 0.023$) over time. In group 2, total bacteria ($P = 0.014$)

and *Blautia* (P = 0.008) significantly decreased on day 20 and *C. hiranonis* (p = 0.001) on day 60. *Blautia* (P = 0.017) remained decreased on day 60. *E.coli* remained unchanged on day 20 (P = 0.999) but decreased on day 60 (P = 0.0001). *Faecalibacterium* remained unchanged on day 20 (P = 0.711) but increased on day 60 (P = 0.0002). In group 3, no statistically significant differences were identified.

Administration of amoxicillin/clavulanic acid or doxycycline had profound effects on certain bacterial groups potentially associated with dysbiosis in these kittens.

Disclosures

No disclosures to report.

ESCG-O-3

Fecal microbial metabolism is altered in dogs with chronic enteropathy

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Several studies have reported intestinal microbial dysbiosis in dogs with chronic enteropathy. Limited data is available about the microbiota gene function in this pathology in dogs. Determining the functional attributes of the microbiome is essential for understanding their role on host metabolism and disease. The aim of this study was to compare the functional roles of the fecal microbiota in healthy dogs and dogs with CE by fecal DNA shotgun sequencing.

Fecal samples were collected from 14 healthy dogs and 20 dogs with chronic enteropathy (CE). Fecal DNA was extracted using a commercial kit (PowerSoil, QIAGEN). Functional characterization of the shotgun sequence reads in the KEGG database was performed using next generation sequencing, in order to identify the relative abundance of specific metabolic pathways. A Wilcoxon test was used for comparison of the gene abundance between groups. Significance was set at q < 0.05.

At phylum level, low abundance of Bacteroidetes was observed in dog with CE, compared to healthy control dogs (48.5 vs 1.6%; q = 0.0006). Fusobacteria was also significantly increased in healthy controls (0.25 vs 0.04%; q = 0.0111). The pathway enrichment analysis of the bacterial metagenomes showed that 130 of 360 (36.1%) total metabolic modules were differentially abundant between studied groups. Genes for carbohydrate metabolism, biosynthesis of amino acids (lysine, threonine, histidine, isoleucine, tryptophan, leucine and serine) and vitamins (ascorbate, thiamine and riboflavin) were decreased in dogs with CE, while genes involved in transport of molecules and homeostasis maintenance during oxidative stress (glutathione biosynthesis) were increased in CE.

Our data presents, as previous reported, an intestinal microbial dysbiosis in dogs with CE. As new finding, our results show an altered microbial metabolism in dogs with CE compared to healthy dogs, characterized by reduction of amino acid biosynthesis and carbohydrate metabolism. Further studies including transcriptomic analysis are warranted to define the consequences of this microbiota dysfunction on dogs with CE.

Disclosures

No disclosures to report.

ESCG-O-4

The pug breed demonstrates a worse response to treatment of protein-losing enteropathy than other breeds of dog

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'Protein-losing-enteropathy' (PLE) is a syndrome caused by various diseases including idiopathic inflammatory bowel disease, primary lymphangiectasia, lymphoma, and severe acute gastroenteritis. Certain breed predispositions, such as the soft-coated Wheaten Terrier, are well known. Our clinical experience suggests that pugs with PLE respond poorly to treatment, but this is not described in the literature. The aim of the current study was to assess whether the pug breed demonstrates a worse response to treatment for PLE than other breeds of dog.

This was a retrospective study comparing the response to treatment in all pugs diagnosed with PLE between 2009 and 2018 in five referral centres in the United Kingdom. Approval for the study was granted by the University of Liverpool Research Ethics Committee. A group of non-pug dogs, also diagnosed with PLE within the same period, was selected for comparison. PLE was defined as any gastrointestinal disease resulting in serum albumin below the laboratory reference interval that could not be explained by another cause. Factors associated with survival were assessed using simple statistics (Mann-Whitney tests and chi-square test as appropriate) and Cox's proportional hazards regression. Initially, factors were tested individually using simple regression; a multiple regression model was then created, subsequently refined by backwards stepwise elimination until the best model was found.

A total of 35 pugs were diagnosed with a PLE between 2009 and 2018 and were compared with 113 dogs from other breeds. On simple regression analysis, factors associated with survival (at P < 0.1) were pug breed (P = 0.002), diet used for treatment (P = 0.022), receiving immunosuppressive therapy (P = 0.089), and treatment with cobalamin (P = 0.005). However, the only factors that remained in the final model were the pug breed and the diet used for treatment. In this respect, dogs of the pug breed were associated with a greater hazard of dying (compared with non-pugs: hazards ratio [HR] 2.67 (CI 1.60-4.47; P < 0.001), whilst being fed a hydrolysed diet was associated with a lesser hazard risk of death than when fed other diets (compared with low-fat diets: HR 0.50, 95%-CI: 0.26-0.97; P = 0.042; compared with other diets including highly-digestible diets: HR 0.35, 95%-CI: 0.17-0.71; P = 0.004).

In conclusion, this study demonstrated an association between the pug breed and a poorer response to treatment for PLE compared to other breeds. Further research should be undertaken as to the underlying cause.

Disclosures

Disclosures to report.

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ESCG-O-5

Is measuring serum folate pointless? Retrospective analysis of prevalence and clinical significance of hypo- or hyperfolataemia in dogs with chronic enteropathies

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Assessment of serum folate (SF) is routinely performed in dogs with chronic enteropathies (CE), most often in conjunction with serum cobalamin. Traditionally, their combination has been used to differentiate intestinal malabsorption from dysbiosis. Despite the fact that the diagnostic and prognostic value of serum cobalamin is well documented, the prevalence of hypo- and hyperfolataemia and the clinical and prognostic value of its assessment has not been scrutinised in dogs with CE. The aims of this study were to determine the prevalence of SF abnormalities in dogs with CE, as well as its relationship to other laboratory parameters and outcome. Files of dogs presented for chronic gastrointestinal (GI) signs (> 3 weeks duration) between 2014 and 2017 were retrospectively evaluated. Exclusion criteria were lack of SF assessment, and supplementation of folate or cobalamin beforehand. 321 dogs (100 FN, 109 MN, 34 FE, 78 ME; median age of 65 m, range 2-171) were included. Hypofolataemia was present in 97/321 (30%), hyperfolataemia in 86/321 (27%), with the remaining 138/321 dogs (43%) having normal SF values. Initially, dogs were divided into groups according to the final diagnosis: CE (n = 215), other GI disease (n = 76), non-GI disease (n = 30). SF values were not significantly different across those groups (Kruskal-Wallis, $P = 0.83$). When dividing CE dogs into different subgroups of food- (FRE), antibiotic- (ARE), steroid-responsive (SRE) or protein-losing enteropathy (PLE), no difference in SF values was observed (ANOVA, $P = 0.92$). When all dogs were grouped by their SF status (low, normal, high), significant differences in serum cobalamin ($P = 0.001$), alkaline phosphatase (p = 0.01), cholesterol (p = 0.03) and total calcium (p = 0.01) were identified (ANOVA, Dunn's post hoc test). Multivariate analysis confirmed the correlation between SF and cobalamin ($P = 0.009$), as well as cholesterol ($P = 0.028$) and total calcium (p = 0.038). However, none of these correlations were linear (Spearman, all $P > 0.05$). Kaplan Maier analysis of follow-up and survival times by SF status showed no significant differences. In conclusion, SF was not associated with GI disease or subgroup of CE. Improvement of clinical signs or survival was not associated with SF status. In this study, no diagnostic or prognostic benefit of assessing SF in dogs with chronic GI signs could be detected. Future prospective studies should assess if folate supplementation in the 30% of CE dogs with hypofolataemia can accelerate clinical improvement or influence outcome/ prognosis.

Disclosures

No disclosures to report.

ESCG-O-6

Dogs with acute haemorrhagic diarrhoea syndrome not receiving antibiotics have a good prognosis despite initial high AHDS-score and systemic inflammation

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Acute haemorrhagic diarrhoea syndrome (AHDS) in dogs is often treated with antibiotics due to the potential risk of bacterial translocation from the gastrointestinal tract to the blood stream. However, recent studies indicate that antibiotics are not always necessary. According to the Danish antibiotic use guidelines for companion animals 2012, antibiotics are not recommended for routine treatment of AHDS but only indicated in hospitalized dogs with severely affected overall condition and signs of systemic inflammation (SIRS)/sepsis.

The aim of this study was to evaluate severity of disease and outcome in hospitalized dogs with acute haemorrhagic diarrhoea that did not receive antibiotics. The study was performed as a retrospective, observational study based on information from medical records for dogs with acute haemorrhagic diarrhoea of unknown aetiology, hospitalized at the University Hospital for Companion Animals during the period 25/2-2014 to 9/10-2018.

Signalments, concurrent diseases, clinical disease at the time of hospitalization and during each consecutive day, treatment prior to and during hospitalization, days of hospitalization/euthanasia and laboratory results were registered for each patient. Clinical disease was scored according to the AHDS-scoring system from 0-18 and the number of SIRS criteria (tachycardia (HR > 120), tachypnea (RR > 40), hyper or hypothermia (T > 39.0°C or < 37.5°C), leucocytosis (WBC > $18*10^9$/L), leukopenia (WBC < $5*10^9$/L, band neutrophilia and/or hypoglycaemia [glucose <4 mmol/L]) were recorded.

One-hundred and seventy-two dogs were excluded from the analysis due to suspected drug induced disease (vaccination(s) (N = 4), anaesthetics (N = 5), corticosteroids or NSAIDs (N = 81)), alimentary foreign body (N = 5) or treatment with antibiotics during hospitalization (N = 128).

Of the 128 dogs, where an obvious cause for the diarrhoea were not found and that only received supportive treatment (intravenous fluid therapy N = 128; antiemetics N = 98; gastroprotectants N = 107), 98% survived to discharge (125/128 dogs). Two dogs were euthanized due to financial constraints and reluctance from the owner to proceed with further treatment due to advanced age and one brachycephalic dog suffered a respiratory crisis with respiratory arrest non-responsive to resuscitation. The surviving 125 dogs were hospitalized for an average of 1.7 days (range 1-4 days) with a mean AHDS-score of 12 at hospitalization (range 4-16). The mean AHDS-score after 24 hours of hospitalization was 5.5 (range 0-14). 29% (37/128) of the

dogs met ≥2 SIRS criteria during hospitalization. None of the 128 dogs had a degenerative left-shift.

These results suggest that antimicrobial therapy in dogs with acute haemorrhagic diarrhoea may not always be necessary in dogs even when 2 or more SIRS-criteria are met.

Disclosures

No disclosures to report.

ESCG-O-7

Faecal bile acid profiles in dogs with acute haemorrhagic diarrhoea syndrome over time and compared to healthy dogs

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Recent studies have shown alterations in faecal bile acid (BA) profiles in dogs with chronic enteropathy, potentially contributing to clinical signs. The study aim was to assess faecal BA concentrations in dogs with acute haemorrhagic diarrhoea syndrome (AHDS-D) over time and to compare it to that of healthy control dogs (HC-D).

Twenty-five AHDS-D and 53 HC-D were enrolled. Faecal BA concentrations were measured by gas chromatography-mass spectrometry on days 0, 2, 7, and 14 in AHDS-D, and on day 0 in HC-D. Statistical analysis was performed with Friedman test for comparison of BA over time in AHDS-D, and unpaired t-test or Mann-Whitney-test for comparison between AHDS-D and HC-D ($P < 0.05$).

On day 0, concentration of lithocholic acid was significantly lower in AHDS-D than in HC-D ($p < 0.001$). On day 2, total faecal BA were significantly increased in AHDS-D ($p < 0.001$) compared to HC-D. More precisely, primary BA were significantly increased in AHDS-D on day 2 ($P = 0.034$) with significantly higher concentration of cholic acid ($p = 0.004$), while secondary BA were significantly decreased ($p < 0.001$) with significantly lower concentrations of deoxycholic acid ($p < 0.001$) and ursodeoxycholic acid (UDCA) ($p < 0.001$). Dogs in AHDS-D showed a significant decrease in total primary BA concentrations ($P = 0.021$) with significantly lower concentrations of cholic acid ($P = 0.027$) and deoxycholic acid ($p = 0.021$) on day 14 compared to day 2.

In conclusion, AHDS-D show alterations in faecal BA profiles compared to HC-D. However, faecal BA profiles normalize rapidly suggesting that BA dysmetabolism does not seem to play a major role in the pathophysiology of AHDS.

Disclosures

No disclosures to report.

ESCG-O-8

Long-term consequences of acute hemorrhagic diarrhea syndrome in dogs

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Destruction of the intestinal barrier and microbiota dysbiosis especially around the time of weaning represent important mechanisms for allergic sensitization. Consequently, 42% of young dogs surviving a canine parvovirus (CPV) infection develop chronic gastrointestinal disorders later in their lives, but it is unknown if adult dogs with severe intestinal lesions also have an increased risk for developing chronic gastrointestinal disorders. The aim of this study was to evaluate, whether dogs with acute hemorrhagic diarrhea syndrome (AHDS) have a higher prevalence of chronic enteropathies later in life.

Forty dogs diagnosed with AHDS, for which a follow-up of at least 12 months was available, were included in the study. A historical control group of 67 dogs without history of gastroenteritis was included to enable risk assessment. Dog owners were asked to complete a questionnaire. The percentage of dogs with signs of chronic enteropathies in both groups were compared using Fisher's exact test.

There was no significant difference between AHDS and control dogs concerning development of chronic enteropathies (AHDS 22.5%; controls 12.0%; $P = 0.177$) during their observation time (AHDS: median 4 years, range 1-12 years; controls: median 5 years, range 1-12 years).

The results of this study suggest that dogs that experience an episode of AHDS do not have an increased risk for developing chronic gastrointestinal disease later in life, which is different to the risk of young dogs with CPV infection. Thus, timing of the intestinal barrier dysfunction might represent one main risk factor.

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Disclosures

No disclosures to report.

ESVC-O-1

Acute effect of oral pimobendan on left atrial function and mitral valve regurgitation severity in dogs with stage B2 myxomatous mitral valve disease - A pilot study

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Myxomatous mitral valve disease (MMVD) is associated with failure of the mitral valve (MV) apparatus. In a proportion of affected dogs, MV regurgitation is severe enough to cause an increase in left atrial (LA) volume and pressure. LA function was reported to decline with increased disease severity. Pimobendan, a phosphodiesterase III inhibitor exerting positive inotropic and vasodilatory effects, was shown to decrease LA pressure, although its effect on LA function is not well established. This study aimed to prospectively evaluate the acute effect of pimobendan on LA function and mitral regurgitation fraction in dogs with stage B2 MMVD.

Sixteen dogs in stage B2 MMVD were included in this prospective interventional study. Echocardiograms were performed at presentation and 3 hours following a single-dose pimobendan. Two-dimensional, M-mode, and Doppler images were recorded from the

right parasternal and left apical standard views. In all dogs, left atrial volume was measured using the biplane area-length method at three time-points: immediately before MV opening, at onset of P-wave, and at MV closure. Reservoir, conduit and active pump functions were calculated, as previously described. In ten dogs, MV regurgitation volume was calculated by subtracting the forward stroke volume (Aortic outflow velocity time integral multiplied by the aortic cross-sectional area) from the total left ventricular (LV) stroke volume (End-systolic LV volume subtracted from the end-diastolic LV volume, both measured using Simpson's method of discs). MV regurgitation fraction was calculated as the percentage of regurgitation volume from the total stroke volume. Paired Student *t*-test and Wilcoxon Signed-Rank Test were used to compare the results.

Compared with baseline, LA volume was significantly lower on post-pimobendan measurements immediately before MV opening ($P < 0.01$), at P-wave onset ($P < 0.01$) and at MV closure ($p = 0.02$). However, LA conduit, reservoir and active pump functions did not change significantly. MV regurgitation fraction post-pimobendan (27.7 +/− 5.3) was significantly lower ($p < 0.01$) compared with baseline (43.7 +/− 4.7), forward stroke volume was significantly increased ($P = 0.015$), while total stroke volume did not change significantly.

This study suggests that despite an acute reduction in LA volume, pimobendan may not exert a measurable effect on LA function. The beneficial decrease in left atrial volume appears to result from an acute reduction in MV regurgitation fraction. The mechanisms by which pimobendan reduces regurgitation fraction are thought to be associated with improved forward flow secondary to its inotropic and vasodilatory effects, in addition to reduced end-diastolic LV dimension, minimising secondary functional mitral regurgitation.

Disclosures

No disclosures to report.

ESVC-O-2

Retrospective evaluation of the safety and tolerability of pimobendan in cats with obstructive versus nonobstructive hypertrophic cardiomyopathy

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Pimobendan is frequently used off-label for treatment of cats with congestive heart failure (CHF) secondary hypertrophy cardiomyopathy (HCM). Concerns exist regarding the safety of pimobendan in the subset of cats with HCM and dynamic outflow tract obstruction (HOCM). The purpose of this study was to evaluate safety and tolerability of pimobendan in cats with CHF secondary to HOCM compared with nonobstructive HCM.

Medical records from 94 cats with CHF (47 with HOCM, 47 with nonobstructive HCM) at two tertiary referral hospitals were reviewed. Demographic, clinicopathologic, echocardiographic, and treatment data were collected and compared between groups, including information regarding possible adverse effects of pimobendan.

Average age of cats (9 +/− 4 years) did not differ between HOCM and HCM ($P = 0.12$). Compared to cats with HCM, cats with HOCM were more likely to manifest CHF as pulmonary edema (44/47 versus 32/47; $P = 0.003$) and less likely to have pleural effusion (13/47 versus 25/47; $P = 0.02$). Other than a higher incidence of heart murmurs in cats with HOCM ($P < 0.001$), clinical variables did not differ between groups. Pimobendan was typically initiated on the date of CHF diagnosis (median time from diagnosis of CHF to initiation of pimobendan was 0 days). Initial dose of pimobendan was 0.25 +/− 0.07 mg/kg every 12 hours; dose (or frequency) was escalated at some point during CHF management in 31/94 (33%) of cases, with no difference between HOCM and HCM cats.

Clinical signs that could potentially represent adverse effects of pimobendan (vomiting, diarrhea, anorexia, lethargy, new-onset arrhythmias) were noted in 13/47 (28%) HCM cats and 9/47 (19%) HOCM cats ($P = 0.34$). Based on patterns of timing and resolution, these signs were generally ascribed to recurrence of CHF rather than pimobendan administration. Pimobendan was discontinued due to adverse effects in only 1 cat with nonobstructive HCM that experienced lethargy and nausea 2-3 hours following pimobendan administration (resolved when pimobendan discontinued). Pimobendan was discontinued in 7 additional cats, either because owners were unable to administer the medication (n = 2) or because CHF had resolved (n = 5 cases where CHF was precipitated by an acute external event, such as fluid overload or injectable glucocorticoid administration). No cats experienced acute adverse hemodynamic effects (hypotension, cardiovascular collapse) following pimobendan administration.

Results of this study suggest that pimobendan is well-tolerated in cats with cardiomyopathy and CHF, regardless of presence of dynamic outflow tract obstruction.

Disclosures

Disclosures to report.

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ESVC-O-3

Aorto-septal angle, isolated basal septal hypertrophy and systolic murmur in 122 cats

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Systolic heart murmurs (SM) are commonly diagnosed in healthy cats. The aim of this study was to determine whether the aorto-septal angle (AoSA) is associated with SM in cats. More, we hypothesised that SM are related to the presence of septal bulge, systolic anterior motion of the mitral valve (SAM) and increased aortic flow velocity (AoV). Between November 2014 and February 2018, 316 client owned cats referred for a cardiology evaluation were prospectively examined. Regardless the presence of a systolic murmur, cats with a

normal echocardiographic exam, normal blood pressure (systolic pressure < 160 mmHg) and euthyroid were included in the study. Cats with normal diastolic thickness of the left ventricle, but isolated basal septal hypertrophy (IBSH) ≥6 mm or SAM, were also included. The AoSA was measured from the right parasternal five chambers view, based on the published guidelines in the dog. A total of 316 cats were examined, 122 fulfilled the inclusion criteria. A left parasternal SM was found in 39 cats (32%). In 10 cats the murmur was audible only after stressed auscultation. SM were associated with a narrower AoSA (P < 0.001), higher prevalence of IBSH (P < 0.01) and higher AoV (P < 0.001), compare to cats without a murmur. The IBSH increases with aging, with an increase of 0,11 mm per year. The AoSA decreases with aging (P < 0.001), with a reduction of 0.55° per year. The AoSA was narrower in cats with IBSH (P < 0.001) and IBSH was always present in cats with AoSA<120°. The AoSA did not differ between cats with or without SAM (P = 0.853). There was a non-linear correlation between AoSA and AoV. In conclusion AoSA angle and the remodeling of the interventricular septum may be correlated and can be a cause of systolic murmur in apparently healthy cats. These findings resemble the sigmoid septum found in human, which can be considered a morphologic variant of the ventricular septum, age related and associated with a systolic murmur.

Disclosures

No disclosures to report.

ESVC-O-4

Accuracy of noninvasively determined pulmonary artery pressure in dogs with myxomatous mitral valve disease (MMVD)

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Development of pulmonary hypertension is an independent predictor of poor outcome in dogs affected by myxomatous valvular degeneration (MMVD). Pulmonary arterial pressure is routinely estimated by applying the simplified Bernoulli equation to the velocity of tricuspid regurgitation (PASP_D). The accuracy of this estimation is unknown in dogs with MMVD, but experimental studies suggest that the method is imperfect. We prospectively enrolled dogs affected by ACVIM stages B2 and C MMVD for which treatment had been unchanged for at least one month. A flow-directed thermodilution monitoring catheter was percutaneously placed in the right jugular vein and advanced to the main pulmonary artery. Pulmonary arterial systolic pressure was recorded (PASP_C). A second operator simultaneously acquired tricuspid regurgitant velocity spectra to calculate PASP_D. Each operator was blinded to the result of the other technique. Twenty dogs were enrolled. Technical difficulties prevented catheterization in 2 dogs. Eighteen measurement pairs were therefore used for comparison of PASP_C and PASP_D through Bland-Altman analysis and linear regression. A statistically significant bias between PASP_C and PASP_D (mean difference = 0.5 mmHg; Confidence interval: −6.5 mmHg, +7.5 mmHg) was not detected. The limits of agreement between the techniques were wide (−27.3 mmHg, +28.2 mmHg). Regression analysis failed to identify a significant linear association between the two techniques (r = 0.11, p = 0.17). In conclusion, PASP_D estimation poorly agrees with PASP_C measurement in dogs affected by MMVD in ACVIM stages B2 and C. In these dogs, PASP_D could under- or over-estimate PASP_C by more than 20 mmHg, and therefore caution should be used when interpreting PASP_D.

Disclosures

Disclosures to report.
Michele Borgarelli receives financial support from Ceva Sante Animale for other work not related to this abstract.

ESVC-O-5

Left atrial tear in dogs with myxomatous mitral valve disease - clinical presentation, echocardiographic features and long-term survival

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Left atrial tear (LAT) is an acute, life-threatening, rare complication in canine myxomatous mitral valve disease (MMVD). This case-control matched multicenter retrospective study focussed on clinical manifestation and long-term survival in LAT dogs compared to control dogs with MMVD but without LAT.
Data were collected from the veterinary cardiology departments' databases, selecting patients with advanced MMVD associated LAT. Signalment, clinical presentation, echocardiography, selected laboratory findings, the cause of death and survival data were evaluated. Parameters were assessed upon LAT diagnosis (time 0, T0) and at the resolution of pericardial effusion (time 1, T1). Control dogs with similarly advanced MMVD were selected based on quantitative echocardiographic parameters matching those of LAT dogs at T1.
Thirty client-owned dogs were included: 15 dogs with MMVD associated LAT and 15 control dogs without LAT. In both groups, 9 dogs were in ACVIM congestive heart failure stage C and 6 in B2 MMVD. Commonly observed clinical signs included dyspnoea, syncope and weakness. No significant differences were found in age (both groups 10 ± 2 years, *P = 0.89*), body weight (both groups 7 ± 5 kg, *P > 0.99*), gender distribution (LAT group: 7 females, control group: 10 females, *P = 0.46*), serum creatinine concentration (LAT group: 131 umol/L ± 72, control group: 108 umol/L ± 39, *P = 0.42*) or echocardiographic variables between the groups. Mean left atrial to aortic diameter ratio was 2.24 ± 0.4 at T0, 2.53 ± 0.4 at T1 for dogs with LAT and 2.26 ± 0.4 for the controls. Left ventricular internal diameter normalized to body weight at end diastole was 1.15 ± 0.3 at T0, 1.29 ± 0.2 at T1 for dogs with LAT and 1.25 ± 0.3 for the controls. Pericardiocentesis was performed in 3/15 dogs. At study termination, all dogs in LAT and 10 in the control group had died from cardiac causes. Five dogs with LAT had died in the first week post admission as compared to 1 control dog. The mean survival time for LAT dogs was 53 days. When excluding the animals that died in the first week, survival times were 427 for dogs with LAT and 371 days for the control dogs (*P = 0.617*).

In conclusion, dogs with MMVD associated LAT bear a high mortality risk, especially in the first week post the event. However, once survived this critical time, LAT does not seem to significantly impact the long-term survival.

Disclosures

No disclosures to report.

ESVC-O-6

Echocardiographic evaluation of left ventricular dimension and systolic function before and 24 hours after percutaneous closure of patent ductus arteriosus in 120 dogs

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One hundred and twenty dogs were enrolled to value the effect of loading condition changes on left ventricular volumes before and 24-hours after the patent ductus arteriosus (PDA) occlusion by ACDO using standard echocardiography. The animals were divided in pure breed (n. 94) and mixed breed (n. 26); subsequently, the pure breed dogs were divided on the basis of the size of the breed of belonging in 3 groups (small size n. 36; medium size n. 8; large size n. 50). Moreover, the animals were divided in three classes based on their age: until 6 months; 6-12 months; over 12 months. A significant reduction of all the examined parameters (LVIDd, LVIDs, EDV, ESV, EDVI, ESVI, FS) was observed after ductal closure. The evaluation of the relative percentage difference (RDP) of the echocardiographic parameters showed at 24-hours after the closure, a significant reduction higher in small size breed than in large size breed dogs. No significant difference related to breed size was observed only for RPD_FS variable. A significant interaction effect, between breed size and age classes, was observed only for RPD_EDVI ($F = 3.4$; $P = 0.039$). Until six months of age there was no significant difference in RPD_EDVI reduction, but over 6 months a significant reduction between small size and large size breed dogs at 24-hours from the occlusion was observed.
In conclusion, our data seem to indicate that small breed dogs show a greater tolerance to congenital volume overload, and for this reason it could be possible to delay the PDA closure of a few months allowing weight gain that makes easier the interventional procedure. On the other hand, the large breed dogs should be submitted to ductal closure as soon as possible, in order to avoid an excessive LV wall stress.

Disclosures

No disclosures to report.

ESVC-O-7

Delayed Electrolyte Depletion and Azotemia in a Furosemide Rate Continuous Infusion Model

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Intravenous furosemide is the mainstay of treatment for acute congestive heart failure in dogs, however, the potential for delayed effects on hydration, electrolytes and renal function have not been studied. This study sought to evaluate these parameters in normal dogs receiving furosemide continuous rate infusion (CRI) with or without renin-angiotensin-aldosterone system inhibitors.
Ten healthy dogs were studied in a 3-way randomized, cross-over design. Dogs orally received either placebo, benazepril, or benazepril +spironolactone for 3 days prior to 5-hour furosemide CRI 0.66 mg/kg/hr. Body weight (BW), renal values, serum electrolytes, packed cell volume and total protein were measured before oral medications, hour 0 and 5 of the furosemide CRI, and hour 24. Variables were compared between time-points and treatments.
Loss of BW during the CRI exceeded recovery at 24 hours and hemoconcentration occurred, with incomplete return to baseline at 24 hours. Blood urea nitrogen and creatinine were unchanged during the CRI but increased 24 $\pm$ 12% at 24 hours. Serum sodium did not change during the CRI but decreased at 24 hours. Serum chloride decreased at hour 5 and did not return to baseline at 24 hours. Hypochloremic metabolic alkalosis and increased anion gap present at hour 5 did not normalize at 24 hours. No differences between treatments were found.
Some furosemide CRI-related biochemical changes were delayed for 24 hours while others evident at hour 5 only partially improved at hour 24 in these normal dogs. These findings have implications for clinical patients with renal dysfunction, or receiving higher doses or longer furosemide infusions.

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ESVC-O-8

Changes in renal endothelin activity with cardiac, renal and other chronic diseases in dogs

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Pathways that disrupt the cardiovascular-renal axis in dogs are incompletely defined. The renal endothelin (ET-1) system may play a key role because it regulates blood pressure and sodium homeostasis in the kidney, but also mediates vascular dysfunction and pro-fibrotic/inflammatory changes that increase cardiovascular risk in people. Urinary ET-1 (UET-1) is a marker of renal vascular and tubular ET-1 activity, and so could provide insight into changes in renal ET-1 signalling that contribute to cardiovascular-renal interactions in dogs. We hypothesised that renal ET-1 activity increases in advanced canine chronic cardiac and renal diseases.
In this pilot study, we compared UET-1 and cystatin C (a marker of renal injury/dysfunction) concentrations in surplus urine from four

groups of dogs presented to the R(D)SVS: healthy (n = 18), chronic kidney disease (CKD; IRIS stages 2-4; serum creatinine 257 µmol/l, IQR395; n = 11) cardiac disease (ACVIM classification B1-C; n = 39) and non-cardiorenal chronic disease (n = 14). Urine was free-catch and owner-collected on the morning of appointment. Samples were excluded if they contained active sediment, and were stored at −80°C before batch analysis.

Both UET-1 and cystatin C were measured by commercial ELISAs and indexed to urinary creatinine concentration (enzymatic method). Comparisons by one-way ANOVA yielded non-transformable residuals so Kruskal-Wallis with Dunn's *post hoc* tests were used (significance P < 0.05).

There was a marked increase in UET-1 excretion in dogs with stage C heart disease (0.69,IQR 1.61 pg/mg; n = 11) compared to healthy dogs (0.02 pg/mg, IQR 0.11; *P* = 0.02). UET-1 excretion was also increased in stage B2 heart disease (0.25 pg/mg, IQR 1.70; *P* = 0.03; n = 19) although to a lesser degree. This was predominantly due to increases in dogs with MMVD (0.38 pg/mg, IQR 1.75; *P* = 0.046; n = 7) rather than those with non-MMVD cardiac diseases (0.23 pg/mg, IQR 0.91). al increases in UET-1 in CKD and chronic disease groups were not statistically significant.

By contrast, CKD markedly increased urinary cystatin C excretion from below limits of detection to (0.53 ng/mg, IQR 2.44; P < 0.0001), while only modest increases (*P* = 0.03) were observed in dogs with chronic disease (0.02 ng/mg, IQR 0.07), and none at all in heart disease stages B1-C.

Renal ET-1 activity increases with congestive heart failure, but surprisingly, also increases in MMVD before congestion develops. Neither increase is associated with renal injury or is a consequence of a chronic disease state. Renal ET-1 may mediate pathophysiological cardiovascular-renal interactions in MMVD that are different to those in isolated CKD and the development of congestive heart failure.

Disclosures

No disclosures to report.

ESVC-O-9

High grade AV Block and third degree AV Block in cats: a retrospective study of epicardial pacemaker implantation (2006-2018) focusing on signalment, presentation and survival

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Third degree atrioventricular block (AVB) is characterised by complete atrioventricular (AV) dissociation, causing independent atrial and ventricular rhythms. Persistent third degree AVB (PAVB) is most commonly described. Another form of AVB, where AV dissociation is intermittent (IAVB) is also recognised. In cats, AVB can be associated with underlying cardiac or systemic diseases. When present, clinical signs associated with these forms of AVB can include weakness, lethargy and syncope. If clinical signs and in particular syncope are present, epicardial pacemaker implantation represents an effective treatment. The aim of the study was to retrospectively assess presentation, echocardiographic data, comorbidities and outcome from cats diagnosed with AVB (PAVB or IAVB) in a single referral hospital, including those that underwent pacemaker implantation. Non parametric testing and Kaplan Meier curves with log rank testing were performed. Sixty-four cats were included over a 12-year period. Forty-three cats had PAVB, 21 had IAVB. Median age of presentation was 13 years, with no difference between AVB type (*P* = 0.752). Thirty-five cats were male and 29 female. Forty-four cats were referred for cardiac complaints (syncope, arrhythmia or dyspnoea), 8 cats had non-specific signs (lethargy) and in 12 cats AVB was an incidental finding. Cats with IAVB were more likely to present with syncopal events (p = 0.005). The median duration of clinical signs prior to presentation was 21 days (1-1138). Twenty-nine cats had echocardiographic changes, left ventricular hypertrophy (17), chamber dilation (12); 13 cats presented with congestive heart failure (CHF). Forty-five cats had one or more comorbidities, the most common were hyperthyroidism (16), diabetes mellitus (9), azotaemia (8). Fifteen cats underwent epicardial pacemaker implantation, mainly cats with IAVB (9/15). Five cats had minor complications (lead dislodgement, pacemaker undersensing, exit block) and 12 cats showed no further clinical signs since implantation. Forty-seven cats died; all-cause mortality median survival time was 799 days (0-2965) and no difference in survival was observed in cats that presented with CHF (*P* = 0.052), IAVB (*P* = 0.082), had comorbidities (*P* = 0.683) or pacemaker placement (p = 0.089). Death due to cardiac cause occurred in fewer cats (17/47), with shorter survival than all-cause mortality (65 days, *P* = 0.003). CHF on presentation was associated with cardiac death (*P* < 0.001). The results of this study showed a variable outcome in cats with AVB. Cardiac death occurred in the minority of cat and was associated with CHF at presentation. Most cats have comorbidities, which did not affect all-cause mortality. Pacemaker implant controlled clinical signs in the majority of cats.

Disclosures

No disclosures to report.

ESVC-O-10

Diet-induced reduction of cardiac wall thickness, Troponin-I and IGF-1 in cats with asymptomatic hypertrophic cardiomyopathy

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Complete and balanced diets, one test and one control, were evaluated in this prospective, randomized, double-blind, multicenter study for effect on clinical, biochemical and echocardiographic parameters in forty-four client-owned cats with asymptomatic hypertrophic cardiomyopathy (aHCM).

Cats with diastolic interventricular septum (IVSd) and/or left ventricular wall (LVWd) thickness ≥ 6 mm were included after informed owner-consent. Examination of non-sedated, fasted cats before and after 6 and 12 months of test or control diet included auscultation, bodyweight (BW), body condition score (BCS) and echocardiography. Wall

thicknesses measured by M- and 2D-mode at basal, mid (LVWd, IVSd) and apical (IVSd) level were recorded as maximum (max-), sum- and number of areas ≥6 mm (n-)). Blood analysis included NT-proBNP, ultra-sensitive troponin-I (c-TnI), serum amyloid A (SAA), insulin, glucose and IGF-1. Linear and generalized mixed models analyzed diet, time and diet-time interactions with significance level of 5%.

There was a significant diet-time interaction for heart-rate (p = 0.032) and IGF-1 (P = 0.020). Test but not control diet showed a significant decrease over time for max-IVSd (P = 0.011), n-IVSd (P < 0.001), sum-IVSd (P < 0.001), M-IVSd (P = 0.023), max-LVWd (P = 0.002), n-LVWd (P = 0.035), sum-LVWd (P = 0.006), M-LVWd (P < 0.001) and n-(IVSd + LVWd) (P = 0.001), IGF-1 (P = 0.005) and cTnI (p = 0.001). No significant changes in BW, BCS or effect of cardiac medication were observed. Cats with left atrium remodeling separately had significantly decreased n-IVSd (P = 0.014), sum-IVSd (P = 0.003) and M-LVFWd (P < 0.001).

A reverse effect on primary echocardiographic parameters of aHCM, with decreased c-TnI and IGF-1 was observed with the test diet. Further research is needed to evaluate the effects on clinical outcome.

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ESVC-O-11

Biomarker discovery in cats with cardiomyopathy

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Cardiomyopathies are frequent in cats and their diagnosis relies on a combination of physical examination, thoracic radiography, electrocardiography and echocardiography. Additionally, in the last decade, assays to measure circulating biomarkers of heart disease, such as cardiac troponin I and N-terminal pro-brain natriuretic peptide (NT-proBNP), have become available for cats. These assays have been proven useful in the diagnosis of feline cardiomyopathies, although in certain cases they may be less reliable. Therefore, the aim of this study was to detect new circulating proteins that may improve the identification of cats affected by cardiomyopathy using biomarker discovery. Client-owned cats were prospectively enrolled. Evaluation comprised complete blood count and biochemical profile with T4 determination, blood pressure measurement, thoracic radiography and echocardiography. Based on diagnosis, cats were allocated to 5 groups, namely symptomatic cardiomyopathy with signs of congestive heart failure (group CM-Sx), asymptomatic cardiomyopathy (group CM-aSx), respiratory diseases (group Resp-Ds), systemic diseases without systemic hypertension (group Sys-Ds), and healthy controls (group Healthy). Plasma samples were processed untreated or following enrichment in low-abundant proteins, submitted to mass spectrometry and their protein profiles compared with statistical software. Putative biomarkers were evaluated by western immunoblotting experiments. Eighty-nine cats were included; 9 in CM-Sx, 9 in CM-aSx, 14 in Resp-Ds, 30 in Sys-Ds and 27 in Healthy. In CM-Sx, 7/9 cats had hypertrophic cardiomyopathy and 2/9 restrictive cardiomyopathy; lung edema was identified in 7, pleural effusion and aortic thromboembolism in 2. In CM-aSx, all cats had hypertrophic cardiomyopathy. By mass spectrometry, several differential proteins were identified among groups. Putative biomarkers were tested by western immunoblotting and 2 of them (Protein 1 and Protein 2), were differentially detected in CM-Sx, compared to the others. In particular, Protein 1 was identified in 6/9 (66.7%) cats in CM-Sx, while plasma samples from cats in CM-aSx, Resp-Ds, Sys-Ds and Healthy had a visible band in 2/9 (22.2%), 2/14 (14.3%), 1/30 (3.3%) and 1/27 (3.7%), respectively (P < 0.001). Protein 2 was detected in the plasma of 4/9 (44.4%) cats in CM-Sx, 1/9 (11.1%) in CM-aSx and 1/30 (3.3%) in Sys-Ds, while no bands were detected in Resp-Ds and Healthy (P = 0.005). These data suggest 2 novel putative biomarkers for differentiating cats with symptomatic cardiomyopathy. Further studies with a higher number of cardiomyopathic cats and different symptomatic status are mandatory to optimize the tests and evaluate sensitivity and specificity of the candidate biomarkers.

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ESVC-O-12

Blood pressure measurement by High Definition Oscillometry in different clinical settings in healthy cats

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Clinical environment can be stressful for cats, thereby affecting blood pressure (BP) recordings.

The aim was to investigate how different clinical settings, and the order (sequence) in which settings are performed, affect BP recordings in healthy cats.

Ninety-six healthy cats were prospectively included. The health examination included physical examination, echocardiography, hematology, and biochemistry. Blood pressure was measured with a high-definition oscillometric (HDO) device, with cuff on tail in three clinical settings; (1) cat taken out of its carrier and placed on examination table with veterinarian present, (2) cat in its own carrier with veterinarian present, or (3) cat in its own carrier without veterinarian present. The owner was

present in all settings. The sequence of clinical settings was randomized and 4-6 recordings were made in each setting.

The combined effect of setting and sequence was associated with lower systolic (SBP), mean arterial (MAP) and diastolic BP (DBP) when BP was measured by the owner with the cat in its own carrier last in sequence (all; $P < 0.0039$). Heart rate was higher when BP was measured with cat on examination table first in the sequence ($P < 0.0006$). When measurements were made with the cat on the examination table, higher coefficients of variation (CVs) were found for SBP, MAP, DBP and HR (all; $P < 0.0001$).

In conclusion, measuring BP with cat in its own carrier gave lower BP, HR and CVs, compared to measurement on examination table. As sequence affected BP with lower values when recordings were made last by owner, time might influence results.

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No disclosures to report.

ESVC-O-13

Does pleural effusion protect against arterial thromboembolism in feline congestive heart failure?

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Aortic thromboembolism (ATE) is a frequently-seen cardiac complication in cats and left atrial (LA) enlargement is considered a risk factor. In dogs, activation of coagulation followed by fibrinolysis occurs in all types of pleural effusions and dogs with pleural effusion of any type or with ascites secondary to congestive heart failure (CHF) show an enhanced systemic fibrinolysis, which may decreases clot formation. Therefore, the aim of this study was to determine whether cardiopathic cats with pleural effusion were less likely to develop aortic thromboembolism (ATE) than cats without pleural effusion.

Cross-sectional study retrospectively evaluating client-owned cats with heart disease presented between 2004 and 2018. All cats included underwent a full echocardiography evaluation and thoracic radiographs. Cats were divided into 3 groups: without CHF (group 1), with cardiogenic pulmonary edema (group 2) and with pleural effusion (group 3). Frequency of ATE among groups was compared by chi-square test. The LA diameter and the LA/aorta ratio (LA:Ao) were also compared between cats with and without ATE (T-test) and also among the 3 groups (Anova followed by Tamhane post-hoc analysis).

In the study were included 629 cats (group 1 = 420, group 2 = 71, and group 3 = 138). Sixty-one cats at time of presentation had ATE, overall prevalence of 9.7%. LA in cats with ATE (20.16 $\pm$ 4.48 mm) was significantly (t = 8.90, $P < 0.001$) bigger than in cats without ATE (15.23 $\pm$ 4.7). LA:Ao in cats with ATE (2.25 $\pm$ 0.49) was significantly (t = 8.16, $P < 0.001$) higher than in cats without ATE (1.68 $\pm$ 0.52). Frequency of ATE was statistically (χ^2 = 47.29, $P < 0.001$) different among the 3 groups (group 1, 30/420 [7.1%]; group 2, 23/71 [32.4%]; group 3, 8/138 [5.8%]). LA was significantly (F = 79.3, $P < 0.001$) increased in cats of group 2 (19.80 $\pm$ 5.31 mm) and group 3 (18.17 $\pm$ 3.97 mm) compared to group 1 (14.20 $\pm$ 3.38 mm), while no significant difference was present between groups 2 and 3. LA:Ao was significantly (F = 66.6, $P < 0.001$) increased in cats of group 2 (2.24 $\pm$ 0.55) and group 3 (2.10 $\pm$ 0.51) compared to group 1 (1.53 $\pm$ 0.43), while no significant difference was present between groups 2 and 3.

As expected, cats with CHF (with edema or pleural effusion) had a bigger LA compared to the cats without CHF. Moreover, also cats with ATE presented a larger LA atrium compared to the cats without ATE. Nevertheless, when pleural effusion is present frequency of ATE remains low despite an enlarged LA.

Disclosures

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ESVC-O-14

Iatrogenic heart murmur: a new cause of systolic murmurs in cats

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Heart murmurs are commonly detected in apparently healthy cats and Doppler echocardiographic evaluation is ultimately required to identify the cause of blood flow turbulence responsible for this clinical finding. However, even Doppler echocardiography can occasionally fail to demonstrate the origin of murmurs in cats. Nevertheless, over the years, we have observed that applying gentle pressure to the right side of the chest wall of a cat with the ultrasound probe ("provocative testing") can induce temporary narrowing of the right ventricular infundibulum and dynamic right ventricular outflow obstruction, subsequently causing blood flow turbulence. We have also observed that a similar phenomenon can be reproduced by gently pressing the stethoscope head against the right wall of the chest, inducing an audible murmur during auscultation. The aim of this study was to evaluate the effect of increased pressure of the ultrasound probe against the chest wall of cats undergoing echocardiographic examination in increasing right ventricular outflow velocity and evoking blood flow turbulence in this anatomical area.

Clinical records of apparently healthy cats with dynamic right-sided systolic heart murmurs that underwent echocardiography between 2010 and 2018 were retrospectively reviewed. Only cats that had blood flow turbulence in the infundibular tract induced by provocative testing during image acquisition of the right parasternal short axis view at the level of the heart base and did not have functional or structural abnormalities during echocardiographic examination were included in this study (n = 61). Their median age was 8.0 (6.0 to 9.3) years and mean body weight was 4.5 $\pm$ 1.22 Kg. The median murmur grade was 2/6.

All cats included in the study presented a laminar blood flow on colour Doppler assessment of the right infundibular tract; however, turbulence could subsequently be visualised following provocative testing. Similarly, the provocative test caused increased peak systolic velocity and a late-peaking "scimitar-like" profile, characteristic of dynamic mid-systolic obstruction.

Outflow peak systolic velocities were normally distributed both pre-testing (1.05 ± 0.26 m/s) and post-testing (1.94 ± 0.51 m/s) and their difference (0.89 ± 0.40 m/s) was statistically significant on paired samples *t* test ($P < 0.0001$).

The result of this study confirms that some murmurs in cats can be of iatrogenic origin, being caused by pressure of the ultrasound probe against the chest wall. We postulate that a similar phenomenon can be evoked by pressing the stethoscope head against the chest wall.

Disclosures

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ESVC-O-15

Point Of Care Ultrasound of the Caudal Vena Cava in Canine Degenerative Mitral Valve Disease

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In human medicine, caudal vena cava (CVC) diameter (CVC_D) and collapsibility index (CVC_{CI}) evaluated via Point Of Care UltraSound (POCUS) are accepted markers of intravascular volume status. In human chronic heart failure, CVC POCUS helps identifying patients requiring hospitalization or at risk of decompensation. Degenerative mitral valve disease (DMVD) is the most common acquired canine cardiac disease. It is typically associated with RAAS-activation and a subsequent hypervolemic state in advanced stages. Intravascular volume status impacts treatment and is likely correlated with prognosis. In dogs, CVC parameters obtained via POCUS are described markers of intravascular volume status. The goal of this study was to investigate CVC parameters evaluated via POCUS at different ACVIM stages of degenerative mitral valve disease in dogs.

Echocardiographic and CVC POCUS findings of dogs with DMVD presented between January 2017 and January 2019 were retrospectively reviewed. ACVIM stage and recent administration of diuretics were recorded. Dogs with significant right sided heart disease or pericardial effusion were excluded. POCUS CVC Cineloops were obtained during the echocardiographic evaluation, using a longitudinal subxyphoid view with dogs placed in right lateral recumbency. CVC maximal and minimal diameter were measured and indexed on aortic diameter ($CVC_{D\text{-}max}$/Ao and $CVC_{D\text{-}min}$/Ao), and CVC_{CI} was calculated. One single observer, unaware of disease severity or ACVIM stage, performed all measurements, and subjectively assessed the CVC as fat, flat or normal. CVC parameters were compared between ACVIM stages using Fisher exact test and Kruskal-Wallis. ANCOVA were used to assess the effect of ACVIM stage and diuretic treatment on CVC parameters. Data are expressed as median and range., 81 dogs with DMVD were included (ACVIM stage B1 (23), B2 (24), C (27), D (7)), 28 had recently received diuretics. CVC parameters were associated with ACVIM stage. $CVC_{D\text{-}min}$/Ao was significantly larger, whereas CVC_{CI} was significantly reduced in dogs with ACVIM stage C or D compared with ACVIM stage B1 or B2 (P-value <0.01). $CVC_{D\text{-}max}$/Ao differed in dogs with ACVIM stage D compared with other stages (p-value <0.01). There was a significant association between a subjectively fat CVC and advanced DMVD stages (ACVIM C or D) (p-value <0.0001). ANCOVA revealed that CVC parameters were influenced by ACVIM stages and not by diuretic administration.

CVC POCUS parameters ($CVC_{D\text{-}min}$/Ao and CVC_{CI}) were correlated with disease severity in patients with DMVD, and could be useful to identify dogs in need of hospitalization or at increased risk of decompensation.

Disclosures

No disclosures to report.

ESVC-O-16

Echocardiographically determined left ventricular volume indices obtained from two views in dogs show good agreement

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Echocardiographic left ventricular (LV) volume estimates can help clinicians identify and quantify cardiomegaly and cardiac function in dogs. Cardiologists can obtain these estimates from different views and index them against body size to normalize them for comparisons between individuals and to classify them into ranges of disease severity. However, to-date, studies comparing estimates obtained from different views have been performed by only one group of investigators examining specific breeds. Therefore, we examined the agreement between two methods of obtaining LV volume estimates in dogs with a range of diseases and disease states. We also generated reference intervals for LV volume indices.

Five investigators contributed echocardiographic data from 199 dogs; one dog was excluded from the analyses. The LV of each dog was measured in triplicate using a Simpson's single plane method, from either the right parasternal long axis view or the left apical 4-chamber view, in systole and diastole. The 3 measurements were averaged, and the two methods were compared using limits-of-agreement analyses. Volumes were indexed to bodyweight and to body surface area. Reference intervals were created from measurements obtained on 73 healthy dogs. Indexed LV dimensions were regressed against LA:Ao in 82 dogs with mitral valve disease.

Systolic and diastolic LV volume estimates obtained by the two methods agreed, showing no fixed or proportional biases. Only 2 dogs showed markedly different LV volumes between the two methods. The 95% limits of agreement were approximately 1.0 ml/kg for diastolic LV volume index and 0.5 ml/kg for systolic LV volume index. Healthy dogs had an upper limit of 4.8 ml/kg and 2.2 ml/kg (left apical view) or 4.3 ml/kg and 2.0 ml/kg (right parasternal view) in diastole and systole, respectively. The 90% confidence intervals of each of these limits included the point estimate of the complementary method. Diastolic, but not systolic, LV volume index increased with increasing LA:Ao in dogs with mitral valve disease ($r = 0.28$). Data provided by all investigators appeared similar with no investigator showing obvious bias.

Our study suggests that little difference exists in estimates of LV volume from the two echocardiographic views, and that the estimates

are interchangeable in dogs with a range of cardiac diseases and disease severities. Additionally, our data provide reference limits for volumes indexed to weight, which is both mathematically and physiologically more appropriate than indexing to surface area.

Disclosures

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ESVC-O-17

Supraventricular tachycardia in 23 cats; comparison with 21 cats with atrial fibrillation (2004-2014)

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The causal mechanisms, predisposing factors and natural course of supraventricular tachycardia (SVT) and atrial fibrillation (AF) are well described in people and to a lesser extent in dogs. SVT has not been well-described in cats, and reports of AF are limited. The aim of this study was to describe the signalment, clinical findings and outcome for cats with SVT versus cats with AF.

Forty-four client-owned cats were included in this retrospective study; 23 cats with SVT and 21 with AF. Cats were examined between November 2004 and April 2014. Inclusion criteria were availability of a 50 mm/s 6-lead ECG and a concurrent echocardiographic study. Continuous variables were compared between groups using a two-sample t-test or Mann-Whitney *U* test and categorical variables were summarised using Chi-squared or Fisher's exact test. Kaplan-Meier survival curves were generated to assess for impact of rhythm diagnosis, presence of ventricular arrhythmia, left atrial diameter, heart rate and congestive heart failure status on cardiac death. Differences in survival between groups were compared using Mantel-Cox logrank comparison of Kaplan-Meier survival curves.

Overall, the most common presentation was respiratory distress, (10 of 44 cats), followed by lethargy (n = 9) and collapse (n = 8). Cats with AF had a slower median heart rate (220 [range 180-260 bpm] compared to cats with SVT (300 [range 150-380] bpm, $P < 0.0001$). All cats with AF had cardiac chamber remodelling whereas 4 cats with SVT had no structural abnormalities. Left atrial diameter was significantly larger in AF cats (23.7(16.2-40.1) mm, compared to 19.1 (12.8-31.4) mm in SVT cats; $P = 0.019$)). Median survival was 58 days (1-780) in cats with AF compared with 259 days (2-2295) in cats with SVT (p = 0.112). Cats presenting with signs of CHF had worse overall survival ($P = 0.001$); rhythm diagnosis, ventricular arrhythmia, left atrial size and heart rate had no impact on survival status.

Most cats with AF or SVT have advanced cardiac remodelling, and median left atrial size was greater in cats with AF than SVT. Some cats with SVT had no evidence of cardiac remodelling, suggesting that SVT in cats is not always a consequence of atrial enlargement.

Disclosures

No disclosures to report.

ESVC-O-18

Electrocardiographic patterns of ventricular pre-excitation in the dog

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Ventricular pre-excitation (VPE) describes the activation of a portion of the ventricular myocardium along an accessory pathway (AP), which occurs sooner than if an electrical impulse only conducted along the normal His-Purkinje system. Ventricular pre-excitation has been documented in 1/3 of the dogs with APs. The aims of our study were to explore the electrocardiographic (ECG) features of VPE and identify ECG criteria to determine AP location in this species.

Records of 26 privately-owned dogs with documented AP were retrospectively reviewed. For all dogs 12-lead ECG and detailed electrophysiologic mapping were analyzed. The dogs were classified in three groups according to the position of the AP: antero and mid-septal (6/26), right posterior and right lateral (9/26) and right postero-septal (11/26). For each ECG, measurements on 3 different beats were performed for the following parameters: P-delta wave (d) interval and segment duration, d duration, morphology and axis (at 20 and 40 ms), d-Q, d-R, d-R' and d-S duration, morphology and axis. Descriptive statistics were performed and on quantitative variables normal distribution of values was assessed by the Shapiro-Wilk W-test and mean, median, quartiles and standard deviations were calculated. Kruskal-Wallis one-way analysis-of-variance-by-ranks test was used to evaluate difference between position and delta wave measured on each lead.

In 20 dogs VPE was manifest and in 6 intermittent. The most common d-QRS complex morphology in lead II was the multiple peak QRS (rR', rRs) found in 18/26 dogs. Right posterior and right lateral APs had a taller d-R and d-S respectively in lead I ($P = 0.04$) and II ($P = 0.01$) Antero and mid-septal APs had a taller d-R' in lead III ($P = 0.04$) and a taller d-S in lead V_2 ($P = 0.01$).

The results of this study suggest that localization of APs using surface ECG criteria is possible. Additional studies are needed to test the use of the d-R, d-S and dR' amplitude to guide subsequent radiofrequency catheter ablation procedure.

Disclosures

No disclosures to report.

ESVC-O-19

Use of the cutting balloon technique for the treatment of subvalvular pulmonary stenosis

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Subvalvular pulmonic stenosis (SPS) is classified into two subtypes: infundibular (ISPS) and sub-infundibular (SISPS). ISPS can be primary, due to fixed obstruction of the right ventricular (RV) outflow tract, or secondary to right ventricular concentric hypertrophy caused by pulmonic stenosis. Furthermore, SISPS is also defined as double

chambered right ventricle (DCRV), a high-pressure superior chamber and a low-pressure inferior chamber. The aim of the study was to describe the treatment of ISPS and DCRV in dogs and cats with a the cutting balloon (CB) technique.

This retrospective study included 7 patients (5 dogs and 2 cats) diagnosed with ISPS or DCRV. All cases underwent dilation with a 8 mm in diameter (length 2 cm) CB followed by dilation with an high pressure-balloon. For each patient maximum outflow velocity (Vmax), maximum pressure gradient (PGmax), tricuspidal regurgitation maximum velocity (TRVmax), right atrial (RA) area, RV systolic function estimation were measured before the procedure (T0) and then 24 hr (T1) and 1 month (T2) after the procedure and right atrial and ventricular pressures during the procedure.

The mean diameter of the stenosis was 6,4 (±1,54) mm for the dogs and 2,1 (±0,14) mm for the cats. At T0 mean Vmax was 5.3 ± 0.9 m/s, mean PGmax was 117.07 ± 40.3 mmHg, mean TRV max was 5.23 ± 0,63 m/s, mean RA area was 599 ± 1225.5 mm^2, mean TAPSE was 11.54 ± 4.39 mm, mean TDI was 0.11 ± 0.007 m/s and mean FAC was 42.16 ± 16.1%. Mean pressure values measured by catheterisation before the procedure were: RA pressure 7.3 ± 1.8 mmHg, RV superior chamber 92.66 ± 40.45 mmHg; RV inferior chamber 34.33 ± 12.09 mmHg. CB dilation could not be completed in 1/5 dogs and 2/2 cats. During the dilation mean RV superior chamber pressure was 60.33 ± 13.42 mmHg. Follow-up at T1 was available for 4/4 treated dogs and at T2 was available for 3/4 treated dogs. Following CB dilation mean RV superior chamber pressure was reduced by 31,5%, whilst mean PGmax was reduced by 51.06%.

The present study described the successful treatment of ISPS and DCRV in 4 dogs by CB dilation with a PGmax reduction of 51.06% and reverse right atrioventricular remodelling at T1. The procedure could not be completed in 1 dog and 2 cats probably because of their small body size and/or their severe RA remodelling which impeded us to advace the stiff CB on the softer guide. These preliminary results indicate a possible application of CB dilation in the treatment of ISIP and DCRV.

Disclosures

No disclosures to report.

ESVC-O-20

Imaging and clinical features of canine right atrial appendage aneurysm: a single-centre cross-sectional study in 10886 dogs

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Aim of this study was to describe the prevalence, morphology, and size of right appendage aneurysm (RAA) in dogs. In this 13-year single-centre cross-sectional study, CT reports of dogs underwent thoracic CT examination for various reasons were reviewed. Patients' characteristics were assessed for likely associations with RAA. Radiographic and echocardiographic studies were also evaluated when available. Continuous data were assessed for normality of distribution with the Shapiro-Wilk test. Dogs with RAA were compared with dogs without RAA for the variable gender and sexual status using Chi-Square test. Wilcoxon-Mann-Whitney test was used to analyse age and body weight of groups. The body condition score (five points scoring system) was analyzed using the Fischer's exact test. Non-normally distributed data were reported as median, interquartile range (IQR), and range. For all statistical analyses, the significance level was set to $\alpha = 0.05$. CT data of dogs with RAA were retrieved from archive and analyzed using various post-processing techniques. RAA location and shape were recorded. Oblique multiplanar views were used such that the area to be measured was orthogonal to the long axis of the RAA. RAA measurements included the neck, maximum height and width. Additional CT features of cardiac and extracardiac structures were also recorded. RAA was detected in 23/10886 dogs having thoracic CT in the selected period of time (0.21% prevalence). Radiographic and echocardiographic studies were available for 10/23 dogs. A mediastinal mass were visible in 2/10 radiographs. Echocardiographic examination disclosed RAA in 4/10 dogs with pulmonary hypertension. The bodyweights of dogs with RAA were significantly lower than those of the remaining 10 836 dogs without RAA (median 8 kg [IQR 5; range 37.6] and median 16 kg [IQR 23; range 116.9] respectively; $P = 0.003$). Female sex was also significantly associated with RAA ($P = 0.03$). Four/23 dogs had also right atrial dilatation. Three different RAA phenotypes were detected: sack-like in 19/23 dogs, focal lump-like in 3/23 dogs, and fusiform in 1/23 dog. Results of CT measurements were: mean of the neck 1.48 cm [IQR 1; range 2.6]; mean height 3.06 cm [IQR 1.39; range 4.85], and mean width 1.64 cm [IQR 1; range 3.1]. RAA is an infrequent (0.21%) and often incidental condition that can have different phenotypes. In this study, female sex and smaller size showed an association with RAA. Further case-control studies are necessary to assess a possible relationship between RAA and pulmonary hypertension.

Disclosures

No disclosures to report.

ESVC-O-21

Predictors of reoccurrence of congestive signs in dogs with ACVIM-Stage C myxomatous mitral valve disease (MMVD)

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The aim of this study was to identify predictors of reoccurrence of congestive signs (CS) in dogs with MMVD and clinically stable heart failure (HF). Congestive signs were defined as tachypnea, dyspnea and cough that resolved with medical treatment for HF.

Medical records of 5122 dogs enrolled in the LOOK-Mitral registry from 1st November 2015 to 31st July 2018 were reviewed to identify dogs with stable ACVIM-Stage C MMVD defined as dogs with CS and unchanged medical treatment for at least four weeks since the first identification of HF. The study population was composed by 186 dogs,

subsequently divided into two groups: reoccurrence group (RG, n = 66) and no reoccurrence group (NRG, n = 120). Reoccurrence of CS was defined by resting respiratory rate > 40 breath/minute and/or dyspnea, within the study period, that resolved after furosemide increase.

Baseline body weight (BW) (OR:1.11, 90%CI:1.06-1.17), presence of cough (OR:1.98, 90%CI:1.05-3.74), left atrial-aortic ratio (LA/Ao)(OR: 3.08, 90%CI:1.38-6.86), left ventricular internal diameter at end-diastole (OR: 3.41, 90%CI:1.22-9.50) and end-systole (OR: 7.61, 90% CI:2.32-24.90) indexed to body size, mitral valve peak E wave velocity (OR: 3.44, 90%CI:1.63-7.27), and furosemide daily dosage (OR:1.32, 90%CI:1.04-1.67) were associated with reoccurrence of CS in the univariate analysis. The BW (P = 0.0003) and LA/Ao (P = 0.0196) remained significant in the multivariate analysis. Increment of 0.1 of LA/Ao or of 1 kg in body weight were both associated with 1.1 increased odds of reoccurrence.

This study suggests that BW and LA/Ao are independent predictors of reoccurrence of CS in dogs with stable ACVIM-Stage C MMVD.

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ESVCN-O-1

Dietary lemon balm and fish peptides enhance the efficacy of L-tryptophan to reduce urinary cortisol, a stress marker in cats

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The objective of this study was to test the efficacy of two diets supplemented either with L-tryptophan (diet U) or with L-tryptophan, plus lemon balm (*Melissa officinalis L.*) and fish peptides (diet US), on the reduction of urinary cortisol, a stress marker in cats.

Ten colony cats firstly received a control adult cat diet (diet C) for five weeks and then were randomly assigned to two groups to test diets U and US for five weeks, in a cross over design, with two weeks washout period. Twenty-four-hour naturally voided urine was obtained at the end of each period, under routine conditions and following the application of mild-stressors (open-field test, overnight fast and blood sampling). Urinary cortisol and serum serotonin concentrations were measured and behaviour tests were performed (open-field test and reaction to the presence of and contact with an unfamiliar person). Mixed models for repeated measurements (SPSS) were used to analyse the data.

Compared with control diet, both supplemented diets were effective to reduce urinary cortisol in almost all the tested situations (P < 0.05). Compared with diet U, diet US resulted in a lower urinary cortisol concentration on average and after an overnight fast (P < 0.05), difference for routine being marginal (P = 0.096). Diet US resulted also in a marginal increase in serum serotonin (0.05 < P < 0.10) compared to the two other diets (C and U). Diet U resulted in a higher average score for unfamiliar person test while diet US mainly increased the tolerance to the presence of the unfamiliar person compared to C.

In conclusion, both supplemented diets are effective to reduce stress markers in cats, with an enhanced effect for lemon balm, fish peptides and L-tryptophan supplementation compared to L-tryptophan alone. Supplemented diets may also help to improve interaction with unfamiliar person. New trials should be conducted in cats suffering from stress-related disorders to confirm the clinical benefits of these dietary supplementations.

Disclosures

Disclosures to report.

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ESVCN-O-2

Metabolic effects of a diet with Enterococcus faecium NCIMB 10415 for healthy adult dogs

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The use of probiotics is believed to have health promoting effects such as stabilising the gut microbiome, increase short-chain fatty acid (SCFA) production and lower circulating cholesterol levels. However, probiotic supplementation may also affect the vitamin B metabolism and there are indications that circulating cobalamin may be depleted to levels below reference levels in dogs. Yet, probiotic effects are highly strain specific. The aim of this study was to investigate the effects of a commercial complete diet containing *Enterococcus faecium* NCIMB 10415 on faecal quality, faecal SCFA (acetate, butyrate and propionate) concentrations as well as serum folic acid, cobalamin, cholesterol and triglycerides.

Ten healthy client owned dogs were included in this randomized prospective double-blinded crossover study. All dogs went through a 7 days acclimatisation period of gradual transit from their regular diet to the control diet. The acclimatisation period was followed by two study periods of each 35 days. In each study period, the dogs were randomly assigned to start being fed the control diet (CD) or probiotic diet (PD) (control diet supplemented with *E. faecium* NCIMB 10415, 10^9 cfu/kg). Blood samples and rectal faecal samples were collected at inclusion (I) at day 0 and at the end of each feeding period (day 42 and 77). The faecal quality was scored daily by the owner. Results are presented as mean ± SD, and a difference of P < 0.05 was considered significant.

A significant reduction in serum cholesterol (I: 6.5 ± 2.22 vs PD: 5.40 ± 1.71 mmol/L) and increased faecal content of butyrate was found in the PD relative to I. For both PD and CD, serum cobalamin was significantly reduced (I: 442 ± 110 vs CD: 381 ± 73 and PD:

359 ± 77 ng/L) but within the reference interval (235-812 ng/L) and faecal concentration of acetate was significantly increased compared to I. No changes were found in serum concentration of triglycerides and folic acid or in the faecal concentration of propionate or faecal quality.

Based on this study, *E. faecium* incorporated in a commercial diet, has a potentially health promoting effect in dogs by reducing the serum concentration of cholesterol and increasing the faecal concentration of butyrate. Circulating cobalamin decreased following intervention irrespective of whether *E. faecium* NCIMB 10415 was included or not, which could indicate that other dietary factors may significantly impact cobalamin levels.

Disclosures

Disclosures to report.

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ESVE-O-1

The Relationship of SDMA and Creatinine in Cats with Subnormal Total T4 After Hyperthyroidism Treatment

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Iatrogenic hypothyroidism in cats is associated with reduced glomerular filtration rate, increased occurrence of azotemia, and shortened long term survival. Previous studies have supported this conclusion but were limited by small sample sizes. Symmetric dimethylarginine (SDMA) has been shown to be an earlier, more sensitive, and reliable renal biomarker of decreased glomerular filtration rate than creatinine, which has been found to be influenced by fluctuations in lean muscle mass. The purpose of this study was to utilize big data to characterize the relationship of SDMA and creatinine (Cr) to subnormal total T4 (TT4) post hyperthyroid treatment. Using the US IDEXX Reference Laboratories database 2, 395 cats were identified with samples that were tested with TT4, SDMA and Cr. From this dataset, a hyperthyroid treated group was identified, from this treated population a total of 479 subnormal TT4 cats were identified, defined as having a TT4 < 0.8 μg/dL post hyperthyroid treatment. The remaining cats were euthyroid after treatment. A four-to-one comparative age-matched control group of post treatment euthyroid cats to subnormal TT4 cats was used to evaluate SDMA and Cr pre-and post-treatment. A McNemar's paired test or Chi-square test was used where appropriate. A significant increase in the number of subnormal TT4 cats with an SDMA concentration above the reference interval was found compared to euthyroid controls post-treatment (39.6% (190/479) vs. 32.1% (616/1916), $P < 0.001$). Cr concentrations were also increased above the reference interval in a significantly greater number of subnormal TT4 cats as compared to controls post-treatment (25.9% (124/479) vs. 12.8% (246/1916), $P < 0.001$). In the subnormal TT4 group increased SDMA concentrations identified 13.7% more cats with potential decrease in GFR and reduction in renal function than Cr. This study confirms that a significant percentage of cats with subnormal TT4 attributed to overtreatment of hyperthyroidism have abnormal renal biomarkers as compared to those that are euthyroid following therapy. The big data used for this study supports the importance of avoiding iatrogenic hypothyroidism. The identified association of reduced renal function with iatrogenic hypothyroidism emphasizes the importance of avoiding overtreatment. Comprehensive renal monitoring including measurement of SDMA should be part of routine management of hyperthyroid cats.

Disclosures

Disclosures to report.

All authors listed are employed full-time for IDEXX laboratories.

ESVE-O-2

Prevalence of 'Atypical' Addison's disease among a population of dogs diagnosed with hypoadrenocorticism

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A diagnosis of 'Atypical' Addison's disease is typically made in patients diagnosed with hypoadrenocorticism with a Na:K ratio > 27 at presentation. The percentage of patients with hypoadrenocorticism which have 'atypical' disease remains unknown. The primary aim of this study was to determine the percentage of dogs diagnosed with atypical disease within a general population of dogs diagnosed with hypoadrenocorticism and to determine if this percentage differed between a population diagnosed in first opinion practice and a population diagnosed at referral practice.

The database of a commercial laboratory in the United Kingdom was searched to find dogs diagnosed with hypoadrenocorticism over a four year period (2015-2018). Dogs were included if an ACTH stimulation test was performed and post ACTH cortisol concentration was <55 mmol/L. The results of serum or plasma sodium and potassium concentrations at the time of initial presentation were also required for inclusion. Dogs were excluded if they were receiving trilostane or any other medication known or expected to interfere with adrenal function testing. Dogs were diagnosed with 'atypical' hypoadrenocorticism if the Na:K ratio at the time of presentation was >27.

Forty-seven dogs with newly diagnosed hypoadrenocorticism were identified and included. Of the 47 cases included, 20 cases (43%) were diagnosed with 'atypical' hypoadrenocorticism. Of the 33 cases diagnosed at referral practice, 16 (48%) were diagnosed with 'atypical' hypoadrenocorticism. Of the 14 cases diagnosed at first opinion practice, 4 (29%) were diagnosed with 'atypical' hypoadrenocorticism.

The overall percentage of cases diagnosed with 'atypical' hypoadrenocorticism may be higher than previous estimates. The percentage of newly diagnosed cases of hypoadrenocorticism at referral practice which have 'atypical' disease appears higher than the percentage of 'atypical' cases diagnosed at first opinion practice. In the absence of electrolyte abnormalities classically associated with 'typical' cases of hypoadrenocorticism, particularly a Na:K ratio < 27, the index of suspicion for the disease may be low. The finding of a higher percentage of patients with 'atypical' disease when considering the population

diagnosed at referral practice raises concern that cases may go undiagnosed at first opinion practice.

Disclosures

Disclosures to report.

One of the authors was employed by the commercial laboratory which allowed its databased to be searched in order to identify cases. This commercial laboratory performed the measurement of serum cortisol concentrations in all included cases.

ESVE-O-3

The predictive role of the transtubular potassium gradient (TTKG) for Addison syndrome in hyperkaliemic dogs: a cross-sectional study

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Addison disease is characterized by a deficiency of both cortisol and aldosterone. The lack of aldosterone results in renal sodium wasting and potassium retention leading to hyponatriemia, hyperkalemia and a body volume depletion. The effect of aldosterone on serum potassium excretion can be evaluated by comparing urine and serum potassium concentrations after correcting the urine potassium concentration for reabsorption of solute-free water by the kidney. This estimation has been called TTKG. Thus, the aim of this study was to evaluate the ability of TTKG in the identification of dogs affected by Addison disease among a population of hyperkalemic dogs.

For this cross-sectional study, we retrospectively search the data base for dogs with a serum potassium concentration > 5.5 mmol/L (reference interval = 3.9-5.1) presented between December 2012 and February 2019. Inclusion criteria were a urine TTKG calculated at hospital admission (which is routinely done in our laboratory) and a final diagnosis available. Based on final diagnosis dogs were divided in newly diagnosed, naturally-occurring Addison's disease (diagnosed by an ACTH stimulation test) and other diseases. Dogs were excluded from the study if they had an history of corticosteroid administration and/or drugs having affecting potassium excretion (e.g., ace-inhibitors, aldosterone-receptor blockers, diuretics, fludrocortisone, IV fluids). Moreover, hyperkalemic dogs were also excluded if urine osmolality was ≤300 mOsm/Kg or urine sodium was ≤25 mmol/L, precluding these values a correct TTKG calculation. TTKG was compared by T-test between dogs with Addison disease and sick hyperkalemic controls dogs. Finally, ROC curve analysis was used to identify the best cutoff value (Youden index) for discriminating dogs with Addison disease from sick hyperkalemic controls dogs without Addison disease. For all analyses the significance was set to $\alpha = 0.05$.

Eighty hyperkalemic dogs were included in this study, 41 with Addison disease and 39 without. TTKG in dogs with Addison disease was significantly ($P < 0.0001$) lower (3.5 ± 1.73) than TTKG in control dogs (5.8 ± 2.07). The Youden index identified through ROC curve analysis for TTKG was 4.55 (sensitivity = 75.6%, specificity = 74.3%; AUC = 0.802, 95% CI, 0.707 to 0.898; $p < 0.0001$).

The present study showed that TTKG, in hyperkalemic patients can be used as a diagnostic tool in the initial discrimination of dogs with Addison disease, from hyperkalemic dogs without Addison disease. In a subset of hyperkalemic dogs, TTKG may be helpful in patient management until confirmatory diagnosis with an ACTH stimulation test is available.

Disclosures

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ESVE-O-4

Comparison of different monitoring methods in dogs with hypercortisolism treated with trilostane

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The monitoring of trilostane treatment, the drug of choice for the medical therapy of canine hypercortisolism (HC), is currently based on the evaluation of the clinical signs and the results of the ACTH stimulation test. However, this method has many limitations and recent data have shown a lack of correlation between ACTH stimulation test results and the clinical signs of dogs with HC treated with trilostane. Over the last years, many different studies investigated possible alternative methods with conflicting results. A single study to compare all these methods in the same canine population is lacking. The aim of this study was to evaluate which of the previous investigated monitoring methods better correlate with a standardized and published clinical score (CS) obtained by an owner questionnaire and could represent the best method to monitor trilostane therapy.

We conducted a prospective multicentre study. Dogs with HC on treatment with trilostane twice daily for at least two weeks were blood sampled and categorized as unwell (sick or over-treated dogs), well and under controlled (no dose or dose increase required dogs, respectively) based on the CS. The results of the CS were compared with: serum cortisol concentration pre-trilostane (T0), 3 h-post-trilostane (T3) and 4 h-post-trilostane (1 h-post-ACTH) (T4) administration, plasma ACTH concentration pre-trilostane administration, plasma ACTH/cortisol (T0) ratio, serum haptoglobin concentration (Hp), chemistry variables (ALP, ALT, GGT), urinary cortisol/creatinine ratio and urine specific gravity (from urine of the evaluation's day and from the morning of the day before). Plasma ACTH and serum and urinary cortisol were measured with a chemilumescent assay (Immulite 2000)., 76 re-evaluations of 37 dogs were included. Unwell dogs were excluded for further analysis. Haptoglobin was the parameter that better correlated with the CS ($r = 0.47$), followed by ALT ($r = 0.34$), T0 ($r = 0.33$) and the UCCR average of the 2 urinary samples($r = 0.33$). ROC curve analysis identified a concentration of Hp of 150 mg/dl and a concentration of T0 of 4 μg/dl as useful value to discriminate well and under controlled dogs with a specificity of 91% and 78% respectively.

Hp seems to be the best parameter to monitor trilostane therapy. However, Hp has likely limited capability to identify over-treated dogs; the concurrent evaluation of cortisol pre-trilostane may be useful in detecting an overdose of trilostane. The combined evaluation of Hp and T0 correctly categorized 85% of the cases and can be used as

alternative monitoring method for dogs with HC under trilostane therapy.

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ESVE-O-5

Feline plasma adrenocorticotropic hormone: validation of a chemiluminescent assay and concentrations in cats with hypercortisolism, primary hypoadrenocorticism and other diseases

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Naturally occurring hypercortisolism (HC) and hypoadrenocorticism are rare conditions in cats and their diagnosis can be challenging. Actually, there is a lack of validation studies for the measurement of feline plasmatic adrenocorticotropic hormone (ACTH). The aims of this study were to validate a commercially available chemiluminescent assay to measure feline ACTH concentrations, determine the normal reference interval (RI) and assess plasma endogenous ACTH concentrations in cats with primary hypoadrenocorticism (PH), HC and cats with other diseases (OD).

Thirty-three healthy cats and 25 cats with OD (9 cats with diabetes mellitus; 5 with hyperthyroidism; 3 each with chronic kidney disease, or gastrointestinal disease; 2 with acute kidney injury; and 1 each with hypovolemic shock, septic shock, or hyperaldosteronism) were included prospectively in the study. Data from 11 cats with PH and 9 with HC (8 pituitary-dependent hypercortisolism - PDH - and 1 adrenal-dependent hypercortisolism - ADH) were retrieved from medical records of three referral centers (University of Bologna, Zurich and Utrecht) that use the same method of measurement (Immulite 2000). Left over samples, collected for diagnostic purposes, were used. The intra-assay coefficients of variance (CVs) ranged from 2.6 to 3.6%, and interassay CVs from 6.6 to 13.2%, for samples with high and low concentrations of ACTH, respectively. Dilution studies performed on two samples with high concentrations of ACTH, using the diluent provided by the manufacturer, showed excellent accuracy (R^2 > 0.99). The RI for plasma endogenous ACTH in healthy cats, established using the Robust Method, was 27-390 pg/mL (median 96 pg/mL). Plasma ACTH concentrations ranged from 23.6 to 400 pg/mL, 21.6 to 355.5 pg/mL, 331 to >1250 pg/mL in healthy, OD and PDH cats, respectively; the only cat with ADH showed an ACTH value of 5 pg/mL (detection limit of the assay). In all the cats with PH the concentration of ACTH was >1250 pg/mL. ACTH concentrations did not show significant differences between healthy and OD groups. Cats with PDH and PH had significantly higher ACTH values than the other groups. There was only 1 PDH cat with a result in the range of healthy and OD cats. Immulite chemiluminescent assay is a valid technique for measurement ACTH in feline plasma and the RI is quite wide. Due to the low overlap between healthy or OD cats and those with PH or HC, measurement of endogenous ACTH seems useful and should be included in the diagnostic workup when PH or HC are suspected.

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ESVE-O-6

Major Histocompatibility Complex (MHC) class II haplotypes associated with increased risk of canine diabetes mellitus - a breed-specific study

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Canine diabetes mellitus (DM) can be classified as insulin resistance or insulin deficiency diabetes, with all cases requiring daily insulin injections to control hyperglycaemia. A number of pathological mechanisms are thought to lead to the development of the disease, including immune-mediated destruction of beta cells. Disease risk differs considerably between breeds, suggesting that genetic factors are involved, but environmental triggers are also thought to play a role. In human Type 1 DM, the region of the genome containing the human leucocyte antigen (HLA) MHC class II genes confers approximately 50% of the genetic risk. Associations with dog leucocyte antigen (DLA) class II haplotypes have also been identified in diabetic dogs, but investigations to date have considered all breeds together. This study aimed to identify breed-specific diabetes-associated DLA haplotypes in an expanded data set. Taking the 18 most highly represented breeds in our previous study, we increased the DLA-typing data from 294 to 736 diabetic dogs and from 501 to 1083 breed-matched non-diabetic controls to enable breed-specific statistical analysis.

Dogs were genotyped for DLA-DRB1, –DQA1 and -DQB1 using sequence-based typing of DNA (extracted from blood surplus to diagnostic requirements). The study population only included dogs from the UK. Female entire cases, expected to have dioestrus diabetes, were excluded. In all breeds, there were at least ten cases and the number of controls was equal to or greater than the number of cases. Genotypes from all three loci were combined to identify DLA class II haplotypes affecting risk of DM across each breed individually and all

breeds together ($P < 0.05$ using Fisher's Exact test and an Odds Ratio [OR] confidence interval entirely <1.0 or > 1.0).

Five breeds were identified as having one or more DLA haplotypes that were significantly different between DM cases and controls. These breeds were Bichon Frise (one risk haplotype, OR = 4.41); Labrador Retriever (one risk haplotype, OR = 1.60); Cavalier King Charles Spaniel (one risk haplotype, OR = 2.73); Cocker Spaniel (one risk haplotype, OR = 6.31 and one protective haplotype, OR = 0.30); Border Terrier (one protective haplotype, OR = 0.21). Combined analysis of all samples identified four DM-associated haplotypes: one risk and three protective, including one protective DLA-DQ haplotype identified in the previous study.

These new DLA associations provide further evidence of a role for the adaptive immune system in canine DM, and highlight potentially different aetiologies between breeds.

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ESVE-O-7

Comparison of nine canine serum thyroxine measurement methods and impact of T4 cross-reacting autoantibodies

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Serum thyroxine (T4) concentration is commonly measured to assess canine thyroid function or monitor therapy. Several reference laboratory and in-clinic immunoassays are commonly used. The pathogenesis of hypothyroidism may result in endogenous T4 cross-reacting antibodies (T4AA) in the sera of a proportion of cases. T4AA may interfere with T4 analysis.

The aims of this study were to compare results between seven total- and two free-thyroxine methods and to investigate the in-vitro effect of T4AA on them.

Five serum pools of predictably equidistant T4 concentration covering the reference interval were created using sequential 50:50 mixing of 2 initial pools of low and high T4 concentration surplus canine serum and analysed by 7 TT4 (Radioimmunoassay (RIA); Immulite Total T4 (ImmTT4); Immulite Canine Total T4 (ImmKT4); Thermo-Microgenics Total T4 (DRI-T4); IDEXX Catalyst T4 (CataT4); IDEXX Snapshot T4 (SnapT4) and Tosoh AIA T4 (TosT4)) and two Free T4 methods (Antech Free T4 by dialysis (FT4d) and Immulite Veterinary Free T4 (ImmVF4)). The mixed pools approach allowed for assessment of internal agreement within each method (linearity) as well as comparison between methods. To determine the impact of T4AA, the same 5 pools were additionally analysed after mixing 50:50 with a T4AA positive canine surplus serum pool.

Total T4 methods did not agree with one another; e.g., highest pool varied between 39 and 59.5 nmol/L. Two TT4 methods (RIA and ImmKT4) demonstrated good linearity with all points agreeing with predicted concentrations. Of 5 methods that demonstrated less good internal agreement, three were linear but with results that did not match predictions across the range (DRI-T4, TosT4, SnapT4) and two were non-linear (ImmTT4, CataT4). ImmVF4 and FT4d were linear, although one data point by FT4d was not as predicted.

An in-vitro effect of T4AA was seen in all methods (including FT4d) causing false low results by ImmKT4, DRI-T4, CataT4, SnapT4, TosT4 and FT4d and false high by RIA and ImmVF4. ImmTT4 generated mid-range (21-24 nmol/l) results across all concentrations in the presence of T4AA.

The choice of analytical method for canine serum T4 is likely to have an impact patient management decisions. Commonly available methods do not agree with one another and several do not agree within themselves. The effect of T4AA varies by method and a combination of false high and false low methods within thyroid test panels could help detect the likely presence of interfering T4AA if they are not measured directly.

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ESVE-O-8

Analysis of GWAS data in Domestic Shorthair and Burmese cats identifies diabetes-associated loci near the DPP9 and within the DPP10 gene

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Diabetes mellitus (DM) in cats resembles human type 2 DM, a complex disease involving a combination of genetic and environmental factors. In a previous genome-wide association study (GWAS), 4 single nucleotide polymorphisms (SNPs) were found to be associated with DM in lean Domestic Shorthair (DSH) cats, and a polymorphism in the MC4R gene was associated with DM in overweight DSH cats in a candidate gene study. Susceptibility genes in Burmese cats, a breed predisposed to DM, had not been identified in the initial GWAS study.

In an attempt to overcome some of the limitations present in the Burmese breed due to inbreeding, data from the previous GWAS of DM in DSH and Burmese cats was combined for analysis. Genotyping was performed using Illumina Infinium 63 k iSelect DNA array, and after quality filtering, 390 diabetic and 390 non-diabetic control DSH cats, as well as 19 diabetic and 21 control Burmese cats were included in the analysis. Controls were significantly older than diabetic cats ($P < 0.0001$). Body condition score (BCS) was known for 718 cats, 62% of these were lean and 38% were obese. Stratified analysis using a Cochran-Mantel-Haenszel (CMH) test within strata defined in the multidimensional scaling (MDS), and logistic regression with MDS coordinates, using BCS and breed as covariates, were conducted for

case:control association testing. P-values from the CMH test were adjusted for genomic inflation (λ = 1.187), and feline standard genome-wide significance was set at P < 10e-5. Max(T) permutations were used to generate corrected empirical P-values following logistic regression; significance was set at P < 0.05. A single significant SNP (chrC1:125033967; P_{raw} [adjusted for genomic inflation] = 9.58x10e-6) was identified in the stratified analysis, and a single significant SNP (chrA2:3535683; P_{genome} [100000 permutations] = 2.75x10e-3) was identified in logistic regression. The SNP in chromosome A2 was associated with DM in the previously reported GWAS of DM in lean DSH, but the association with the SNP in chromosome C1 in both breeds is new. These SNPs are located within and close to DPP10 and DPP9 genes, respectively, which are related to DPP4, an enzyme involved in degradation of incretins. This further analysis has revealed new potential candidate genes in both breeds, highlighting the usefulness of this alternative approach to the GWAS analysis. Both DPP9 and DPP10 have been implicated in metabolic and immune functions, suggesting that further investigation of these genes in the pathogenesis of feline DM is warranted.

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ESVE-O-9

Glycemic variability in newly diagnosed diabetic cats treated with the GLP-1 analogue exenatide extended-release

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Glycemic variability (GV) refers to glycemic excursions with episodes of hypoglycemia and hyperglycemia throughout the day or on different days with no apparent causal link and is considered to be an indicator of glycemic control.

In humans with diabetes mellitus (DM), adding a glucagon-like peptide-1 (GLP-1) analogue to the conventional therapy results in significant reduction in GV. In cats knowledge on GV is scarce and the influence of different treatment modalities has not been studied. The objective of this study was to evaluate GV in cats receiving the GLP-1 analogue exenatide extended-release (EER) additionally to insulin therapy. Blood glucose curves from a recent prospective placebo-controlled clinical trial were evaluated for GV 1, 3, 6, 10 and 16 weeks after starting therapy. Cats were treated with EER (200 μg/kg) or 0.9% saline, administered subcutaneously, once weekly. Both groups received insulin glargine twice daily and a low-carbohydrate diet.

To assess GV, mean glucose concentrations and standard deviations (SD) were calculated and compared between treatment groups and, in the EER group, between cats achieving or not achieving remission. Both dependent variables (mean and SD) were analyzed using a repeated mixed linear model which included the fixed effects of treatment, week, their interaction and repeated animal effect. Data were reported as least-squares means and SE. Thirty cats with newly diagnosed DM were included, 15 of which received EER and 15 received placebo. Six of 15 (40%) cats and 3 of 15 (20%) achieved remission in the EER and placebo group, respectively (P = 0.427). The mean of the whole study period was lower in the EER group compared to placebo (9.6 $\pm$ 0.8 vs. 12.4 $\pm$ 0.8 mmol/L; P = 0.024); by considering the single time points, the mean was lower at week 6 (5.0 $\pm$ 1.7 vs. 12.9 $\pm$ 1.6 mmol/L; P = 0.003). The GV-SD of the whole study period was lower in the EER group compared to placebo (2.4 $\pm$ 0.3 vs. 3.3 $\pm$ 0.3 mmol/L; P = 0.035).

In the EER group, cats achieving remission had lower mean (7.6 $\pm$ 1.1 vs. 10.9 $\pm$ 0.8 mmol/L; P = 0.026) and lower GV-SD (1.7 $\pm$ 0.4 vs. 2.9 $\pm$ 0.3 mmol/L; P = 0.032) than those not achieving remission.

In conclusion, the GLP-1 analogue EER leads to a reduction in GV and better glycemic control in cats with DM. Furthermore, lower GV is associated with higher remission rates in cats treated with EER. The results of this study therefore suggest that adding a GLP-1 analogue to the conventional therapy may be advantageous in the treatment of cats with DM.

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ESVE-O-10

Clinical performances of flash glucose monitoring system in diabetic dogs

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Flash glucose monitoring system (FGMS, FreeStyle Libre®) was recently validated for use in diabetic dogs (DD). It continuously measures the interstitial glucose concentrations for up to 14 days. The aim of this study was to evaluate the clinical usefulness of FGMS in monitoring DD.

Twenty DD on insulin treatment were prospectively enrolled in the study. The FGMS was placed on the neck for up to 14 days. Duringthe 1st-7th-14thdays, blood glucose curves (BGCs) have been performed simultaneously in the hospital with FGMS and a validated portable blood glucose meter (PBGM) (OptiumXceed, Abbott®). During the 5th-6th and 12th-13thdays the owners performed a BGC using the FGMS at home. The BGCs performed with PBGM and FGMS in

hospital and those performed with FGMS at home and in hospital were compared. Each BGCs has been evaluated as optimal considering: 1) 50% of the values between 90-250 mg/dL or 2) glucose nadir between 90-180 mg/dL. The glucose nadirs obtained from the data downloaded by the software (DDS), the FGMS scans and the PBGM were compared. Moreover, the glucose day-time (GDTNs) and night-time nadirs (GNTNs) were compared.

The evaluation of the BGCs performed in hospital with FGMS and PBGM, led to the same decision on insulin adjustment in 77% and 80% of cases considering the percentage of values in the range 90-250 mg/dL and the glucose nadir, respectively.

The evaluation of the BGCs performed at home and the following day in the hospital with the FGMS, led to the same decision of insulin adjustment in 68% and 64% of cases considering the percentage of values in the range 90-250 mg/dL and the glucose nadir, respectively.

The glucose nadirs were identified in 81% of cases by the DDS and in 65% and 35% of cases using FGMS scans and PBGM, respectively.

The medians of GNTNs were significantly higher than the GDTNs.

The hypoglycemic episodes obtained from the DDS were 39% more than those immediately showed on the display of the FGMS.

In conclusion, adjustments in insulin dose based on BGCs obtained with FGMS and with PBGM are similar. The FGMS detects the nadirs and the hypoglycemic episodes more frequently than PBGM and it allows the assessment of glucose variations also during different consecutive days. Therefore, FGMS is a potentially valuable tool in the monitoring of canine diabetes mellitus.

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ESVE-O-11

Whole Genome Sequencing to explore genetic risk factors in canine diabetes mellitus

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Canine diabetes mellitus (DM) is more prevalent in certain breeds, suggesting an underlying genetic basis, although environmental factors may also be involved. Notably, dog breeds with low DM risk are over-represented in studies of neoplastic transformation of pancreatic beta-cells (insulinoma). This suggests that beta-cell survival may be an important contributing factor in canine DM, and that similar genes may be involved in canine DM and insulinoma. Previous genetics work in canine diabetes mellitus (DM) has focused on candidate genes and genome-wide association studies, employing a case-control design within individual breeds. However, this design does not account for the fact that, within high risk breeds, a fixed genetic risk of DM may exist, resulting in only minimal genetic differences between cases and controls.

The aim of this study was to develop a new model for identification of genetic risk variants in complex disease, in order to identify new canine DM genes. This was achieved by exploring the genetic differences between dog breeds at very high risk of DM (Samoyed - Odds Ratio 15.2) and exceptionally low risk of DM (Boxer - Odds Ratio < 0.01), as part of the Canine Diabetes Genetics Partnership initiative.

Whole genome sequencing (WGS) at 30X coverage was undertaken on 6 diabetic Samoyeds and 6 Boxers with insulinoma, using Illumina HiSeqX technology. Six Samoyeds and 6 Boxers without DM or insulinoma underwent WGS as controls. DNA was extracted from blood samples that were surplus to requirements for clinical purposes. A custom bioinformatics pipeline was developed to annotate and prioritise variants for follow-up, based on the Genome Analysis ToolKit. Variants were annotated according to their minor allele frequency (by breed or case-control status), predicted impact on gene function and location near a region with a plausible role in beta-cell function or diabetes risk. In Samoyeds, >4000 breed-unique high or moderate impact variants were identified, >3000 of which were present in more than one Samoyed, and 173 of which were found exclusively in the diabetic group. In Boxers, >1500 breed-unique high or moderate impact variants were identified, >150 of which were present in more than one Boxer, and 68 of which were found exclusively in the insulinoma group. Replication and functional studies are in progress to validate candidates and investigate underlying mechanisms. This study demonstrates that WGS offers a promising route for investigation of complex diseases where genetic risk may be fixed at a high or low level within breeds.

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the CDGP and none stand to make any direct financial gain from the data in this abstract. All authors have seen the information presented in this abstract and have been given the opportunity to comment on it. In the interests of full disclosure, please note that a preliminary findings abstract on Whole Genome Sequencing in canine diabetes (submitted by the Canine Diabetes Genetics Partnership) has been accepted for poster presentation at the Canine and Feline Genetics and Genomics meeting in Bern 2019. However the ECVIM abstract submitted here is based on screening variants against a larger data set, so uses data from additional dogs and breeds. It also includes bioinformatics improvements, allowing better quantification and annotation of candidate genetics variants.

ESVE-O-12

Evaluation of 1,2-o-dilauryl-rac-glycero glutaric acid - (6′-methylresorufin) ester (DGGR) lipase assay in dogs with naturally occurring hypercortisolism

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Canine pancreatic-specific lipase (ScPL) is considered the most sensitive and specific test for the diagnosis of pancreatitis in dogs. However, relatively high costs and long turnaround times can limit everyday clinical use. Other more accessible serum assays are available for daily usage like 1,2diglyceride lipase assay (total lipase; TL) and 1,2-o-dilauryl-rac-glycero glutaric acid-(6′-methylresorufin) ester (DGGR) lipase assay (DGGRL). Recently a study compared ScPL to DGGRL obtaining a high agreement. For this reason DGGRL is actually widely used. A recent study observed that in 35% of dogs with hypercortisolism, ScPL concentrations were elevated (≥400 μg/L) resulting in false positive results for pancreatitis.

The aim of the present study is to evaluate serum DGGRL and TL activity in dogs affected by naturally occurring hypercortisolism. Dogs with acute pancreatitis (AP) and healthy dogs (HD) were used as controls.

Left over samples used for diagnostic proposals and stored at −20°C were selected retrospectively, following stability period recommendations, and analyzed from 19, 21 and 23 dogs with hypercortisolism, AP and HD, respectively. Serum DDGRL and TL were measured using an automated analyzer. Diagnosis of hypercortisolism was performed combining more than 2 typical clinical signs with a positive specific endocrine test (ACTH stimulation test and/or LDDS test). Dogs with hypercortisolism were included only if they had absence of clinical and ultrasonographic signs suggestive of AP. The diagnosis of AP was based on the presence of suggestive clinical, clinicopathological (excluding the study variable) and ultrasonographic findings. Data were expressed as median and (range) and compared using nonparametric statistics ($P < 0.05$ considered significant).

Median DGGRL concentration (U/L) was 234 (55-874), 263 (37-1768) and 44 (19-209) in dogs with hypercortisolism, AP and in HD, respectively. Median TL concentration (U/L) was 416 (144-1932), 469 (84-3472) and 270 (58-544) in dogs with hypercortisolism, AP and in HD, respectively. DGGRL and TL concentrations were significantly different among HD and the other two groups, but resulted not different between dogs with hypercortisolism and AP ($P = 0.58$; $P = 0.74$). DGGRL concentrations were above the reference interval (10-130 U/L) in 63.2%, 66.6% and 8.7% dogs with hypercortisolism, AP and HD, respectively. TL concentrations were above the reference interval (70-700 U/L) in 26.3%, 33.3% and 0% dogs with hypercortisolism, AP and in HD, respectively.

Because of the high rate of false positive results, DGGRL concentrations should be evaluated with caution when AP is suspected in dogs with hypercortisolism.

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ESVE-O-13

Comparison of measurement of free thyroxine concentration by a chemiluminescent analogue immunoassay to equilibrium dialysis in dogs with non-thyroidal illness

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Measurement of free thyroxine (T4) concentration by equilibrium dialysis (fT4d) is technically demanding and expensive. An automated free T4 analogue (fT4a) immunoassay (Immulite 2000 Veterinary Free T4, Siemens) has recently gained popularity. However, there are concerns regarding its ability to differentiate hypothyroidism from non-thyroidal illness (NTI).

The aim of this study was to evaluate the changes in fT4a concentrations in dogs with NTIs for comparison with simultaneous total T4 and fT4d concentrations. It was hypothesized that fT4a would provide good and poor agreement with total T4 and fT4d concentrations, respectively.

Initially fT4a concentrations were measured in surplus serum samples from 150 dogs with various NTIs in which hypothyroidism was not suspected. Total T4 concentration (Immulite Canine Total T4, Siemens) and fT4d (Free T4 by Equilibrium Dialysis, Antech Laboratories) were subsequently measured in a subpopulation of 75 dogs selected with a range of fT4a concentrations. Reference intervals were 7.7 to 47.6 PMol/L, 7.0 to 40.0 PMol/L and 15.0 to 50.0 nmol/L for fT4a, fT4d and total T4, respectively. The Kruskal Wallis or chi-squared tests were used for statistical analysis, as appropriate.

In 150 dogs with NTI, fT4a was significantly ($P < 0.001$ and $P = 0.023$, respectively) lower in dogs with severe compared to mild and moderate NTI. The proportion of dogs with values below the reference interval was significantly ($P = 0.013$ and 0.0032, respectively) less for fT4d (n = 25 [33.3%]) compared to fT4a (n = 40 [53.3%]) and total T4 (43 [57.3%]). The proportions did not differ significantly ($P = 0.62$) between fT4a and total T4. Among the dogs with low total T4 concentration, fT4a and fT4d were normal in 5 (11.6%) and 19 (44.2%)

dogs, which was significantly ($P < 0.001$) different. Among the 40 dogs with low fT4a, 15 (37.5%) dogs had normal fT4d values. Bland-Altman difference plot revealed that fT4a underestimated and overestimated free T4 concentration as measured by fT4d at low and high concentrations, respectively.

Considering the lack of agreement observed, fT4a and fT4d cannot be used interchangeably. As opposed to fT4d, fT4a provided no more diagnostic information than that gained by measurement of total T4 in dogs with NTI. Based on these results, fT4a cannot be recommended to differentiate NTI from hypothyroidism in dogs with a low total T4 concentration. This represents the population in which measurement of free T4 concentration is most commonly recommended.

Disclosures

Disclosures to report.

Carmel Mooney provides consultancy services for Dechra Veterinary Products. Helen Evans is Veterinary Services Operation Manager at NationWide Specialist Laboratories.

ESVE-O-14

Organoid cultures of follicular-cell thyroid carcinoma: a novel canine model for translational thyroid cancer research

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Growing patient-derived tissue in 3-dimensional cell culture systems (organoids) has revolutionized *in vitro* cancer research. In contrast to 2-D cell lines, organoids can be grown more efficiently and conserve important features of the original tumor, such as tissue architecture and cellular heterogeneity. In human medicine, organoid cultures provide a unique platform for personalized cancer therapy. In this study, we aimed to culture and characterize organoids derived from follicular-cell thyroid carcinoma (FTC) in dogs.

Tissue samples of follicular type (n = 1) and compact type (n = 1) FTCs derived from two euthyroid dogs were frozen in DMSO-containing freezing medium within 24 h of thyroidectomy and stored at −150°C until processing. After thawing, the tissue was digested with collagenase IV and dissociated mechanically. Cells were subsequently seeded in Cultrex® Basement Membrane Extract and cultured to organoids in Advanced DMEM supplemented with N-acetylcysteine, B-27 supplement, EGF, Noggin, Rspondin-1 and Rock-inhibitor. Organoids were split and passaged every 9 to 14 days. After 15 to 24 days, organoids were formalin-fixed, pelleted in 2.5% agarose, paraffin-embedded and processed for hematoxylin-eosin staining. Immunohistochemistry (IHC) for thyroid transcription factor-1 (TTF-1), thyroglobulin (Tg), calcitonin, vimentin and Ki-67 was performed on sections of the primary tumors and organoids.

Organoids of FTC were cultured efficiently using our protocol. Organoids of both tumors formed follicle-like structures composed of a single epithelial cell layer. These epithelial cells were round to cuboidal, had variably distinct cell borders and abundant eosinophilic to foamy cytoplasm. Anisocytosis and anisokaryosis were not observed in either of the organoid lines but were present in the FTC of compact type.

Nuclear expression of TTF-1 in both organoid lines confirmed thyroid origin. The organoids derived from the compact FTC, which had approximately 30% of cells positive to Tg, showed no Tg expression while the organoids derived from the follicular FTC, which had Tg expression in >95% of cells, showed Tg expression in about 50% of the organoids. Vimentin expression was observed in both organoid lines (30-60% of cells) and was higher than in the primary tumors, where only up to 10% of tumor cells were positive. IHC for calcitonin and Ki-67 was negative in both organoid lines.

Organoids derived from naturally occurring canine FTC are able to conserve histological and immunohistochemical features of the primary tumors providing an interesting *in vitro* model to better understand the pathogenesis and optimize treatment of thyroid cancer in dogs. The culture protocol likely requires further optimization.

Disclosures

No disclosures to report.

*Both authors contributed equally to this work.

ESVIM-O-1

Pulmonary deposition of nebulized 99mTc-DTPA after pharmacologically induced airway narrowing in healthy dogs

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In humans, airway narrowing due to asthma or COPD commonly results in heterogeneous deposition of aerosols within the lung. Consequently, uneven distribution of inhaled medications may lead to variable and inadequate drug levels in certain lung areas. Currently it is not known if the same effects occur in dogs with lower airway disease.

The purpose of this study was to evaluate the effects of pharmacologically induced airway narrowing on pulmonary deposition of a nebulized radiopharmaceutical (ie. the amount and distribution) in healthy dogs.

The prospective study was conducted in ten healthy beagles. Radiopharmaceutical inhalation was performed on 2 occasions with a wash out period of at least 1 week between both experiments (ie. without and immediately after induction of airway narrowing). Narrowing of the airways was achieved by nebulizing increasing concentrations of carbachol into a barometric whole body plethysmography chamber harboring the animal until airway narrowing was detected by the system (Buxco FinePoint®; endpoint: increase of PENH >300% over baseline).

On both occasions dogs inhaled a dose of nebulized ^{99m}technetium-diethylenetriamine-pentaacetic acid (^{99m}Tc-DTPA) through an Aerochamber® Medium attached to a customized nose-muzzle mask for 3 minutes. Immediately afterwards dogs were scanned with planar scintigraphy in right lateral, left lateral and sternal recumbency. The deposition of ^{99m}Tc-DTPA in the head region, the lungs, the stomach, as well as the whole body distribution were recorded and quantified

by manual or isocontour region of interest (ROI) analysis. Deposition calculated as percentage of delivered dose in the ROI was compared. The distribution of deposition within the lungs was scored independently by 3 blinded observers for the degree of asymmetry between right and left lung (0-2) and patchiness (0-2) of individual images as well as possible differences in the scan image pairs (ie. before and after airway narrowing; 0-2) in random order. For analysis, individual scores were averaged.

Mean percentage uptake of the delivered dose in the lungs was not significantly different before and after airway narrowing. Before airway narrowing, the average asymmetry score was 0.8 whereas none of the dogs had a patchy distribution (score 0). After airway narrowing, the scores for asymmetry and patchiness significantly increased (1.7 and 1.1, respectively). Comparison of the scan image pairs before and after airway narrowing revealed a significant difference (score 1.8).

This study indicates that airway narrowing in dogs results in heterogeneous aerosol deposition within the lungs, although the overall amount of drug deposition is not affected.

Disclosures

Disclosures to report.

Lecturer for Improve (Hirt).

ESVIM-O-2

Assessment of lung microbiota in healthy dogs: impact of breed and living conditions

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The lung has been recognized to host a diverse, low biomass bacterial population, identified as the lung microbiota (LM). In human chronic lung diseases (CLDs), the LM is altered compared with that of healthy patients. However, whether alterations are a cause or a consequence of the disease is still unclear. In dogs, the LM has been described mostly in healthy experimental beagles. However, in a previous work from our team, an impact of the living conditions and/or of the breed was suspected between healthy beagles and West Highland white terriers (WHWTs). Recent studies in mice and horses showed modifications in the LM according to the living conditions. These modifications in the LM could predispose individuals to certain CLDs. So, we aimed to assess the breed impact and the influence of living conditions, either experimental or domestic, on the LM, in healthy dogs.

Healthy dogs were sampled, for a total of 48 dogs, and categorized into 5 groups: experimental Beagle (EB), Shepherd (S), Terrier (T), Brachycephalic (Br) and WHWT dogs, a breed with high susceptibility for canine idiopathic pulmonary fibrosis (CIPF). Bronchoalveolar lavage fluid (BALF) was obtained under anaesthesia in each dog. After DNA extraction from BALFs, a PCR targeting the V1-V3 region of the 16S rDNA was performed. Amplicons were then sequenced on a MiSeq Illumina sequencer. Taxonomical assignation and microbiota community analysis were done with MOTHUR V1.40 with an OTU clustering distance of 0.03.

Results showed that the bacterial load was higher in EB dogs ($P < 0.0001$). The AMOVA results indicated differences between EB group compared with S and T groups ($P < 0.005$). Significant differences in relative abundances at the family and the genus level were found. The genus *Hydrogenophilus* was higher in EB and the genera *Brochothrix*, *Limnohabitans*, *Parabacteroides* and *Curvibacter* were higher in WHWT compared with other groups ($P < 0.05$). In each group, specific genera were found as indicators of discrimination ($p < 0.05$). Bacterial richness was higher in WHWT than in EB, S and T groups ($P < 0.001$), but there were no significant differences for the evenness and the α-diversity between groups.

Our study demonstrated an effect of the living conditions on the LM. Breed differences were also shown. This LM modifications might be related to breed susceptibility to lower respiratory diseases, such as CIPF in WHWTs and need to be considered in future analyses on the role of LM disturbances in diseases.

Disclosures

No disclosures to report.

ESVIM-O-3

Protein biomarkers in regurgitation, vomiting, and cough: proteomic characterization of canine gastric fluid by liquid chromatography mass spectrometry (LCMS)

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Reflux and aspiration in people cause and exacerbate respiratory diseases. Protein biomarkers in humans detect extra-esophageal reflux (EER) in the absence of overt dysphagia, regurgitation, or vomiting. In dogs, reflux likely contributes to respiratory disease pathogenesis and progression.

Our study objectives were as follows: (1) Perform comprehensive analysis of the canine gastric fluid (GF) proteome. (2) Compare the oropharyngeal (OP) proteome in normal, vomiting/regurgitating and coughing dogs to identify potential biomarkers for EER and aspiration. (3) Compare biologic function of proteins between sites.

Twenty-three client-owned dogs were prospectively enrolled. Canine GF samples (n = 5) and OP swabs in normal (n = 6), vomiting/regurgitating (n = 7), and coughing (n = 5) dogs were evaluated. Protein digests were analyzed by liquid chromatography mass spectrometry (LCMS). Data were searched against the NCBIMus database. Differential abundance (DA) of proteins and functionality between groups was evaluated by Fisher Exact test and ANOSIM respectively. A $P < 0.0004$ and $p \leq 0.01$ respectively were considered significant after correction for multiple comparisons.

Across sites, 504 individual proteins were identified. Normalized spectral abundance demonstrated pancreatic proteins were increased compared to pepsin in GF. Significant differences in DA between groups was noted ($P < 0.0001$): GF vs normal (n = 130), cough vs. normal (n = 22), reflux/vomiting vs. normal (n = 20). Marked between-dog variation was observed for proteins with DA. Functional proteome was dissimilar between all groups ($P \leq 0.01$).

In conclusion, the proteomic composition of OP swabs varies between health and disease. Variable abundance may impact utility of individual gastric proteins as disease biomarkers and investigation into biomarker panels is warranted.

Disclosures

Disclosures to report.

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ESVIM-O-4

Serum 25-hydroxyvitamin D in dogs with sinonasal aspergillosis

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Vitamin D has important roles in both innate and adaptive immune responses. Hypovitaminosis D in people has been associated with increased susceptibility and severity of illness with a variety of inhaled respiratory pathogens. Sinonasal aspergillosis (SNA) is a common cause of chronic nasal disease that remains a challenge to treat and is associated with substantial morbidity and non-survival in dogs. There is a lack of biomarkers that can predict the development of SNA and treatment outcome in dogs. Therefore, the 2 objectives of this study were 1) to compare serum 25(OH)D concentrations in dogs with SNA and healthy control dogs and 2) to determine if serum 25(OH)D concentrations can predict first time treatment outcome.

Nine dogs with SNA and 8 healthy control dogs were in included in the retrospective arm of this study. Serum samples were obtained from these 17 dogs between 2017 and 2019 and stored at −80° C until analysis. Diagnosis of SNA was based on presence of compatible clinical signs and per-endoscopic identification of fungal plaques with turbinate destruction and fungal culture. Treatment was deemed successful if there was resolution of clinical signs and absence of fungal plaques on follow-up rhinoscopy. Serum 25(OH)D was measured using HPLC.

Seventy-eight percent of dogs with SNA had complete fungal debridement. Seven dogs had follow-up available of which 57% (4/7) had first time treatment success. Dogs with SNA had significantly decreased serum 25(OH)D concentrations (mean, SD; 26.7 ng/ml, 15.3 ng/ml) compared to healthy control dogs (40.2 ng/ml, 7.4, p = 0.03), two-tailed *t*-test). Serum 25(OH)D concentrations was not associated with first time treatment outcome ($P = 0.33$, logistic regression).

These results suggest that serum 25(OH)D is significantly decreased in dogs with SNA as well as a potential immunologic contributory role in the disease. In addition, these results support pursuit of the prospective arm of the study that will investigate potential mechanisms for decreased serum 25(OH)D in dogs with SNA and its role in treatment outcome.

Disclosures

No disclosures to report.

ESVIM-O-5

Plasma mRNA cathelicidin expression in hospitalized critically ill dogs

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Cathelicidin is an antimicrobial peptide essential to the innate immune system and whose circulating concentrations are well documented to have a positive association with 25-hydroxyvitamin (OH) D in people. Decreased 25(OH)D concentrations are predictive of survival in people and dogs with critical illness. Decreased production of cathelicidin has been proposed to be an important mechanism linking decreased 25(OH)D concentrations with survival in critically ill people. Therefore, we aimed to evaluate the association between plasma cathelicidin mRNA expression and serum 25(OH)D concentration and survival in critically ill dogs.

Nineteen dogs (sepsis, n = 7, critically ill without sepsis n = 5, healthy controls n = 7) from a cohort (n = 99) with previously measured 25(OH)D concentrations were randomly selected. Plasma mRNA was extracted, reverse transcribed and resulting cDNA was used as template for real-time PCR using specific primers for canine cathelicidin.

There was not a significant difference in cathelicidin concentrations between dogs with sepsis (mean, SD; 8.29, 0.47), critically ill without sepsis (8.90, 0.82), and healthy controls (8.52, 0.62; $P = 0.32$; one-way ANOVA). There was not a significant correlation between 25(OH)D and cathelicidin expression (rs = −0.14, $P = 0.55$; spearman correlation). Thirty-three percent (4/12) of critically ill dogs did not survive to discharge. Cathelicidin concentrations were not predictive of survival to discharge ($P = 0.64$; logistic regression). Dogs with sepsis (mean, SD; 26.1 ng/ml, 9.2, $P = 0.001$), and critically ill dogs without sepsis (18.7 ng/mL, 9.4, $P < 0.001$) had significantly decreased serum 25(OH)D compared to healthy controls (53.9 ng/ml, 15.7, one-way ANOVA). Severity of illness defined by APPLEfast scores ($P = 0.22$), serum 25(OH)D concentration ($P = 0.25$), and presence of sepsis ($P = 0.42$; logistic regression) were not predictive of survival.

Based on this small cohort, there was no association between plasma cathelicidin mRNA expression and serum 25(OH)D concentrations or outcome in critically ill dogs. The negative results from this preliminary investigation should be interpreted with caution as the number of dogs enrolled was small and type II error might have been a contributing factor to a lack of association.

Disclosures

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ESVIM-O-6

Hereditary methemoglobinemia in dogs caused by cytochrome b5 reductase deficiency associated with variants in CYB5R3

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Methemoglobin forms when hemoglobin iron is oxidized from ferrous iron (Fe^{2+}) to ferric iron (Fe^{3+}), which is incapable of binding O_2. Hereditary methemoglobinemia in dogs is most commonly caused by cytochrome b_5 reductase (CYB5R) deficiency. We recently reported on 2 likely-causal *CYB5R3* variants in 1 dog. Scattered case reports show varied clinical consequences of hereditary methemoglobinemia in dogs. Therefore, the objectives of this study were to determine the metabolic and molecular bases and characterize clinicopathological features of canine CYB5R deficiency.

Twenty-five dogs from various breeds were investigated for unexplained mild to severe non-cardiopulmonary cyanosis. EDTA blood was used to determine methemoglobin concentrations (methb%) and CYB5R enzyme activities, and analyze *CYB5R3* gene for disease-causing mutations. The previously documented nonsynonymous Ile190Leu *CYB5R3* variant was homozygous in 16 of the dogs and heterozygous in 3 dogs. The other previously identified variant was not found in any other dogs. Whole genome sequencing analysis identified 2 novel nonsynonymous *CYB5R3* variants: Arg219Pro (homozygous in 7 dogs) and Thr202Ala (homozygous in 1 dog).

The mean methb% was 23.8% (±11.5 [SD]) and median CYB5R activity was 10.9% (Q1, Q3;5.8,24.1). Dogs homozygous for the Arg219Pro variant had significantly greater methb% (36.2± 9.9%) and more clinical signs than those homozygous for Ile190Leu variant (15.7%, 6.0, $P < 0.001$). Beside cyanosis, 70% of dogs exhibited ≥1 other clinical sign.

In conclusion, this study reveals the clinical, metabolic, and molecular genetic variation seen in CYB5R deficiency in dogs. The Arg219Pro variant appeared to cause a more severe phenotype than the Ile190Leu variant. The 4 DNA variants identified can been readily used as genetic screening tests.

Disclosures

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ESVIM-O-7

Differences in clinical presentation of common dog breeds diagnosed with primary IMHA

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Canine primary immune-mediated haemolytic anaemia (IMHA) is a common haematological emergency and the incidence differs among breeds. It is unknown whether dogs of predisposed breeds present with different degrees of disease severity, different clinical signs or differences in clinicopathological findings. This information could be used in the clinic to guide treatment and provide prognostic advice for owners. The primary aim of this retrospective study was to identify any differences in these findings and outcomes in some of the commonly-affected breeds. We hypothesised that spaniel breeds would have findings associated with more severe disease and poorer outcomes compared to other breeds.

Electronic records of canine patients of a tertiary referral centre were searched over a 10 year period for IMHA-related keywords. A recently-published algorithm was used to assess the confidence of diagnosis of IMHA. Cases with sufficient features of IMHA were analysed further but were excluded if investigations revealed possible underlying disease or were incomplete. Cases were also excluded if they had received immunosuppressive or blood transfusion therapy more than 48 hours prior to presentation. For cases fulfilling inclusion criteria, the most frequent five breeds were selected for analysis, as well as 'cross breeds' to serve as a control group. Data were collected on the historical and clinical signs, clinicopathological findings and outcomes.

For continuous variables, one-way ANOVA with a Dunnett's multiple comparisons test was performed to compare each breed to the control group. For categorical variables, z-tests were used to compare proportions.

689 records were identified containing IMHA keywords, of which 253 fulfilled all inclusion criteria. Cocker Spaniels (n = 36), Springer Spaniels (n = 25), Labradors (n = 15), Jack Russell Terriers (n = 13) and Shih-tzus (n = 11) were selected for final analysis, alongside crossbreeds (n = 24) as a control group. Labradors were significantly older when compared to the control group (mean 8.6 v. 5.9 years, $P = 0.029$). Surviving Cocker Spaniels had significantly shorter hospitalisation periods when compared to the control group (mean 5.97 v. 8.91 days, $P = 0.031$). Compared to cross breeds, a greater proportion of Jack Russell terriers survived to discharge (100% v. 75%, $P < 0.05$).

Spaniel breeds did not appear to be affected more severely, with surviving Cocker Spaniels requiring shorter hospitalisation periods. Jack Russell Terriers may have a more favourable outcome but this finding could be confounded by the effect of treatment and requires confirmation.

Disclosures

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ESVIM-O-8

Diagnostic imaging findings in a referral population of dogs diagnosed with immune-mediated haemolytic anaemia

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Diagnostic imaging in dogs diagnosed with immune-mediated haemolytic anaemia (IMHA) is indicated to rule out systemic diseases mimicking or triggering the IMHA. However, studies, providing descriptions of thoracic and abdominal abnormalities, supporting a clinical decision making in dogs with IMHA are lacking.

The aim of this study was to describe changes documented on thoracic and abdominal imaging of dogs with confirmed IMHA.

Medical records from a referral hospital were searched from 2015 and 2018 for all dogs that were diagnosed with IMHA and underwent

thoracic and abdominal imaging by radiography, ultrasound or computed tomography (CT). To be included, a complete history, clinical and routine laboratory examination findings had to be available as well.

A total of 51 client owned dogs fulfilled the inclusion criteria and were included in this retrospective study. The median age of dogs was 6.4 years (range, 7 months to 11.4 years). There were 9 sexually intact females, 26 spayed females, 8 sexually intact and neutered males, respectively. CT of thorax and abdomen were performed in 7 and radiographs of the thorax and abdominal ultrasound in 44 dogs. Fine needle aspirates (FNAs) were collected in 28 dogs.

CT and radiographs of the thorax revealed abnormalities in 11/51 dogs. Sternal lymphadenopathy and cardiomegaly were documented in 4 dogs respectively and pleural effusion in 2 dogs.

CT and ultrasound of the abdomen revealed abnormalities in 44/51 dogs. Hepatomegaly and peritoneal effusion were present in 20 dogs respectively, gallbladder wall thickening in 17, gallbladder sludge in 15, splenic nodule in 13, diffuse steatitis in 11, enlarged pancreas and abdominal lymphadenopathy in 8 and splenomegaly in 7 dogs.

Hepatomegaly and splenomegaly were further investigated via FNA in 19/20 dogs and revealed extramedullary haematopoiesis in 12 hepatic and 14 splenic samples. Cholestasis, vacuolar hepatopathy and reactive lymphoid hyperplasia were documented in less than 50% of samples collected from both organs. Round cell neoplasia was documented via FNA of liver, spleen and abdominal lymph nodes in one dog. Cholecystocentesis was performed in 10/17 dogs with gallbladder wall thickening and revealed positive bile culture in 3 dogs.

In this population of dogs with IMHA thoracic imaging abnormalities were uncommon. Hepatomegaly with gallbladder wall thickening and peritoneal effusion were the most common abdominal imaging findings with bactibilia confirmed in one third of collected bile samples. Hepatosplenomegaly and abdominal lymphadenopathy were uncommonly associated with neoplasia.

Disclosures

No disclosures to report.

ESVIM-O-9

Determination of Blood Groups DEA 1, DEA 4, DEA 5, Dal, and Kai 1/2 in Different Canine Breeds

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Dogs have >12 blood group systems, but little is known about their frequency in Europe. Here we report on an extensive typing survey with available reagents and established or new clinical kits in purebred dogs.

Leftover EDTA blood samples were examined using an immunochromatographic strip method for *DEA 1*, a gel column technique for *Dal* and *Kai 1/2*, and new agglutination card test for *DEA 4* and *DEA 5* (and partially compared with gel column). Monoclonal antibodies were used for *DEA 1* and *Kai 1/2* typing and polyclonals for all other types.

Among the 164 examined dogs, 62% were *DEA 1+*, 100% *DEA 4+*, 11% (card)/14% (gel) *DEA 5+*, 97% *Kai 1+* and 2% *Kai 2+*. Blood from most *DEA 1+* dogs bound strongly positive on the strips. Agglutination reactions for card and gel tests were concordant for *DEA 4+* samples. In contrast, the reactions for *DEA 5* were generally less in the agglutination test than in the gel column test with some discordant reactions. None of the dogs were *Kai 1+/2+*, and only one was *Kai 1−/2-*. *Dal*-dogs were found in Cane Corso, Dalmatian, Doberman, Maltese, Mastiff, Shih Tzu, and Pug dogs. Various blood group patterns were observed in other breeds.

In this first extensive blood typing survey of purebred dogs from Europe, the proportion of positive and negative blood types were similar to those in US. The newer typing techniques seem to work well (*DEA 5* cards are being improved) and will be useful to detect and prevent specific blood type incompatibilities in clinics.

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ESVIM-O-10

Systemic AA-amyloidosis in shelter cats and shedding of amyloid fibrils

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Systemic AA-amyloidosis is a protein misfolding disease of humans and animals arising from the formation of amyloid fibrils from the acute phase protein serum amyloid A. In animals it is common in chickens and cheetahs kept in captivity. Latest studies showed that transmission of systemic AA-amyloidosis occurs in cheetahs and involves fecal shedding of amyloid fibrils. Client-owned domestic shorthair cats are rarely affected by systemic AA-amyloidosis. Whether systemic AA-amyloidosis is common in domestic shorthair cats living in shelters and shedding of amyloid fibrils occurs is unknown. Thus, aims of the study were to determine the frequency of systemic AA-amyloidosis in different cat shelters and to investigate excretion of amyloid fibrils.

Any cat from 3 shelters was included if necropsy was performed within 6 hours from death. Liver, kidney, spleen and bile were obtained during necropsy. Clinical and laboratory results were retrieved from available medical archives. AA-amyloid was identified in tissues by hematoxylin-eosin and Congo-red staining, and confirmed by immunoblotting. Shedding of amyloid fibrils was investigated with immunoblotting in bile samples. Descriptive statistics and non-parametric tests were used.

Deposition of AA-amyloid was observed in the liver, kidney or spleen of cats that died in each of the 3 shelters; in particular, the prevalence was 40% (8/20 cats), 50% (8/16 cats) and 85.7% (6/7 cats), respectively. In 13 cats all 3 organs were involved, in 4 cats 2 organs, and in 5 cats 1 organ. In cats with any of the 3 organs being AA-amyloid-positive, sensitivity and specificity of AA-amyloid identification in the bile was 83.3% and 58.3%, respectively; in those with only the liver involved, 87.5% and 50%. Of note, semiquantitative analysis of AA-amyloid in the bile showed that none of 12 cats without organ deposition of AA-amyloid had a score > 1+, while scores 2+ or 3+ were identified in 66.7% (8/12 cats) of those with any of the 3 organs involved (P = 0.001). In 18 cats with AA-amyloid-positive organs medical records showed that 9 had kidney or liver involvement: 4 had azotemia, 3 proteinuria, and 3 hemoabdomen due to spontaneous hepatic rupture.

In conclusion, the prevalence of systemic AA-amyloidosis appears to be elevated in shelter cats and shedding of amyloid fibrils occurs in the bile. Cats with systemic AA-amyloidosis are more likely to shed higher amounts of AA-amyloid. Whether bile excretion of AA-amyloid indicates that horizontal transmission is relevant to the disease in shelter cats remains undefined.

Disclosures

No disclosures to report.

ESVIM-O-11

Treatment of non-lactate metabolic acidosis in hypovolemic and normovolemic dogs: chloride-free iso-osmolar solution with elevated Strong Ion Difference versus Ringer's Lactate solution

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The use of sodium bicarbonate-based solutions for treating metabolic acidosis is currently debated. The alkalizing effect of solutions with elevated Strong Ion Difference (SID) has recently been described. The primary aim of the study was to evaluate the safety and efficacy of a chloride-free iso-osmolar solution with elevated SID (Hyper-SID), compared to Ringer's lactate used for the treatment of metabolic acidosis.

Hyper-SID solution was prepared by adding 145 mEq/L of sodium, 145 mEq/L of lactate, 10 mEq/L of potassium and 10 mEq/L of aspartate to the sterile water for injections. The calculated SID was 155 mEq/L.

This prospective, multicenter, randomized study evaluating the efficacy and safety was authorized from the Ethical Committee. Hospitalized dogs with an excess of bases ≤ −10 mEq/L were enrolled in the study if the baseline lactate level was not greater than 4 mEq/L. Patients were classified by ultrasonographic methods as hypovolemic or normovolemic and randomized with random number generation in the type of fluid to be received (Ringer's Lactate in the RL group and Hyper-SID in the H-SID group). Normovolemic and hypovolemic dogs received 4 hours infusion at the rate of 4 or 10 mL kg^{-1} hr^{-1} respectively. Blood gas analysis, before fluid infusion (T0) and after 4 hours (T4), were compared for the following parameters: Be-ecf, pH, PCO2, Sodium, Potassium, Chloride, Lactate, SID^3, SID^4. After normality distribution analysis, variables were described and evaluated using non-parametric statistics (significance level set to 5%).

Forty dogs were included in the analysis, median age was 110 months (12-192) and median weight 16 kg (3-39). Dogs classified as hypovolemic were 9/40 in the RL group and 13/40 in H-SID group, while dogs classified as normovolemic were 8/40 in the RL group and 10/40 in the H-SID group. The basal Be-ecf was not different between the treatment groups (P > 0.05). In normovolemic patients the median increase in Be-ecf at T4 in the RL and H-SID group was 0.8 mEq/L and 4.5 mEq/L (P = 0.004) respectively, whereas in hypovolemic patients was respectively of 1.3 mEq/L and 11,5 mEq/L (P = 0.0001). Lactate value greater than 5 mEq/L was not documented in any patient during the study.

The Hyper-SID solution proved to be effective and superior to the RL solution in the treatment of non-lactate metabolic acidosis in hospitalized dogs. In the sample of subjects belonging to the H-SID group, no side effects or raising lactatemia was reported during the infusion period.

Disclosures

No disclosures to report.

ESVNU-O-1

Serum symmetric dimethylarginine in dogs and cats with acute kidney injury treated with intermittent hemofiltration

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Serum symmetric dimethylarginine (SDMA) concentration has been shown to be a marker of renal dysfunction in dogs and cats. In humans, dialysis lowers SDMA but less efficiently than urea because the former has a larger distribution volume. Information concerning SDMA in animals undergoing extracorporeal renal replacement therapy is yet lacking. The aim of this study was to describe the dynamic of SDMA concentration in dogs and cats with acute kidney injury treated with intermittent hemofiltration (IHF).

Reports of IHF performed between October 2017 and 2018 were collected. Medical records were reviewed and cases with pre- and post-IHF biochemical profiles were included. IHF was performed according to standard methods. Urea, creatinine and SDMA reduction-ratio (URR, CreaRR, SDMARR) was calculated for each IHF. In dogs and cats separately, first sessions of IHF were included in group A, second sessions in group B and third sessions in group C. URR, CreaRR and SDMARR were compared within each group with non-parametric tests.

Overall, 39 IHF sessions were performed in 14 dogs and 8 cats; specifically, 13 dogs and 8 cats in group A, 9 dogs and 5 cats in group B, 3 dogs and 1 cat in group C. Five (35.7%) dogs and 3 (37.5%) cats survived. Median SDMA concentration pre-IHF was 62 μg/dl in dogs

(range: 15- > 100) and 78 µg/dl in cats (range: 41- > 100). In dogs, median values of URR, CreaRR and SDMARR were 37.1% (range: 12.0-66.3), 35.6% (range: 12.0-67.7) and 16.7% (range: −86.7-42.6), respectively; in cats, their median values were 38.6% (range: 20.4-52.4), 41.5% (range: 21.2-54.1) and 33.8% (range: 7.1-50.0). In 8 of 25 (32%) IHF sessions in dogs, SDMARR was either negative (n = 5) or 0 (n = 3). These 8 sessions were performed in 6 dogs: 4 had leptospirosis and 2 poisoning (grape and ethylene glycol), 4 died. The median SDMARR was lower than URR in groups A and B in dogs (P = 0.021, P = 0.008) and not different in cats. In group A in dogs, SDMARR was also lower than CreaRR (P = 0.007). No other differences were observed in both species.

In conclusion, SDMARR did not represent an accurate marker to assess efficiency of IHF in dogs. The partial inability of IHF to clear SDMA, the large distribution volume of SDMA or other unknown conditions affecting post-IHF SDMA concentration might have contributed to this result. Differently, SDMARR may be reliable to evaluate IHF efficiency in cats.

Disclosures

No disclosures to report.

ESVNU-O-2

Non-obstructive 'acute on chronic' kidney disease in the cat: is it possible to predict survival?

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Cats with chronic kidney disease (CKD) are often presented when a superimposed acute uremic crisis happens and any prognostic indicator would be of relevance to feline practitioners.

The aim of this study was to determine if some variables commonly assessed in azotemic cats in that particular context could be accurate predictors of survival.

Medical records over 4 years from azotemic cats hospitalized for at least two days with acute signs and confirmed CKD based on ultrasonographic findings and/or previously documented azotemia were retrospectively reviewed. Cats with potential toxic, neoplastic or obstructive cause of azotemia were not included. Signalment, clinical signs (combined into a clinical severity score [CSS]), selected laboratory and diagnostic imaging results and outcome were registered. A machine learning-based classification and regression trees method was used to assess predictors of survival at 7, 30, 90 and 180 days.

Thirty-two cats were included in this study. Overall median survival time was 28 days (range: 0 to 1566 days). Plasma creatinine concentration after 2 days of hospitalization ($Crea_{48}$) was the best predictor of survival. A $Crea_{48}$ of less than 64 mg/L best predicted survival at 7, 30 (Se = 0.87; Sp = 0.77) and 90 days. A $Crea_{48}$ of less than 38 mg/L best predicted survival at 180 days. When $Crea_{48}$ was censored from the analysis a lower CSS and a younger age were also predictive of survival up to 90 days.

This study confirms that $Crea_{48}$ is a good predictor of short and medium-term survival in cats with CKD presented in uremic crisis.

Disclosures

Disclosures to report.

M.Faucher: Webconference for Boehringer Ingelheim A.Combes: Speaker for Veterinarius.

ESVNU-O-3

Effect of measurement location on systolic blood pressure (SBP) readings in out-patient and in-patient dogs

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Non-invasive blood pressure (BP) measurement is widely used to diagnose hypertension, yet many factors affect measurement accuracy. ACVIM guidelines propose a standard protocol which suggests BP should be performed with the owner present. Studies have not been performed to evaluate this in dogs nor to evaluate whether SBP varies in different hospital locations.

The aims were: 1) to determine whether SBP was different in out-patient dogs when measured in the consult room with the owner or a quiet room away from the owner and 2) to determine whether SBP measured in in-patient dogs was different when measured in their kennel or in a quiet room away from other dogs.

Each dog had their BP measured using high definition oscillometry in 2 locations consecutively; for the 25 out-patients, in the consult room with the owner and in a quiet room and for the 25 in-patients, in their kennel and in a quiet room. The measurement location order was randomized. Blood pressure measurement methods were standardized and based on the ACVIM consensus statement and instrument manufacturer recommendations. The BP cuff was placed on the tail. All measurements were taken by a single veterinary surgeon using the same cuff and body position for each dog in both locations. The pulse waveform was visualized during measurement and 5 valid readings were collected and averaged in each location. Descriptive statistics were performed and the number of dogs with >20% difference between locations was calculated. In addition, SBP was classified using ACVIM consensus statement hypertension categories.

For out-patients, SBP was a mean of 6 mmHg (SD: 20) higher in a quiet room. In 6 dogs (24%), SBP was >20 mmHg different between the locations (3 were higher in the consult room). Fourteen out-patients (56%) changed hypertension category when the location changed (9 were higher away from the client).

For in-patients, the mean difference in SBP between the locations was 0 mmHg (SD: 18), however there was >20 mmHg difference in SBP in 7 dogs (28%) (4 were higher in kennels). Eleven in-patients (44%) changed hypertension category when the location changed (6 were higher in kennels).

Although the mean difference in SBP in both locations was small for both in- and out-patients, approximately a quarter of dogs in both groups had >20% differences between locations and approximately half changed hypertension category. This suggests that to measure changing trends in any individual animal, the location of measurement should be kept constant.

Disclosures

Disclosures to report.

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ESVNU-O-4

Immune-complex glomerulonephritis in cats: a retrospective study based on clinico-pathological data and morphological features

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Chronic kidney disease (CKD) has typically a non-immune mediated origin in cats and immune-complex glomerulonephritis (ICGN) has been scarcely described. To date, few descriptions of ICGN have been published in cats, with membranous glomerulonephropathy (MGN) being most commonly reported. Therefore, aims of this study were to characterize morphological diagnoses of ICGN by light and electron microscopy in a large cohort of cats and to identify associations with clinical and laboratory findings. Additionally, comparisons were performed with cats affected by non immune-complex glomerulonephritis (non-ICGN).

Renal biopsies of cats with ICGN and non-ICGN examined between 2010 and 2019 were considered if both light and electron microscopy were available. Data collected for analysis included breed, sex, age, feline immunodeficiency virus (FIV) and feline leukemia virus (FeLV) status, serum creatinine concentration, urine protein-to-creatinine (UPC) ratio and systolic blood pressure (SBP). Differences between morphological diagnoses of ICGN and non-ICGN, including tubulointerstitial damage score, were investigated with Kruskal-Wallis and chi-squared tests. The likelihood of diagnosing ICGN versus non-ICGN was explored with logistic regression.

Sixty-eight cats were included, 37 (54.4%) with ICGN and 31 (45.6%) with non-ICGN. In cats affected by ICGN, 18 (48.6%) had MGN, 14 (37.8%) membranoproliferative glomerulonephritis (MPGN) and 5 (13.5%) mesangioproliferative glomerulonephritis. Breed, sex, age, FIV and FeLV status, creatinine, UPC ratio and SBP were not different among morphological diagnoses. In cats with non-ICGN, 11 (35.5%) had end-stage CKD, 9 (29%) focal and segmental glomerulosclerosis, 6 (19.4%) global mesangiosclerosis, 2 glomerular atrophy and renal dysplasia (6.5%) and 1 (3.1%) amyloidosis. Eight (25.8%) cats with non-ICGN had grade 1 tubulointerstitial damage, 13 (41.9%) grade 2 and 10 (32.3%) grade 3; creatinine and UPC ratio were positively associated with grades ($P = 0.001$, $P < 0.001$). Cats with ICGN were more likely to have FIV or FeLV infection than those with non-ICGN (13/37 vs. 0/21; $P = 0.002$) and had higher mean UPC ratio (7 ± 3.2 vs. 3.6 ± 2.3; $P < 0.001$). Mean age of cats with ICGN was lower than those with non-ICGN (9.2 ± 3.3 vs. 10.9 ± 3.3 years; $p = 0.042$).

In conclusion, MGN and MPGN were the two most common morphological diagnoses of ICGN in cats, but clinical and laboratory findings did not allow their differentiation. In cats with non-ICGN, serum creatinine concentration and UPC ratio were associated with the degree of tubulointerstitial damage confirming previous literature. Cats with retrovirus infections, higher UPC ratio and younger age were more likely affected by ICGN than non-ICGN.

Disclosures

No disclosures to report.

ESVNU-O-5

Short course of immune-suppressive doses of prednisolone is associated with renal hyperfiltration and changes in hydration and electrolyte status in healthy beagle dogs

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Glucocorticoids influence renal function and are frequently prescribed in dogs. The extent and duration of their effects is scarcely described. Our objectives were to assess prednisolone effects on renal, electrolytic and hydration status in healthy dogs.

24-hours urine collection, total body water content (tBWC) using pharmacokinetic equation, glomerular filtration rate (GFR) using plasma exogenous creatinine clearance, serum symmetric dimethylarginine (SDMA), complete plasma biochemistry and urinalysis (including urine protein-to-creatinine ratio) were obtained in 14 beagles dogs in a 2x2 blinded against placebo cross-over design. One group received a 7-days course of immune-suppressive prednisolone while the second received the placebo. After a 4-weeks wash-out period, groups were switched. Blood and urine were collected before and after each treatment period and during wash-out. A general linear model was used to test period, sequence, treatment, dog and weight effects. A Dunnett test was used to evaluate the effect of steroid with day 0 serving as control. Correlations were assessed using Pearson's coefficient.

Prednisolone significantly affected body weight ($P < 0.001$; mean difference − 1.1 kg), GFR ($p = 0.01$; +0.6 mL/kg/min), SDMA ($P < 0.001$; −2.4 µg/dL), creatinine ($P < 0.001$; −14.4 µmol/L), urea ($P < 0.001$; +1.53 mmol/L), chlorides ($P < 0.001$; −8.9 mmol/L), bicarbonates ($P = 0.006$; +1.4 mmol/L), magnesium ($P < 0.001$; +0.19 mmol/L), total proteins ($P < 0.001$; +6.3 g/L), albumin ($P < 0.001$; +8.7 g/L), tBWC ($p = 0.022$; −43 mL/kg), urine specific gravity ($P < 0.007$; −0.015) and voided urine ($P < 0.001$; +274 mL). SDMA ($r = -0,51$) and creatinine ($r = -0,74$) correlated significantly ($p = 0,001$) with GFR. All variations became non-significant after the wash-out.

Prednisolone at immune-suppressive dosage rapidly decreases tBWC and body weight and induces relevant reversible renal hyperfiltration and clinically significant variations of several analytes.

Disclosures

Disclosures to report.

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ESVNU-O-6

Characterization and in vitro susceptibility of clinical feline UPEC isolates to an E. coli probiotic as a potential therapeutic for urinary tract infection

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The rise in antibiotic resistance amongst urinary tract infections (UTIs) in both cats and dogs underscores the need for non-antibiotic approaches to UTIs. The probiotic *Escherichia coli* Nissle-1917 (EcN) has many benefits including antimicrobial activity against many human pathogens including uropathogenic E. coli (UPEC). The aim of this study was to phylogenetically characterize UPEC in feline UTI cases and investigate the in vitro susceptibility of these isolates to EcN.

Twenty-two cats with positive *E. coli* urine cultures were included in the study. Samples used in this study were obtained from surplus urine collected for routine evaluation of possible urinary tract infections. Characterization of UPEC isolates was performed by clade analysis, serotyping and virulence factor analysis by multiplex PCR testing. EcN effectiveness against UPEC isolates was tested in vitro using microcidin plate analysis.

Clinical signs were consistent with lower urinary tract infection in all cats. Serogroup and virulence factors correlated with clade analysis as reported in human UPEC studies. Fifty-nine percent of UPEC isolates were susceptible to the EcN probiotic in vitro. Average zone of growth inhibition from the EcN probiotic was 5.54 mm (Range 2.33-10.61 mm).

UPEC isolates from feline patients were similar to isolates in human patients in pathogenicity, susceptibility, and genetic background. in vitro susceptibility of feline UPEC isolates were frequently susceptible to the EcN probiotic through growth rate characteristics and/or microcin production. These findings suggest the potential use of Nissle as a novel therapeutic to treat feline urinary tract infections.

Disclosures

Disclosures to report.

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ESVNU-O-7

Prognostic factors in dogs with common causes of proteinuria

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Little is known about the impact of increased urine protein:creatinine ratios (UPC) on survival of dogs with different underlying diseases, and associated risk factors for death are not established. Therefore, the aim of this study was to assess dogs with severe proteinuria (UPC at least once during the disease measured >2.0) for their survival time, underlying diseases and possible prognostic factors like UPC at time of diagnosis, creatinine, urine specific gravity, albumin and haematocrit.

Between 2014 and 2015, 89 dogs with severe proteinuria were retrospectively analysed. Among them, 46 dogs were diagnosed with glomerulopathy (median UPC: 6.0; range: 1.4-21.2), 16 dogs with Cushing's disease (median UPC: 4.4; range 2.1-14.1), 11 dogs with leishmaniosis (median UPC: 4.7; range 2.1-19.8) and 16 dogs with various diseases (median UPC 3.1; range 1.9-9.5).

Over all dogs, increased UPC was identified as a risk factor for death ($P < 0.01$). Median time of survival was 42 days. UPC and time of survival did not differ significantly between the groups. Among dogs with glomerulopathy, identified significant risk factors for death included increased UPC ($P = 0.03$), increased creatinine ($P < 0.01$), low haematocrit ($P = 0.04$) and low urine specific gravity ($P = 0.03$). In dogs with Cushing's disease, only urine specific gravity was a significant risk factor for death ($p = 0.05$). In dogs with leishmaniosis, increased UPC and creatinine were significant risk factors for death ($P < 0.01$; $p < 0.01$).

Increased UPC is a risk factor for death in dogs with glomerulopathy and leishmaniosis, but not with Cushing's disease. This can be explained by different pathogenesis leading to proteinuria.

Disclosures

Disclosures to report.

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ESVNU-O-8

The effect of dietary sodium on urinary calcium and calcium oxalate relative supersaturation (CaOx RSS) in dogs

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Calcium oxalate (CaOx) recurrence is challenging to manage in dogs. Relative supersaturation (RSS) is a measure of crystallization risk, and lower urinary concentration of CaOx precursors can decrease CaOx RSS. Urinary dilution is the primary strategy to decrease CaOx RSS, and can be achieved by increased sodium content of pet food to drive water intake. However, humans with CaOx renoliths are advised to decrease sodium intake because of the potential increase in renal calcium excretion, which could increase the risk of the disease.

The aim of this study was to compare the effect of two dry pet foods differing only in sodium chloride content on urinary volume, urinary calcium excretion and concentration, and CaOx RSS in a cross-over study using 8 healthy colony dogs. A base diet was produced with a sodium content of 0.3% as fed (LSD). This formula was supplemented with NaCl to achieve high-sodium diet (HSD) (1% sodium as fed - comparable to commercially available high-salt urinary diet). The dogs were fed each diet for 7 days followed by 3 days of urine collection. Urinary minerals were measured on pooled 3-day samples using ionic chromatography. CaOx RSS was calculated using SUPERSAT software. The impact of diet on urinary volume, calcium excretion and concentration, oxalate concentration, and CaOx RSS was evaluated using the Wilcoxon Rank-Sum test (significance set at $P < 0.05$). Data are presented as median[25th,75th percentile].

The HSD led to an increase in urine volume (LSD: 19.1[15.0, 23.3] vs HSD: 37.0[33.7, 46.9] ml/kg BW/day, $P < 0.05$). Urinary calcium excretion was not significantly different between diets (LSD: 78.6

[72.5104.9] vs HSD: 88.8[79.3,96.4] µmol/kg BW/day, $P = 0.84$), but urinary calcium concentration was lower in HSD (LSD: 4.8[3.2,6.0] vs HSD: 2.5[2.0,2.9] mmol/L, $P < 0.05$). Urinary oxalate concentration was lower in HSD (LSD: 1.4[1.1, 1.8] vs HSD 0.9[0.7, 1.2] mmol/L, $P < 0.01$). CaOx RSS was significantly lower with the HSD (LSD: 24.1 [13.1, 32.7] vs HSD: 12.7[10.4, 12.4], $P < 0.01$).

The results of this study do not support an increase in urinary calcium excretion with an increase in dietary sodium at commercially relevant levels. The increase in urine volume seen with the high salt diet decreased the urinary calcium and oxalate concentrations, and could explain the decrease in CaOx RSS. An increase in dietary sodium therefore decreases the risk of CaOx urolithiasis in short-term feeding trials. Further studies are required to assess the effect of increased dietary sodium when diets are fed for a longer period of time.

Disclosures

Disclosures to report.

All authors are current employees of Royal Canin, Mars Petcare.

ESVNU-O-9

Proliferative urethritis in dogs: long-term follow up and prognosis

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Proliferative urethritis (PU) is an uncommon inflammatory disease with unknown etiology. Data on prognosis of affected dogs are currently lacking. The aim of this study was to describe clinical, endoscopic and histopathological findings and investigate their influence on long-term outcome in dogs with PU.

Medical records of dogs that underwent cystoscopy and were diagnosed with PU by histopathology were retrospectively reviewed (2015-2019). Dogs with a documented follow up period of at least 12 months after diagnosis were included. Results regarding signalment, clinical, cystoscopic, microbiological and histopathological findings were collected. An endoscopic score (ES) for grading urethral lesions was used: mild localized lesions (ES = 1), complete involvement without stenosis (ES = 2), urethral stenosis (ES = 3), and urethral obstruction (UO, ES = 4). Treatments, short-term complications (<6 months), survival time after diagnosis and variables associated with survival (12 months) were investigated. Data were analyzed using descriptive statistics and reported as median and (range). Survival analysis was performed using Cox proportional hazard regression analysis (P value <.05 considered significant).

Thirteen female dogs (5/13 spayed) met the inclusion criteria. Median age was 96 months (72-168), while median body weight was 15.2 kg (10.2-35). Main clinical signs were dysuria (11/13) and stranguria (8/13); urethral obstruction was detected in 5/13 dogs. In 7/8 dogs for which results were available, urine microbial cultures were positive. All dogs had endoscopic evidence of irregular tissue projections into urethral lumen that extended along the entire urethra in 6/13 dog (ES = 2); urethral strictures were noted in 3/13 cases (ES = 3); 4/13 dogs had UO (ES = 4). At histopathology, lymphoplasmacytic (8/13), lymphoplasmacytic and neutrophilic (3/13), and granulomatous (2/13) inflammation was detected. Medical treatment was started in all dogs after diagnosis and included antimicrobials (10/13), glucocorticoids (5/13), nonsteroidal anti-inflammatories (9/13) and immunosuppressive medications (2/13). Median survival time was 12 months (5-36). Short-term complications included relapsing of clinical signs (11/13) and recurrent UTI (11/13); 5/13 dogs required additional interventional or surgical treatments to restore urethral patency. Variables associated with survival were age (HR 1.037, 95% CI 1.0068-1.068, $P = .016$), body weight (HR 0.858, 95% CI 0.746-0.987, $P = .03$), and developing of UO within 6 months after diagnosis (HR 6.050, 95% CI 1.065-34.481, $P = .04$).

The severity of ES at time of diagnosis did not affect long-term outcome of dogs with PU; however small size older dogs that developed UO after diagnosis were more likely to have a worst prognosis.

Disclosures

No disclosures to report.

ISCAID-O-1

Detection of canine and feline parvovirus shedding in asymptomatic shelter cats in Australia using a minor groove binder probe real-time PCR assay

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Canine parvovirus (CPV) and feline parvovirus (FPV) cause severe, often fatal, enteritis in their hosts. A UK study reported faecal shedding of CPV, but not FPV, by 1 in 3 asymptomatic shelter cats. In contrast, an Australian study, utilizing a similar conventional PCR (cPCR) assay reported no CPV shedding and FPV shedding in 1.8% asymptomatic shelter cats.

The aim of this longitudinal study was to determine whether low-level parvoviral shedding, undetectable by cPCR, occurs in Australian shelter cats. Residual faecal samples from the previous Australian study (n = 152), collected from asymptomatic shelter cats on three sampling days (SD1 n = 47, SD2 n = 84, SD3 n = 21) over 12 months, were tested. SD3 occurred during an FPV outbreak.

Parvoviral DNA was detected using quantitative PCR (qPCR), then a minor groove binder real-time PCR assay differentiated FPV, CPV2, CPV2a, CPV2b and CPV2c. Parvoviral DNA was detected in 25 (16.4%) faecal samples (SD1 0/47, SD2, 5/84, SD3 20/21). All positive samples from SD2 and SD3 were CPV2b, and FPV, respectively. Viral loads of CPV2b (1.85-1.18 x 10^5; median 4.62 x 10^2 copies/µL template DNA) were lower than those of FPV (1.26 x 10^3-8.96 x 10^9; median 9.7 x 10^5 copies/µL template DNA). Quantitative PCR was more sensitive than cPCR to detect low-level parvoviral shedding. FPV was shed by 95% asymptomatic shelter-housed cats during an FPV outbreak. A low prevalence of CPV shedding was detected (3.3%). The potential role of cats as a reservoir of CPV infection in dogs varies between populations tested.

Disclosures

Disclosures to report.

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ISCAID-O-2

Prognostic value of systemic inflammatory response syndrome (SIRS) presence, serum acute phase proteins, cholesterol and total thyroxine concentrations in cats with feline panleukopenia: a retrospective cohort study in 70 cats (2010-2018)

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Feline parvovirus (FPV) is a common infectious agent and can be lethal. The aim of this study was to assess the prognostic value of SIRS presence, serum concentrations of serum amyloid A (SAA), haptoglobin, cholesterol and total thyroxine (tT4) in feline panleukopenia.

Retrospective cohort study enrolling cats with feline panleukopenia presented between January 2010 and January 2018. Definitive diagnosis of feline panleukopenia required a positive direct ELISA assay on feces and/or a positive PCR on feces and/or blood. According to their survival status at 28-days from presentation cats were divided into survivors and nonsurvivors. The prognostic importance at presentation of the variables age, sex and reproductive status, presence of SIRS, serum concentrations of SAA, haptoglobin, cholesterol and tT4 (measured on left over serum sample) was investigated univariately and by multivariabile Cox proportional-hazards regression model. Finally, ROC curve analysis was used to identify the best cutoff value (Youden index) for discriminating survivors from nonsurvivors for the prognostic variables resulted statistically significant in multivariable analysis. For all analyses the significance was set to $\alpha = 0.05$.

Seventy cats were eligible for the study, 47 (67%) survivors and 23 (33%) nonsurvivors. At presentation, nonsurvivors were significantly (U = 314.5, $P = 0.005$) younger than survivors, while no difference in reproductive status was found. There was a significant difference ($\chi^2 = 8.02$; $P = 0.005$) in the number of cats fulfilling SIRS criteria between survivors (9/47, 19%) and nonsurvivor (12/23, 52%). SAA concentrations were significantly lower (U = 749, $P = 0.009$) in survivors (median = 83.3 μg/dL; range, 0.1″248.4 μg/dL) compared with nonsurvivors (median = 138.3 μg/dL; range, 3.2″235.8 μg/dL). Serum haptoglobin concentrations were significantly lower (t = −3.24, $P = 0.002$) in survivors (mean = 136.13 ± 69.80 mg/dL) compared with nonsurvivors (mean = 190.09 ± 55.25 mg/dL), while no difference in serum cholesterol concentrations were found. Finally, serum tT4 concentrations were significantly higher (t = 3.546, $P = 0.001$) in survivors (mean = 1.38 ± 0.66 μg/dL) compared with nonsurvivors. In the Cox proportional-hazards regression model only serum tT4 concentration was significantly associated with survival (HR = 0.26, $P = 0.014$). The Youden index identified through ROC curve analysis for serum tT4 concentration was 0.82 μg/dL (sensitivity = 73.9%, specificity = 82.9%; AUC = 0.783, 95% CI, 0.668 to 0.873; $P < 0.0001$).

The present study showed that serum tT4 at presentation in cats with FPV could be use as prognostic factors in predicting the disease outcome.

Disclosures

No disclosures to report.

ISCAID-O-3

Clinical and epidemiological features of the first reported outbreaks of feline calicivirus virulent systemic disease in Australia and in vitro efficacy of three antiviral compounds: nitazoxanide, 2'-C-methylcytidine and NITD-008

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Feline caliciviruses (FCV) are common feline pathogens causing "flu"-like signs. Rarely, FCVs cause virulent systemic disease (FCV-VSD) characterised by jaundice, facial/limb oedema, skin ulceration and death. The pathogenesis of FCV-VSD is poorly understood. The aims of this study were to 1) characterise clinical and virological features of two nosocomial outbreaks of FCV-VSD in Australia in 2015 and 2018, 2) assess efficacy of three antivirals against representative outbreak strains *in vitro*.

Cats presenting with ≥1 sign consistent with FCV-VSD during the outbreaks were included. Viral isolation and whole genome sequencing were performed on residual diagnostic oropharyngeal swabs and/or necropsy tissue. One virus from each outbreak was tested in plaque reduction assays against nitazoxanide (NTZ), 2'-C-methylcytidine (2CMC) and NITD-008. For each antiviral, EC_{50} was determined. Therapeutic index (TI) was derived from the EC_{50} and half maximal cytotoxic concentration in CRFK cells.

Twenty cases (NSW n = 8, QLD n = 12) were identified (age: 1-72 months, median 16.7). Onset of signs was ≤10 days after hospital admission for surgery in 19/20 cases. FCV vaccination status, where known, was current in 15/16 cases. Overall mortality was 45%. Phylogenetic analysis of full FCV genomes revealed co-infections of two FCV lineages in NSW and a separate lineage in QLD. Dose-response inhibition of both FCV-VSD strains was obtained with all antivirals; NTZ $EC_{50,}$ 0.4-0.6 μM, TI 21, 2CMC $EC_{50,}$ 2.7-5.3 μM, TI >18, NITD-008, EC_{50} 0.5 to 0.9 μM, TI >111.

FCV-VSD outbreaks continue to occur in vaccinated cats and are associated with high mortality. Three antivirals were potent inhibitors of FCV-VSD outbreak strains tested.

Disclosures

Disclosures to report.

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ISCAID-O-4

Antibody response to feline calicivirus vaccination in healthy adult cats

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It is unknown how cats in the field react to feline calicivirus (FCV) vaccination.

This study evaluated prevalence of FCV antibodies in healthy adult cats and their antibody response to FCV vaccination.

Cats >1 year (n = 111) that had not received vaccinations within 12 months received a vaccine containing inactivated FCV antigen strains 431 and G1.

On day 0, 7, 28, FCV antibodies were determined in leftover samples by virus neutralization (VN) using isolate KS20, and by p66 antigen ELISA.

Factors associated with presence of antibodies and response to vaccination were determined by uni- and multivariate statistical analysis.

Pre-vaccination antibodies were detected in 62.7% (69/111) of cats (95%CI: 52.9-70.1) by VN and 77.2% (71/92; 95%CI: 67.5-84.6) by ELISA.

A ≥ 4-fold titre increase after vaccination was observed in 13.6% (15/110; 95%CI: 8.3-21.4) by VN and 33.7% (28/83; 95%CI: 24.5-44.5) by ELISA.

Cats ≥2 years were more likely to have pre-vaccination VN antibodies than cats <2 years (OR: = 7.194; P = 0.021). Presence of VN antibodies was also correlated with the cats´ vaccination status (OR: 3.472; P = 0.014).

Presence of pre-vaccination ELISA antibodies was associated with time since last vaccination (OR: 5.672; P = 0.043).

Outdoor cats were more likely to have a ≥ 4-fold titre increase in ELISA (OR: 5.556; P = 0.005).

Many cats have pre-vaccination FCV antibodies even if vaccination was performed >1 year ago.

Prevalence of antibodies depends on previous vaccinations and seems to increase with age.

A ≥ 4-fold titre increase after vaccination was rarely observed and influenced by the cat´s lifestyle.

Disclosures

Disclosures to report.

Hervé Poulet is Global Head of Companion Animals and Equine Biologicals R&D at Boehringer Ingelheim who provided antibody testing by ELISA. Boehringer Ingelheim played no role in the collection and interpretation of data, or in the decision to submit the manuscript for publication. Katrin Hartmann has given talks for MSD, Merial, Boehringer Ingelheim, Idexx. She participated in research funded by or using products from MSD, Merial, Boehringer, Zoetis, Megacor, Biogal, Scil. Michèle Bergmann has given talks for Merial. She participated in research funded by or using products from MSD, Merial, Boehringer, Zoetis, Megacor, Biogal, Scil. There is no commercial conflict of interest as the information generated here is solely for scientific dissemination. The authors declare that they have no competing interests.

ISCAID-O-5

Correlation of feline coronavirus shedding in faeces with serum coronavirus antibody titre

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Feline coronavirus (FCoV) infection is very common in multi-cat households. It has been proposed that cats with higher antibody titres are more likely to shed FCoV in their faeces. Aim of the study was to determine a possible correlation between FCoV serum antibody titres and faecal FCoV shedding.

Four faecal samples from 72 cats originating from 18 German catteries were examined for FCoV by quantitative reverse transcriptase polymerase chain reaction (RT-PCR). Serum antibody titres were determined by immunofluorescence assay.

There was a weak positive correlation between height of antibody titre and mean faecal virus load (Spearman r = 0.3394; P = 0.0035). Antibody titres were significantly higher if cats shed FCoV more frequently (Kruskal Wallis test P = 0.0042). Twenty-two cats were RT-PCR-negative in all four faecal samples. Those cats had significantly lower antibody titres than cats shedding continuously (in all four samples) (Dunn's test; P < 0.05). When analysing FCoV-shedding cats (shedding at least once), cats that were FCoV RT-PCR-positive continuously in all four samples had significantly higher antibody titres (Mann-Whitney U test P = 0.0026) and significantly higher mean faecal virus loads (Mann-Whitney U test P = 0.0383) than cats that were FCoV RT-PCR-positive in only one, two, or three samples. Eight cats had no detectable antibodies but were shedding FCoV.

Height of antibody titre was correlated to faecal virus load. Chronic FCoV shedders had higher antibody titres and shed more virus. This knowledge can be of importance for the management of FCoV infection in multi-cat environments. However, measurement of serum antibodies cannot replace faecal RT-PCR.

Disclosures

Disclosures to report.

Dr. Christian Leutenegger was the Head of Molecular Diagnostics at IDEXX Laboratories, Inc. Dr. Nikola Pantchev is employed at IDEXX Laboratories, Ludwigsburg. This laboratory offers the FCoV real-time RT-PCR on a commercial basis and performed the RT-PCR-testing in this study.

ISCAID-O-6

A retrospective multi-centre study on treatment and outcome in disseminated aspergillosis in 41 dogs

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Disseminated aspergillosis (DA) in dogs has a guarded prognosis. The aim of this study is to describe DA treatment regimens and their association with post diagnosis survival times.

This retrospective study evaluated dogs diagnosed with DA from 13 veterinary referral centres (private and University) from around Australia over a 10-year period (January 2007 to June 2017). Inclusion criteria included consistent diagnostic findings and a positive culture for *Aspergillus* from a sterile site or a positive serum galactomannan assay.

The data were analysed using survival analysis. The outcome of interest was the length of time (in days) between the date of diagnosis and the date of death or euthanasia due to DA. Factors influencing survival time post diagnosis were quantified using a Cox proportional hazards regression model. Here the data were organised into counting process format which allowed us to quantify the effect of each anti-fungal agent on survival time.

A total of 41 dogs were included in the study. The most common breed was German shepherd dogs (n = 24, 59%). The most common organism cultured was *A. terreus* (n = 24 out of 31 positive cultures, 77%).

Treatment was started in 27 dogs, whereas 9 dogs received no specific treatment, and 5 dogs were lost to follow up. Twenty-four dogs (89%) were treated with itraconazole as first-line treatment (single agent in 12 dogs). A total of nine dogs (33%) were treated concurrently with terbinafine. Median survival time post diagnosis was 273 days. Age at diagnosis had no significant effect on survival time. The daily hazard of death from DA for dogs with an elevated serum creatinine concentration at diagnosis was 18 (95% CI 3.8 to 83) times that of dogs with normal serum creatinine concentration. The daily hazard of death from DA for dogs treated with itraconazole was 5.7 (95% CI 1.7 to 19) times that of dogs that were treated with other anti-fungal treatment. The daily hazard of death from DA for dogs treated with terbinafine was 0.21 (95% CI 0.05 to 0.97) times that of dogs that were not treated with terbinafine.

Consistent with previous studies, we found that most DA cases were German Shepherds. *A. terreus* was the most commonly isolated organism. Although itraconazole has previously been the drug of choice to treat DA, our findings show that combination therapy with terbinafine or newer anti-fungal treatment improves survival times. Serum creatinine is a useful prognostic indicator of survival time.

Disclosures

No disclosures to report.

ISCAID-O-7

Canine Trichuris vulpis infection: a retrospective study of 45 cases

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Trichuris vulpis (*Tv*) is a parasite of canids large intestine with a worldwide distribution. Despite its well-known epidemiology, its pathogenic impact in dogs remains controversial.

The aim of the current retrospective study was to describe clinical and biological signs and treatment response in dogs naturally infected with *Tv*.

Our medical database was searched for cases with *Tv* eggs identified on fecal analysis between 2002 and 2018. Dogs were classified as mono-infected by Tv (G1) or poly-infected (*Tv* and other parasitic species; G2) and the intensity of *Tv* fecal excretion (FE) was quantified (number of *Tv* eggs/5 g of feces). Dogs diagnosed with other comorbidities potentially contributing to systemic or digestive signs, or to biological abnormalities such as anemia or hypoalbuminemia, were excluded. Clinical signs, biological abnormalities and course of the disease were recorded and compared between groups. Associations between these variables and the FE were statistically evaluated and the level of significance was set at $P < 0.05$.

Forty-nine dogs were positive for *Tv* on fecal analysis and 45 were included in the study (25 dogs in G1 and 20 dogs in G2). In G2, concurrent isolated parasites were *Toxocara canis* (47,8%), Ankylostomatidae (43,5%), *Capillaria* spp (13%), Tænidae (4,3%), *Isospora* spp (34,8%) and *Giardia duodenalis* (21,7%). The median age of infected dogs was 4 years. Overall, clinical signs included diarrhea (49%), weight loss (38%), hematochezia (29%), inappetence (24%), vomiting (18%) and polyphagia (13%). Intussusception was diagnosed in 2 dogs, only in G1. Two dogs showed lethargy as the only clinical sign, likely due to a moderate to severe anemia. Digestive signs were acute (less than 10 days) in 34% of dogs and chronic in the remaining dogs. Biological abnormalities included anemia (23%), eosinophilia (30%), hypoalbuminemia (60%) and Na/K ratio < 24 with normal ACTH-stimulation test (12%). Four percent of dogs had neither clinical nor biological signs. The only difference between G1 and G2 was that weight loss was more frequent in G2 ($P < 0.05$). No significant association was found between clinical signs or biological abnormalities and FE. However, a Na/K ratio < 24 with normal ACTH-stimulation test was only found among dogs with massive FE. Complete recovery after antiparasitic treatment was obtained in 94% of dogs with available follow-up (n = 18). One dog died of intussusception.

Our results suggest that *Tv* is pathogenic in dogs, leading to both acute and chronic digestive clinical signs, hypoalbuminemia, anemia and electrolytes disorders.

Disclosures

No disclosures to report.

ISCAID-O-8

Patterns of antimicrobial use for selected canine diseases in Switzerland in 2016

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Antimicrobial resistance is an increasing threat for human and animal health. Both over- and misuse of antimicrobials can foster resistance. The objective of this retrospective study was to investigate patterns of antimicrobial use for selected canine diseases in Switzerland in 2016.

Antimicrobial prescriptions for respiratory and urinary tract infections (UTI) and acute diarrhea from two Swiss university hospitals and 14 private practices during 2016 were reviewed. Classes of antimicrobials, dosage and treatment duration were assessed. A justification score (JS) was applied, where sufficient clinical information was available, to define the appropriateness of antimicrobial therapy based on current guidelines (1 = appropriate, 2 = incorrect dosage and/or duration, 3 = inappropriate antimicrobial choice, 4 = overall wrong treatment decision).

Of 274 dogs with proven or suspected upper or lower respiratory tract infections, 171 (62%) were treated with antimicrobials of the following classes: potentiated aminopenicillins (68.4%), fluoroquinolones (15.8%), non-potentiated (NP) aminopenicillins (20.4%), tetracyclines (22.2%), first-generation cephalosporins (2%), third-generation cephalosporins, lincosamides, macrolides and nitroimidazoles (0.8% each). In 48.2% (132/274), therapeutic decisions regarding antimicrobial therapy were judged appropriate (JS-1), in 23.3% (64/274) inappropriate (JS-2 n = 4; JS-3 n = 26).

Of 245 dogs with proven or suspected UTI, 215 (87.8%) received antimicrobials. In only 36.3% of these, a bacterial etiology was confirmed via culture and/or sediment examination. Antimicrobial classes used were potentiated aminopenicillins (61%), fluoroquinolones (22%), NP-aminopenicillins (11%), first-generation cephalosporins (5%), third-generation cephalosporins (1%), lincosamides (0.8%), amphenicols and potentiated sulfonamides (0.4% each). Antimicrobial susceptibility testing was performed in 73/85 (86%) of the cases where bacterial culture was performed. In 20% (49/245), antimicrobial therapy was judged appropriate (JS-1), in 16% (39/245) inappropriate (JS-2n = 5; JS-3 n = 30).

Antimicrobials were prescribed in 89.5% (247/276) of the dogs with acute diarrhea. Classes used were nitroimidazoles (72.1%), potentiated aminopenicillins (29.1%), fluoroquinolones (7.6%), NP-aminopenicillins (3.6%), tetracyclines (1%) and third-generation cephalosporins (1%). Antimicrobial therapy was significantly associated with the presence of bloody diarrhea ($P < 0.001$). It complied in 43.5% (120/276; JS-1) with current guidelines restricting therapy to the use of aminopenicillins in suspected sepsis.

The results of this study show that antimicrobial use commonly does not comply with current treatment guidelines in dogs with respiratory or urinary tract infections and acute diarrhea. Consequently, there is an urgent need for antimicrobial stewardship initiatives.To support the prudent use of antimicrobials in animals, antibioticscout.ch, a comprehensive online tool based on current guidelines was launched in December 2016. The impact of this tool on veterinary prescribing habits will be assessed in future studies.

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ISCAID-O-9

Comparison of antimicrobial prescription in selected diseases in cats in Switzerland between 2016 and 2018: a trend towards more prudent antimicrobial use

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Overuse of antibiotics is a common problem in veterinary medicine contributing to the development of antimicrobial resistance. To foster prudent antimicrobial use by veterinarians, a freely accessible online tool containing Swiss consensus guidelines for prudent antimicrobial use was launched in December 2016 (http://www.antibioticscout.ch). The aim of this study was to compare antimicrobial prescription in cats in Switzerland before and after the introduction of antibioticscout.ch.

Cats presented to one university clinic and eight private practices/clinics in 2018 with acute upper respiratory tract disease (aURTD), feline lower urinary tract disease (FLUTD) and abscesses (only in private practices/clinics) were included. Signalment, clinical symptoms, diagnostic work-up, diagnosis and antimicrobial therapy were assessed and the data compared to published data from cases belonging to the same disease categories presented to the same practice/clinic in 2016 (Schmitt et al., 2019). A justification score was applied to evaluate accordance of prescription with the guidelines (JS1 = complete accordance, JS2 = different dosage/duration, JS3 = different antimicrobial choice, JS4 = antimicrobial use/non-use in disagreement with the guidelines, JS5 = prudent use not assessable).

Data from 485 cats (aURTD, n = 152; FLUTD, n = 205; abscesses, n = 128) presented in 2018 were compared with data from 2016 (469 cases). The frequency of antimicrobial prescription decreased from 72% (2016) to 61% (2018; $P < 0.001$); this decline was found at the university clinic (68% to 48%, $P = 0.003$) as well as in private practices/clinics (73% to 64%, $P = 0.010$) and in each disease category (aURTD, 72% to 61% $P = 0.030$; FLUTD, 56% to 44%, $P = 0.012$;

abscesses, 97% to 88%, $P = 0.008$). More specifically, the prescription of critically important 3rd generation cephalosporins decreased (2016, 18%; 2018, 13%; $P = 0.017$). Urine analyses were more frequently performed in private practice/clinic in cats with FLUTD (2016, 26%; 2018, 48%; $P < 0.001$). Despite this, the number of prescriptions in accordance with the guidelines (JS1) did not significantly increase from 2016 to 2018 (aURTD, 25% and 31%; FLUTD, 21% and 28%; abscesses, 18% for both years).

The present study reports a reduction in antimicrobial prescription and use of 3rd generation cephalosporins in cats in Switzerland which coincided with antibiotic stewardship activities including the launch of antibioticscout.ch. However, the adherence to consensus guidelines was still poor. An oral preparation of a non-potentiated aminopenicillin for cats was not available in 2018 in Switzerland and could have hampered compliance with the guidelines. Thus, although the present study indicates a trend towards less frequent prescription of antimicrobials in cats, further efforts are necessary to promote antimicrobial stewardship in small animal medicine.

Disclosures

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ISCAID-O-10

Evaluation of hand hygiene compliance in small animal clinics and practices in Switzerland using the CleanHands application

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Small animal veterinarians are commonly faced with contagious or zoonotic diseases and infections with multidrug resistant organisms. Hand hygiene (HH) is considered one of the most important infection control measures in healthcare. Recent studies suggest that HH compliance in small animal veterinary institutions is poor, but comprehensive data from small animal clinics and practices in Europe is lacking.

The present study evaluated HH compliance according to the World Health Organization (WHO) five moments for HH in small animal clinics and practices in Switzerland using the CleanHands application.

Three small animal clinics (A-C), one medium-sized (D) and one small primary opinion practice (D) were included. A minimum of 500 observations in clinics A-C (100 observations per study area: pre-operative preparation area, intensive care unit [ICU], animal housing area, examination area, consultation area) and 130 observations in practices D-E (consultation area and animal housing area) were obtained. The WHO five moments for HH included: before touching a patient, before clean/aseptic procedures, after body fluid exposure/risk, after touching a patient and after touching patient surroundings. Hand disinfection with alcohol-based handrubs or hand washing with water and soap were considered successful HH procedures. Frequency of HH [95% confidence intervals] based on Jeffreys approach were estimated using the software R version 3.4.4.

A total of 1772 observations revealed an overall HH compliance across all institutions of 32% [29-34], ranging from 26-47%. Highest compliance was observed in the consultation area (45% [40-49]), followed by ICU (32% [27-37]), examination area (29% [24-34]), animal housing area (26% [22-31]) and pre-operative preparation area (20% [15-24]). HH was most commonly performed after contact to body fluids (42% [36-47]) and after patient contact (37% [34-41]), and was least common prior to clean/aseptic procedures (12% [8-15]). Veterinarians showed a higher adherence to HH (37% [34-40]) than veterinary assistants (25% [22-29]).

The study indicates an overall poor adherence to HH in small animal clinics and practices in Switzerland. HH was remarkably poor in critical areas such as the pre-operative preparation area and before clean/aseptic procedures. In contrast to observations in human hospitals, adherence to HH was lower in nursing staff than in doctors. In conclusion, the study highlights the need to promote HH in small animal medicine, with special emphasis on training of nursing staff and personnel in pre-operative preparation areas.

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ISCAID-O-11

Evaluation of infection prevention and control standards and carriage of multidrug-resistant organisms in working staff in small animal clinics and practices in Switzerland

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Intensive medical care of companion animals and their close contact to people pose a risk for the selection and zoonotic transmission of multidrug-resistant organisms (MDROs). Infection prevention and control (IPC) concepts are key measures to reduce the spread of MDROs, but data on IPC standards in small animal clinics and practices is sparse. The goals of the study were to assess IPC standards, environmental MDRO contamination and prevalence of MDRO carriage in veterinary personnel in small animal clinics/practices in Switzerland.

Three large clinics (A-C), two medium-sized clinics (D-E) and two primary opinion practices (F-G) were included. Structured one-day IPC audits were performed and environmental samples collected from high-touch surfaces. Nasal and faecal MDRO carriage in veterinary staff (institutions A-C, G) was assessed by providing kits for sampling. The samples were analysed for methicillin-resistant (MR) *Staphylococcus aureus* (MRSA), MR *S. pseudintermedius* (MRSP), MR coagulase-negative staphylococci

(MRCoNS), MR *Macrococcus* spp., colistin-resistant *Enterobacteriaceae*, and extended-spectrum beta-lactamase (ESBL)- and carbapenemase-producing *Enterobacteriaceae* (CPE) using enrichment and selective procedures. Species identification was performed by MALDI-TOF MS analysis. Antibiotic resistance genes were identified by PCR and sequencing. Genetic relatedness was assessed by rep-PCR (for *Enterobacteriaceae*) and multilocus sequence typing (for MRSP).

Frequency of MDRO detection on high-touch surfaces in clinics/practices A-G was 3%, 27%, 28%, 0%, 26%, 4% and 6%, respectively. In clinics with high environmental contamination, CP-*E. coli* (bla_{OXA-48}, $bla_{OXA-181}$), CP-*K. pneumoniae* (bla_{OXA-48}) and MRSP (*mecA*, ST551)(clinic B), MRCoNS (*mecA*)(clinic C), and *Macrococcus* spp. (*mecB*, *mecD*) and MRCoNS (*mecA*)(clinic E) predominated. IPC audits revealed deficits in IPC organization, cleaning/disinfection, hand/personal hygiene, medication preparation and antimicrobial use guidelines in these clinics.

In veterinary personnel, faecal carriage of colistin-resistant *E. coli* (6%), ESBL-producing *E. coli* (6%) and CP-*E. coli* (1%), and nasal carriage of MRSA (7%), MRSP (1%) and MRCoNS (5%) were found. MDRO carriage was not associated with work place or profession of the person. The CP-*E. coli* ($bla_{OXA-181}$) from a staff member of clinic B was related to environmental isolates.

This study documents major deficits in IPC standards in small animal clinics in Switzerland and extensive, but variable, environmental contamination with MDRO. We report for the first time the detection of CPE in environmental samples in a small animal clinic and the faecal carriage of a related CPE isolate in a staff member. The present study highlights the potential of small animal clinics to spread MDRO and the need to promote IPC concepts in these institutions.

Disclosures

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ISCAID-O-12

Prevalence, acquisition and persistence of rectal and naso—/oropharyngeal carriage of multidrug-resistant organisms in dogs and cats presented to veterinary practices and their owners

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Multidrug-resistant organisms (MDRO) represent a significant threat to human and animal health. Extended-spectrum β-lactamase (ESBL), extended-spectrum cephalosporinase (3CG) and carbapenemase-producing (CP) *Enterobacteriaceae* are of particular importance as their easily transmitted resistance genes mediate resistance to many antimicrobial classes. The aims of this study were: to assess the prevalence of MDRO carriage in dogs and cats at presentation to veterinary clinics/practices, to monitor MDRO acquisition during hospitalization, to study MDRO transmission among pets and owners and to determine the duration of carriage.

For this prospective, longitudinal, observational study, rectal and nasal/oropharyngeal swabs were collected from 88 cats and 183 dogs presented to 5 veterinary care facilities and 187/271 animals were resampled at discharge. Participating owners sampled themselves (nasal swab/stool sample). Carriers were resampled for up to 8 months. Nasal/oropharyngeal swabs were analysed for the presence of methicillin-resistant (MR) *Staphylococcus aureus* (MRSA), MR *S. pseudintermedius* (MRSP), MR coagulase-negative staphylococci (MRCoNS), MR *Macrococcus* spp. and rectal swabs/stool for ESBL-producing and CP *Enterobacteriaceae*. After enrichment, isolates were selected on CHROMID ESBL, OXA-48 and CARBA plates or MRSA selective agar. Isolates were identified via MALDI-TOF MS. Antibiotic susceptibility was tested by measurement of MICs and clonality determined using rep-PCR, ERIC-PCR and whole genome sequencing (WGS).

The overall admission prevalence of MDRO carriage was 15.5% (95%CI 11.4-20.4) with ESBL-producing *E.coli* (5.5%) and MRCoNS (8.5%) predominating. The overall discharge prevalence was 32.6% (95%CI 26-39.8; range 17.2-42.7%). In the institution with the highest discharge prevalence predominant hospital-acquired isolates were: ESBL-producing *K. pneumoniae* (13.7%) and ESBL-producing *E.coli* (16.7%). *E.coli* isolates commonly displayed CP-encoding genes ($bla_{oxa-181}$, bla_{oxa-48}, bla_{NDM-5}) and showed clonality (ST410, bla_{oxa181}), suggesting transmission from a common source rather than *de novo* selection. Persistence of ESBL-producing or CP *E.coli* or *K. pneumoniae* was shown in 7/34 MDRO positive animals for up to 138 days. Resistant bacteria were isolated from 10/46 owners (6/10 ESBL-producing *E.coli*; 3/10 MRCoNS; 2/10 MRSA); carriage persisted in one owner for 68 days. Transfer of ESBL-producing *E.coli* between owner and dog was suspected in one case. Further analysis is ongoing to determine the relatedness of the isolates.

In this cohort, the acquisition rate of 3CG-resistant and/or CP *Enterobacteriaceae* was high, but varied between institutions. MDRO carriage was observed in 22% of owners and carriage persisted for several months. These findings show that veterinary hospitals play a significant role in the selection and transmission of MDRO amongst veterinary patients, including bacteria with very problematic resistance profiles.

Disclosures

Disclosures to report.

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SCH-O-1

Lactulose drives a reversible reduction and qualitative modulation of the faecal microbiota diversity in healthy dogs

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Hepatic encephalopathy (HE) is a syndrome of neurologic dysfunction and an important contributor to patient morbidity in dogs with liver

diseases. The prebiotic lactulose, a nonabsorbable synthetic disaccharide, is a frequently employed treatment for canine HE, yet with incompletely understood mechanisms of action. In humans, HE is linked with dysbiosis, which has been associated with worsened morbidity and mortality. The impact of lactulose in ameliorating this HE-associated dysbiosis or general modulation of the intestinal microbiota is controversial. It is unknown if dysbiosis is present in canine HE and the influence of lactulose on the intestinal microbiota has also not been assessed in dogs. The aims of this study were therefore to examine the changes in faecal microbiota composition before, during and after lactulose treatment in healthy dogs.

A total of 21 healthy privately owned dogs were enrolled in a prospective cohort study (12 females, 9 males, median age 5 years [range 2-10]) with 18 completing the study fully. Faecal samples were collected weekly, while dogs were either on their usual diet (week 1), followed by a standardised commercial diet (weeks 2-9), with added oral lactulose (0.5 mL/kg every 12 hours) in weeks 6-7. Faecal bacterial DNA extraction was followed by PCR amplification of the V4 region of the 16S rRNA gene. Illumina standard 16S library prep and sequencing was performed on the MiSeq platform and data analysed using the QIIME2™ pipeline.

After 2 weeks of lactulose treatment (week 7) significantly lower faecal microbiota richness/diversity was observed based on the alpha diversity metrics: observed operational taxonomic units, Shannon/ Chao1 indexes and Pielou's evenness. Beta diversity, based on UniFrac distances, was also significantly different in week 7 compared to weeks 1, 5 and 9. At the bacterial phylum and family levels, week 7 was associated with a significant increase of Firmicutes and Actinobacteria (*Veillonellaceae* and *Bifidobacteriaceae*), and decrease of Bacteroidetes and Fusobacteria (*Bacteroidaceae*, *Fusobacteriaceae*, *Ruminococcaceae* and *Alcaligenaceae*), when compared to weeks 5 and 9. Finally, an extrapolated cirrhosis dysbiosis ratio (CDR) was calculated, for which lower values are associated with dysbiosis and linked with worse outcomes in humans. CDR was increased in week 7 compared to weeks 1, 5 and 9.

In conclusion, lactulose induced a reversible qualitative and quantitative change of the faecal microbiota in healthy dogs, possibly explaining its potential benefit in the management of HE.

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SCH-O-2

Prevalence of bactibilia in apparently healthy dogs

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Bacterial cholecystitis is a debilitating disease in dogs. The presence of bacteria in bile in ill dogs would be significant if bile was considered sterile; however, the prevalence of bactibilia in healthy dogs is unknown. The primary purpose of this study was to determine the prevalence of bactibilia in healthy dogs. Secondary aims were to determine if differences between bactibillic and non-bactibillic healthy dogs occur with regards to serum liver enzymes activities; and liver and gallbladder histopathology.

Fifty-five healthy, abandoned dogs euthanased for non-medical reasons were included in this cross-sectional, prospective study. Dogs were deemed healthy based on clinical and necropsy examinations. Whole blood, bile, gallbladder wall and liver samples were collected aseptically from all dogs within 30 minutes of euthanasia and submitted for bacterial culture, cytological, biochemical (alkaline phosphatase (ALP), alanine aminotransferase (ALT) and gamma-glutamyl transferase (GGT)) and histopathological analyses. Agreement between cytology and culture was assessed with Cohen κ analysis and analysis of variance of serum liver enzymes activities between dogs with bactibilia and without was performed using the Mann-Whitney test.

The prevalence of bactibilia was 16.36% (9/55), with 10.91% (6/55) of dogs diagnosed on cytology and 10.91% (6/55) on bile culture. There was poor agreement between bile cytology and culture (0.439, Cohens kappa; $P = 0.001$). No significant differences in liver enzyme concentrations were found between bactibilic and non-bactibilic dogs. No significant hepatobiliary histopathological abnormalities were present in bactibilic dogs.

The prevalence of bactibilia in asymptomatic dogs was 16.36%, with no significant elevation in liver enzymes or histopathological changes.

Disclosures

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Co-author F. Kettner has a financial relationship with and indirectly benefits from the laboratory service (Vetdiagnostix) used in this study to perform the bacterial culture, cytological, biochemical and histopathological analyses discussed in the abstract. He has indirect shareholding in Vetdiagnostix and is involved in managing the Vetdiagnostix Cape Town branch.

SCH-O-3

Hyaluronic acid as a liver function test to asses extrahepatic portosystemic shunt closure in dogs after surgical attenuation

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Liver function tests do not always normalize after successful surgical attenuation of portosystemic shunts (PSS). Currently the gold

standard to demonstrate absence of portosystemic shunting is portal scintigraphy. Serum hyaluronic acid concentrations (sHA) in dogs with PSS are increased compared to those in healthy dogs. A preliminary study reported that sHA decreased 2 weeks after surgical attenuation of extrahepatic PSS (EHPSS).

The aims of the current study were: 1/ to serially evaluate sHA in dogs with surgically attenuated EHPSS and to determine differences in sHA in dogs with closed versus open (persistent or multiple acquired) PSS; 2/ to compare sHA in patients with EHPSS versus other liver diseases.

Twenty dogs with surgically treated EHPSS and 10 dogs with other liver diseases were included. Dogs with EHPSS had a blood sample taken at diagnosis, 1, 3 and 6 months postoperatively. At the 3-month control visit a transsplenic portal scintigraphy was performed to determine shunt closure status. Dogs with other liver diseases were only sampled at a single time point and comprised of: Maltese dogs with moderately increased postprandial bile acids and no liver disease based on imaging (n = 3), dogs with histologically confirmed portal vein hypoplasia (n = 4) or histologically confirmed chronic hepatitis (n = 3). All samples were analysed in batch using a commercially available ELISA kit (Hyaluronan Quantikine, R&D systems, Minneapolis).

At EHPSS diagnosis, median sHA was 337.20 ng/mL (158.02-790.66 ng/mL). After successful surgery (closed PSS), sHA dropped to 36.62 ng/mL (13.51-92.24 ng/mL) whereas in dogs with persistent portosystemic shunting, sHA remained higher (median 135.70 ng/mL; 56.44-312.04 ng/mL). Kruskal-Wallis tests revealed a significant difference between sHA in dogs with closed versus open EHPSS ($P = 0.008$, $P = 0.005$ and $P = 0.025$ at 1, 3, and 6 months postoperatively, respectively). The median sHA of dogs with other liver diseases was 119.64 ng/mL (48.44-160.00 ng/mL), which was significantly lower compared to dogs at the moment of EHPSS diagnosis ($P = 0.009$).

In dogs with EHPSS, sHA seems to be a promising non-invasive biomarker to determine EHPSS closure after surgical attenuation. In addition, it might also be valuable to differentiate dogs with EHPSS from dogs with other liver diseases.

Disclosures

No disclosures to report.

ESVONC-O-1

BRAF-mutation in carcinomas of various sites in the canine urinary tract

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The oncogenic mutation of the BRAF-gene is well described in canine transitional cell carcinomas (TCC) of the urinary bladder and urethra (Mochizuki et al. 2015, Aupperle-Lellbach et al. 2018, 2019). This study investigates the prevalence of BRAF-mutation in carcinomas distributed over the whole canine urinary tract.

Dogs included in this study were 23 terriers (7 Scottish, 7 Jack Russel, 3 West Highland White, 2 Airedale, 3 Fox, 2 Yorkshire, 1 Welsh, 1 Irish Soft Coated Wheaten) and 108 dogs of other breeds (eg, 40 mongrels, 6 Beagle, 5 Bearnaise Mountain Dogs, 4 Cocker Spaniel) in the age of median 11 years. Formalin fixed samples from carcinomas of the urinary tract (renal tubular carcinoma (RCa, n = 10), TCC of renal pelvis (RPCa, n = 6), TCC of urinary bladder (ubTCC, n = 78), TCC in urethra (uTCC, n = 28), and TCC simultaneously in urethra and urinary bladder (sTCC, n = 9) were histopathologically diagnosed. DNA-isolation was performed by using a QIAamp DNA FFPE Tissue Kit. Exon 15 of chromosome 16 was examined for the presence of BRAF-mutation c.1799 T > A by TaqMan SNP assay. Statistical analyses were performed using GraphPad Prism version 7.03.

Histological diagnosis showed tubulopapilliform RC and solid high-malignant PRCa in all cases. Most TCC were high-malignant (55/78 ubTC, 28/28 uTC, 8/9 sTCC). BRAF-mutation was detected in 0/10 RCa, 1/6 RPCa, 36/78 ubTCC, 16/28 uTCC, 6/9 sTCC. It was significantly ($P \leq 0.05$) more often found in neoplasms of the lower urinary tract than in the kidney. However, there was no significant difference in the prevalence of BRAF-mutation between the different sites of TCC in urinary bladder and/or urethra. Statistical correlation of histological degree of TCC in LUT and BRAF-mutation was not obvious. BRAF-mutation was identified significantly more frequent in ubTcc of terriers (20/25, 75%) than in other breeds (21/57, 35%) ($P < 0.005$). In uTCC the breed differences in BRAF-mutation were not significant, probably due to the small number of terrier cases (2/28 dogs with BRAF mutation).

In conclusion, renal carcinomas are mostly not caused by BRAF-mutation. Thus, tubular and pelvic carcinomas of the kidney cannot be detected by BRAF-mutation analysis of cells excreted within urine. In contrast, BRAF-mutation is often involved in pathogenesis of TCC in urinary bladder and/or urethra, and can be used as an excellent diagnostic tool with enormous specify for malignancy and site of carcinomas in lower urinary tract.

Disclosures

Disclosures to report.

The authors H. Aupperle-Lellbach, J. Grassinger, L. Kempker and H. Erhard are employed at LABOKLIN GmbH & Co KG, who provides the BRAF-test.

ESVONC-O-2

Re-irradiation is a valuable treatment option for dogs and cats with cancer after failing first line therapy

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Re-irradiation is getting a more recognized treatment option in patients with recurrent neoplastic disease. However, information about efficacy and risk for chronic side effects is limited in veterinary

medicine. The aim of this study was to evaluate outcome and reported late side effects in dogs and cats receiving re-irradiation because of progressive tumor recurrence. Small animal cancer patients treated with re-irradiation between 2006-2017 were included into this retrospective study. Patient characteristics, tumor type and localization, pretreatment, time interval between initial radiotherapy and re-irradiation, radiation protocols, late side effects, cause of death and survival times were analyzed. Forty-nine patients (27 dogs, 22 cats) were included into this study and the majority of them suffered from head- and neck tumors (61%). Re-irradiation protocols were - with one exception -palliative and five animals received a second course of re-irradiation. Fifteen patients were initially radiated definitively and 34 animals with palliative intent. Mean time interval between initial radiotherapy and first re-irradiation was 264 days and between first and second re-irradiation 207 days. Mean total biologic-effective-dose for late responding tissue (BED_3) was 126 Gy for initial radiotherapy and re-irradiation. Late side effects were most commonly observed in skin/hair but always scored as mild (score 1, VRTOG). Chronic ocular side effects were reported in seven patients (score 1-3). Median tumor-specific survival time of all patients was 529 days. In conclusion, this study demonstrate that palliative re-irradiation can be offered as treatment option for incurable recurrent tumors. Late side effects were mostly mild and not life-threatening.

Disclosures

No disclosures to report.

ESVONC-O-3

Impact of Repeated Cycles of EGF Bispecific Angiotoxin (eBAT) Administered at a Reduced Interval from Doxorubicin Chemotherapy on Tolerability and Survival of Dogs with Splenic Hemangiosarcoma

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Targeted toxins are promising agents designed to target receptors that are uniquely or highly expressed by cancer cells, improving tumor specificity with reduced adverse events (AEs). As their name implies, bispecific ligand-targeted toxins have dual targeting ability that confers greater binding affinity and killing ability compared to monospecific counterparts. eBAT is a bispecific epidermal growth factor (EGF) angiotoxin developed as a second generation biologic drug to specifically target tumor cells and associated vascular and inflammatory stroma for sarcoma therapy. It consists of human EGF, targeting the EGF receptor (EGFR), human amino terminal transferase (ATF) of urokinase, targeting the urokinase plasminogen activator receptor (uPAR), and a genetically modified, de-immunized *Pseudomonas* exotoxin, leading to inhibition of protein synthesis. We previously reported that eBAT was safe and improved overall survival for dogs with splenic hemangiosarcoma (HSA) in the minimal residual disease setting when added to standard of care (SOC) therapy in a single cycle of three treatments. Studies with *Pseudomonas* exotoxin in humans have suggested that repeat cycles of administration may prolong remissions whereas the optimal timing between administration of targeted toxins and chemotherapy is unclear. The SRCBST-2 (sarcoma bispecific toxin trial-2) study described herein was undertaken to prospectively determine if multiple cycles of eBAT at the biologically active dose (50 ug/kg) given intravenously, concomitant with a reduced interval between administration of the targeted toxin and doxorubicin chemotherapy would be well-tolerated and further improve outcomes of dogs with splenic HSA. Eligibility was expanded to dogs with stage-3 HSA, provided that macroscopic lesions could be surgically excised. Treatment included three planned cycles of eBAT, each administered on a Monday/Wednesday/Friday schedule starting upon recovery from splenectomy, and continuing one week prior to the 1st, 2nd, and 5th doxorubicin events. The interval between first eBAT (given on day 1) and first doxorubicin was reduced compared to the previous trial using a single cycle of eBAT (with doxorubicin starting on day 8 instead of day 21). Twenty-five dogs were enrolled; six experienced acute hypotension with two requiring hospitalization. Self-limiting elevation of ALT was observed in one dog. A survival benefit was not seen in this study: overall survival was comparable to that of a contemporary control group of dogs with stages 1-3 hemangiosarcoma treated with SOC alone. Repeated dosing cycles of eBAT led to greater incidence and severity of AEs and reduced efficacy as compared to a single cycle of eBAT with delayed commencement of chemotherapy.

Disclosures

Disclosures to report.

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ESVONC-O-4

Humoral hypercalcemia of malignancy in canine lymphoma WHO types and its impact on survival

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Humoral hypercalcemia of malignancy (HHM) is a common paraneoplastic syndrome in canine lymphoma and has always been considered a negative prognostic factor. However, studies analyzing the role of HHM in WHO lymphoma types are lacking. The aim of this study was to evaluate the incidence of HHM among different lymphoma types and its prognostic impact on survival. In a retrospective study data of dogs diagnosed with untreated lymphoma between 2008 and 2019 were analyzed. Inclusion criteria were availability of WHO type and serum/plasma calcium concentration.

139 cases were included. HHM was present in 14/139 (10%) patients. Most notably only patients with T-lymphomas were affected. 9/14 (64%) suffered from peripheral T-cell lymphoma (PTLC) and 5/14 (36%) from T-lymphoblastic lymphoma (T-LBL). HHM occurred in 43% (9/21) of PTLC and 38% (5/13) of T-LBL cases. 86% of patients with HHM showed symptoms attributable to hypercalcemia, predominantly polyuria/polydipsia, neurological (somnolence, weakness), and gastrointestinal manifestation (inappetence). In dogs with HHM, median total and ionized calcium were 3.81 mmol/l (reference interval 2.4-3.0) and 1.8 mmol/l (reference interval 1.25-1.5), respectively. Under chemotherapy calcium concentration returned to normal in 93% of cases. No significant differences in progression free survival (150 vs. 60 days, $P = 0.29$) or lymphoma specific survival (170 vs. 135 days, $P = 0.42$) were observed between hypercalcemic and normocalcemic dogs of both subtypes. According to our results, HHM is not an unfavorable prognostic factor but it is associated with aggressive T-cell lymphoma types. Prospective studies assessing the role of vitamin D3, PTHrP and PTH in HHM are warranted.

Disclosures

No disclosures to report.

ESVONC-O-5

High risk mast cell tumours with favourable outcome in 16 young dogs

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Mast cell tumour (MCT) represents the most common canine skin neoplasia and typically affects adult-geriatric dogs (median age 9 years). MCTs are rarely reported in puppies and junior dogs, and therefore little information exists about their biologic behaviour and treatment requirements. The aim of this retrospective study was to describe clinical and histopathological features, proliferation markers, c-kit mutations and outcome of MCTs in dogs less than 1 year-old.

Sixteen dogs were included in the study: 13 with cutaneous MCT and 3 with subcutaneous MCT. The median age at first presentation and diagnosis was 7.6 months (range 2-11.9) and 9 months (range 2-36.1), respectively. There were 5 males (4 entire, 1 neutered) and 11 females (6 entire, 5 neutered) and the most common breeds were Labrador (5) and Golden Retriever (2). Of the thirteen cutaneous MCTs, 3 were grade II (Patnaik), 6 were grade II/low-grade (Patnaik/Kiupel), 2 were grade II/high-grade (Patnaik/Kiupel), and 1 was high-grade (Kiupel); 3had mitotic index >5/10HPFs(median mitotic index 7/10HPFs). Of the three subcutaneous MCTs, two had an infiltrative growth pattern and 1 had mitotic index of 10/10HPFs.

The regional lymph nodes were assessed in 13/16 cases by means of cytology or histopathology; metastases were identified in 4 dogs (2 cutaneous, 2 subcutaneous).Ki-67 was assessed in 10 cases and it was above the cut-off in 9 (8 cutaneous, 1 subcutaneous MCT).Of the 9 cases screened, a c-Kit mutation was identified in 6 (all cutaneous, exons 9,11,12). Patients underwent different treatment modalities: surgery +/− corticosteroids(8), surgery + chemotherapy (5) + radiotherapy (2) and radiotherapy/chemotherapy(1). Recurrence was identified in 3 cases after a median time of 632 days (range 15-730): one patient received only corticosteroids following recurrence (15 days after the initial surgery), achieving complete and durable clinical remission (1013 days); the other 2 dogs received surgery, which was followed by chemotherapy in one case.The median follow-up time for this cohort of dogs was 1115 days (range 282-2655). All patients were alive, and with no evidence of MCT at the end of the study period.

This study suggests that MCTs in puppies and junior dogs might have a more favourable outcome despite the presence of clinical, pathological or genetic characteristics that would predict an aggressive biological behaviour in adult-geriatric dogs. Based on this, prognostic factors might need to be stratified for life stages.

Disclosures

Disclosures to report.

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ESVONC-O-6

Diphenhydramine Does Not Reduce Infusion-Related Ventricular Arrhythmias in Dogs Treated with Doxorubicin

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Doxorubicin (DOX) is one of the most effective chemotherapeutics for canine high-grade lymphoma. Besides dose-dependent chronic cardiotoxicity, DOX can trigger acute cardiac arrhythmias during drug infusion. Diphenhydramine premedication is commonly used, as histamine release is a proposed mechanism for DOX-associated arrhythmogenesis. The study objectives were to evaluate the incidence and severity of DOX infusion-related cardiac arrhythmias in dogs with

high-grade lymphoma, and the effect of diphenhydramine premedication on arrhythmia number and severity during and after DOX infusion.

Dogs with cytologically/histopathologically confirmed high-grade lymphoma were screened with an echocardiogram and concurrent electrocardiogram for this randomized prospective cross-over study. Group-A received no premedication for DOX#1 and was premedicated with diphenhydramine for DOX#2; Group-B received diphenhydramine with DOX#1 and no premedication for DOX#2. For both visits, Holter monitor data was collected 1 hour before DOX and 3 hours post-administration and analyzed by Burdick Holter Analysis Software. Commercially available software (Prism7.0) was used for normality testing and paired-sample analysis with each individual acting as its own control.

Seventeen dogs were enrolled and 10 dogs [Group-A(6), Group-B(4)] completed the protocol. There was no statistical difference between groups A and B when evaluating total ventricular premature complex (VPC) numbers ($P = 0.34$), change of VPCs/hour ($P = 0.25$), total atrial premature complex (APC) numbers ($P = 0.5$), change of APCs/hour ($P = 0.06$), or arrhythmia severity score ($P > 0.99$).

This study demonstrates that in dogs with appropriate pretreatment cardiovascular screening, DOX infusion does not induce significant arrhythmias. Furthermore, these data suggest diphenhydramine may not alter arrhythmia number or severity in canine DOX recipients.

Disclosures

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ESVONC-O-7

Time to change from WHO staging to Ann-Arbor system in canine nodal diffuse large B-cell lymphoma?

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Ann Arbor system (AAS) remains the best anatomic staging of human diffuse large B-cell lymphoma (DLBCL). AAS divides patients into four stages based on localized disease (I), multiple nodal sites on one side (II), disseminated nodal disease on both sides of diaphragm (III), and in other extranodal sites (IV). In comparison to the veterinary WHO staging system (WHOS) AAS considers a splenic infiltration as nodal involvement, but not as a higher stage.

The aim of this retrospective study was to compare WHOS and AAS to predict treatment response and survival in 54 canine nodal DLBCL treated with CHOP between 2008-2019. Because of low number of stage I/II patients (n = 3) in both systems, these were excluded.

There were nine stage III, 28 stage IV, and 17 stage V cases in WHOS, and 23 stage III, and 31 stage IV cases in AAS. No association between WHOS and grade, substage, B-symptoms and treatment response were found. Higher AAS stage was associated with substage B ($P = 0.03$) and B-symptoms ($P = 0.002$) and negatively with treatment response ($P = 0.001$). Higher AAS decreased progression free survival (PFS) (116 vs. 332 days, $P = 0.001$) and lymphoma specific survival (LSS) (180 vs. 489 days, $P = 0.001$). Higher WHOS showed a tendency toward shorter PFS (135 vs. 180 vs. 380 days, $P = 0.36$) and LSS (194 vs. 250 vs. 396 days, $P = 0.43$). In conclusion, AAS could predict more accurately prognosis in canine nodal DLBCL. Prospective studies assessing AAS in larger cohort of patients with standardized staging and in other lymphoma types are warranted.

Disclosures

No disclosures to report.

ESVONC-O-8

Accuracy of PET for Detection of Lymph Node Metastasis in Canine Oral Malignant Melanoma

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Positron emission tomography (PET) is commonly used for lymph node (LN) metastasis detection in human medicine. Studies report a range of accuracy depending on tumor type and grading system. Species and tumor-specific studies are needed to define the role of PET in staging veterinary oncologic patients. The aim of this study was to evaluate the accuracy of PET for metastasis detection in canine oral malignant melanoma (OMM).

Client-owned dogs with cytologically/histologically diagnosed OMM were eligible for this prospective study. A PET scan of the head/neck using ^{18}Fluorine-fluorodeoxyglucose (^{18}F-FDG) was performed followed by computed tomography. Bilateral mandibular lymphadenectomy was performed for histopathologic assessment. Scans were evaluated by two independent observers. First, observers were blinded to primary tumor laterality and graded subjectively comparing ^{18}F-FDG uptake to background. Subsequently, observers were unblinded to primary tumor information and utilized standard uptake value (SUV) quantification for evaluation. Interobserver agreement and receiver operating characteristics (ROC) analysis were performed.

Twelve dogs were enrolled, and metastatic melanoma was identified in 6 mandibular lymph nodes in 5 dogs. The interobserver agreement was higher when SUV quantification was employed (K = 0.58 versus 0.54). The area under the curve improved for both observers using this method (0.92 and 0.97 vs 0.86 and 0.90). The ROC analysis identified the SUVmax value of 3.3 as a cutoff leading to a sensitivity of 100% and a specificity of 83%.

In conclusion, including assessment of the oral cavity and use of quantification improves the accuracy of PET for metastasis detection in canine patients with OMM.

Disclosures

No disclosures to report.

ESVONC-O-9

Efficacy of diosmectite in the management of chemotherapy-induced diarrhoea in dogs: an open-label randomised clinical trial

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Chemotherapy-induced diarrhoea (CID) is one of the most frequent adverse events associated with chemotherapy in dogs. Yet, there is currently no consensus regarding its management. Metronidazole is frequently prescribed, however there is no evidence supporting its use, which could actually be associated with concerning gastrointestinal dysbiosis. Diosmectite is a natural medical clay, which is widely used for the treatment of acute diarrhoea in humans. There is strong evidence, both in vivo and in vitro, and in multiple species, supporting the use of diosmectite as an antidiarrheal.

The aim of this prospective study was to investigate the efficacy of diosmectite for the management of CID in dogs. We hypothesised that diosmectite would decrease the duration of CID compared to our standard management.

Dogs diagnosed with non-gastrointestinal neoplasia and undergoing maximum-tolerated dose chemotherapy between June 2017 and January 2019 were randomised into 2 groups ("diosmectite" and "standard" groups), and were randomly re-allocated if they developed another CID event. Diosmectite was administered at 0.5 g/kg/day PO divided in 2-3 doses to be initiated at the start of CID. "Standard" management consisted of a course of metronidazole at 10-15 mg/kg PO q12h to be initiated if the diarrhoea was not improved after 48 hours. Dogs were assessed weekly with standard quality of life (QOL) and diarrhoea diary forms filled by the owner, and physical examination performed by the clinician. The Waltham faeces scoring system was used to grade diarrhoea.

Sixty-one dogs were recruited during the study period. Twenty-three and 20 grade ≥ 4 diarrhoea events were recorded among the "diosmectite" and "standard "groups, respectively. Median duration of diarrhoea was significantly shorter (12h *versus* 96h) in the "diosmectite" group compared to the "standard" group ($P < 0.001$). Median QOL score was significantly higher (9/10 *versus* 7.5/10) in the "diosmectite" group compared to the "standard" group ($P = 0.0032$).

Management of CID in dogs with early administration of diosmectite was associated with a faster resolution of diarrhoea compared to our standard management with metronidazole, confirming our initial hypothesis. Diosmectite appears to be effective in the first-line management of CID in dogs, leading to an improved quality of life whilst decreasing antiobiotic usage.

Disclosures

Disclosures to report.

The diosmectite used for this study was kindly provided by VBS Direct LTD in the form of VBS Clay 100 g powder pots. VBS Direct LTD had no involvement in the design or performance of the study, writing the abstract, or the decision to submit it for presentation.

ESVONC-O-10

Contrast-enhanced ultrasound for sentinel lymph node identification in the routine staging of canine mast cell tumours: a feasibility study

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Regional lymph node (LN) assessment is part of the routine staging of canine mast cell tumours (MCTs). However, regional LNs are often determined based on their anatomical location and the draining LN(s) may not be accurately identified. In order to overcome this issue, different techniques of sentinel lymph node (SLN) detection have been reported. Contrast-enhanced ultrasound (CEUS) has been described to be a sensitive and specific technique in human patients, but has not been widely used in veterinary medicine.

The primary objective of this prospective study was to report the SLN detection rate of CEUS in dogs diagnosed with a cutaneous/subcutaneous MCT. A secondary objective was to assess the safety of this technique. We hypothesised that CEUS will identify at least one SLN in >80% of dogs, and that adverse reactions would occur in <5% of cases.

Dogs undergoing routine staging of cutaneous/subcutaneous MCT between June 2017 and March 2019 were recruited. Routine staging included bloodwork, urinalysis, fine-needle aspirate (FNA) of regional LN(s), thoracic radiographs, and abdominal ultrasound with FNA of liver and spleen. Regional LN assessment was completed with CEUS for identification of SLN(s). Premedication with intramuscular chlorpheniramine was administered, and 1-2 mL of sulfure hexafluoride microbubbles (SonoVue™) was injected around the tumour, followed by a local massage. A resident in training in diagnostic imaging examined with ultrasound the anatomical regions with potential draining LNs recommended by a resident in training in oncology. The injection site was checked following the procedure and before the discharge of the dog, and the owners were recommended to monitor the site for any local reaction for the following couple of days.

Sixty-five dogs diagnosed with a cutaneous/subcutaneous MCT were recruited. At least one SLN was identified in 61 (94%) of the dogs. Sixteen dogs (26%) had 2 SLNs and one dog (1.6%) had 3 SLNs identified. No adverse reaction to the procedure was recorded. Among the 30 dogs that had histopathological assessment of all the SLN(s) identified, 18 (60%) were diagnosed with nodal metastasis.

CEUS is a sensitive and safe technique for the identification of SLNs in dogs with MCTs, confirming our initial hypothesis. This technique may easily be incorporated to the routine staging of canine MCT, but additional studies are warranted to confirm its clinical benefit.

Disclosures

No disclosures to report.

ESVONC-O-11

Dorsal rhinotomy in 18 dogs with intranasal tumors

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Tumours of the nasal cavity and paranasal sinuses account for approximately 1% of all canine neoplasms. The treatment of choice for intranasal tumors is radiotherapy. Rhinotomy is associated with morbidity and a shorter survival than radiotherapy. The objective of this study was to retrospectively analyze the outcome of dogs with nasal tumours treated surgically. Patients with clinically low-grade nasal tumours, defined by clinical signs persistent for more than 6 months and no signs beyond local disease, were treated with surgery.

Eighteen dogs were included. Twelve had carcinomas (9 adenocarcinomas, 2 transitional type carcinomas and 1 adenosquamous) and 6 sarcomas (1 chondrosarcoma, 1 peripheral nerve sheath tumour and 1 haemangiosarcoma). The main complains were sneezing, nasal discharge and epistaxis. Fifty-five percent (n = 10) of dogs presented with epistaxis. Clinical signs were reported 2 months to 3 years (mean 8 months) prior to the surgery. Surgery was the sole treatment for 83% of dogs while 3 had radiotherapy and surgery. All dogs had blood analysis before surgery including haematology, biochemistry and coagulation times. One dog had planned auto-transfusion after surgery. CT imaging was performed in 94% (n = 17) of dogs, one had rhinoscopy. Dorsal rhinotomy was performed in all dogs without serious complications excluding one dog that required blood transfusiion after surgery. Nasal packing was placed and left for 24 hours. Most of the dogs developed mild to moderate facial emphysema and nasal discharge which resolved within two or three weeks.

Thirteen dogs died (72%) of which 11 died of local tumour progression. One each died from GI bleeding and renal carcinoma. Two dogs had a second surgery following relapse. At the time of data submission, three dogs were still alive at 900, 723 and 141 days and two dogs were lost to follow up 564 and 856 days after surgery. Overall median survival time was 893 days. There was no significant difference between the survival outcomes according to carcinoma or sarcoma diagnoses.

Historically, canine nasal tumours treated surgically achieved a median surival time of 7-9 months which is inferior to the outcome reported using radiotherapy. In our case series severe surgical complications were infrequent. This study reveals that some patients experience prolonged survival following surgery for intranasal tumours. More studies are necessary to better define this patient group.

Disclosures

No disclosures to report.

ESVONC-O-12

The Use of Low-dose Radiation Therapy for the Treatment of Small & Intermediate Cell Gastrointestinal Lymphoma in Cats

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Alimentary lymphoma is one of the most common forms of neoplasia in feline patients. Chemotherapy has been the main stay of therapy for decades for feline alimentary lymphoma. Overall response rates reported from 50-92% and overall survival times range from days to >3 years depending on the protocol utilized and the histologic form. For small cell lymphoma, treatment generally has involved the use of glucocorticoids in combination with chlorambucil (Leukeran). There is currently no standardized protocol for the use of these drugs with some clinicians choosing to continue the therapy for a defined period of time (6-12 months) or indefinitely until disease progression in the patient. The aim of this study is to establish a foundation for use of low-dose radiation therapy for feline intermediate and small cell alimentary lymphoma as an alternative to chronic chemotherapy or in patient's refractory to medical management.

TAMU medical records searched 2012 to 2018 for cases utilizing helical tomotherapy for treatment of intermediate and small cell alimentary lymphoma in feline patients. Exclusion criteria, disease extension beyond intestines and liver. Recorded information included patient signalment, baseline bloodwork, date, method of diagnosis, staging diagnostics, date of treatment initiation with chemotherapy and radiation therapy, any hematological abnormalities, first response duration to radiation therapy, and date of progression and/or death if available for the patient.

Ten cats diagnosed with alimentary lymphoma were treated with low-dose radiation. Seven cats diagnosed with small cell lymphoma were treated with 3 Gy total and three diagnosed with intermediate cell lymphoma were treated with 4 Gy total. Acute effects associated with radiation therapy were not observed. Median overall survival time for cats treated with radiation therapy was 1368 days, the median overall survival time for the control population receiving chemotherapy alone was 1161 days.

Radiation therapy appears to be efficacious and well tolerated with no clinically relevant adverse effects reported. In addition to the favorable adverse event profile, low-dose radiation was extremely convenient for the patients and clients as a number of these clients have to travel long distances for treatment, monitoring and follow-up. Client compliance and satisfaction improved due to a variety of factors such as reduced number of at-home medications, hospital visits and financial burden. Further investigation into the potential use of low-dose radiation therapy for intermediate and small cell alimentary lymphoma as a salvage or an alternative to oral chemotherapy for feline patients is warranted.

Disclosures

No disclosures to report.

POSTER RESEARCH COMMUNICATIONS

ESCG-P-1

Foxp3 and histopathological lesions in relation to outcomes in canine immunosuppressant-responsive enteropathy (Ire): prospective analysis in 57 dogs

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Canine immunosuppressant-responsive enteropathy (IRE) is an intestinal idiopathic inflammation, in which diet and antibiotic trials failed and immunosuppressants are needed. The number of transcription factor forkhead box P3 (Foxp3)-Positive Regulatory T lymphocytes have been investigated in IBD dogs in association with mortality. The aim of the study was to evaluate the clinical significance and prognostic role of histopathological changes and Foxp3-positive T cell in the clinical response and relapse. CCECAI and CIBDAI scores have been

assessed at presentation (T0) and 1 (T1), 3 (T3), 6 (T6) and 12 months (T12) from diagnosis. Endoscopic biopsies histopathology and features were classified using WSAVA guidelines score. Moreover four morphologic features were evaluated: presence of crypt distension (CD), lacteal dilation (LD), mucosal fibrosis (MF) and intraepithelial lymphocytes (IL). Immunohistochemistry for Foxp3 were performed in all cases. Dogs were classified as responders (decreased CCECAI and CIBDAI scores >25% at T1 compared to T0) and non-responders (decreased CCECAI and CIBDAI <25% at T1 compared to T0). Relapse was evaluated as follows: from T3, if clinical scores was >3, differences (D) between CCECAI and CIBDAI at T3, T6 and T12 and the previous closest time point were calculated obtaining DCCECAI and CIBDAI T3-T1, T6-T3, T12-T6. A DCCECAI and CIBDAI $^{3}2$ were considered relapse. Associations between response or relapse and categorical data were evaluated using Fisher's exact test and chi-square test. Fifty-seven dogs were prospectively enrolled. At T1, 9 and 8 dogs belongs to non-responders according to CIBDAI and CCECAI score, respectively. CIBDAI and CCECAI scores at T0 were not associated with T1 clinical response. Patients who relapsed were 5 (T3 and T6) and 4 dogs at T12, respectively. CIBDAI and CCECAI at T1, T3 and T6 were not associated with relapse. Dogs with histological WSAVA moderate had a higher response rate than severe dogs (P = 0.009, OR 6.5). However, histological scores were not associated with relapse rate. The 4 histological features were not associated neither with response nor with relapse rate. Presence of IL was associated with higher CIBDAI scores (P = 0.022). The percentage of Foxp3-positive cells was not associated with T0 CCECAI and CIBDAI or histological scores and morphologic features. The number of Foxp3-positive cells were not different between responders and non-responders and not related with relapse. Between the evaluated parameters, only histological grade seems to predict clinical response at T1. Furthermore, none of the clinical or histological parameters, including Foxp3, seems to predict relapse in IRE dogs.

Disclosures

No disclosures to report.

ESCG-P-2

Prevalence and significance of increased TLI concentrations in clinical practice

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TLI lacks specificity in individuals with certain gastrointestinal conditions, as has been previously reported in cats with intestinal and/or hepatic disease. In humans with chronic enteropathies, it has been suggested that enterocytes can synthetize small amounts of trypsin but is has not been verified for other species.

The aim of the present study was to evaluate the prevalence and significance of increased TLI in cats and dogs from clinical practice.

Results of serum TLI measurements were retrospectively reviewed from samples from animals evaluated for different diseases from two Veterinary Hospitals, performed in an outside laboratory (Idexx Barcelona) using a radioimmunoassay (feline) and chemiluminescent-assay (canine). Clinical records from patients with elevated TLI levels (>45 ng/L for dogs and > 82 µg/L for cats) were reviewed.

383 samples from 317 dogs and 66 cats were evaluated, and TLI was increased in 19.8% (70 dogs and 6 cats). Cobalamin was available for review in 181 of total patients, without correlation between cobalamin and TLI results (P = 0.143).

In 38 of the 76 patients with increased TLI, cPLI was also determined (semi-quantitative and/or quantitative analyses) with results consistent with pancreatitis in 50%(19/38). Among patients with normal cPLI results, pancreatitis was suspected in 2 based on pancreatic histology (n = 1) and abdominal ultrasound (n = 1), and non-cirrhotic portal hypertension (n = 1) and gastrointestinal disease (n = 16) were diagnosed in the other 17 patients. Gastrointestinal disease was confirmed in 11 cases [food-responsive enteropathy (5), IBD (4), gastrointestinal neoplasia (2)] and pressumptive in 5 patients (chronic enteropathy).

Pancreatitis was not suspected in any of the 38 patients with elevated TLI without cPLI analyses. In 30 patients underlying disease was confirmed: gastrointestinal disease (15), hepatopathy (4), renal disease (3), hypoadrenocorticism (1), insulinoma (1), neoplasia (2) and poliartritis (1). In the 11 patients with presumptive diagnosis, the most likely underlying disease was chronic enteropathy (n = 9).

In the present study 76,9% (40/52) of cases with increased TLI and without pancreatitis or azotemia had confirmed or suspected underlying gastrointestinal disease, coincident with previous feline and human findings. In fact TLI was performed in those cases to rule out exocrine pancreatic insufficiency. Although cobalamin deficiency has been linked to increased feline TLI (that normalized after supplementation), this study didn´t detect a significant correlation between cobalamin and TLI levels. Other yet undefined mechanisms likely explain the production of TLI in absence of pancreatic inflammation, and TLI levels must be carefully interpreted in animals with gastrointestinal disease.

Disclosures

No disclosures to report.

ESCG-P-3

Serum 25-hydroxyvitamin D3 in dogs with acute gastrointestinal diseases

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Hypovitaminosis D has been linked to systemic inflammation, chronic enteropathy (CE) and cancer in dogs. It is unknown if changes in serum 25-hydroxyvitamin D_3 [25(OH)D_3] are seen in dogs with acute enteropathy (AE).

This study aimed to investigate [25(OH)D_3] in dogs with AE and compare these to normal dogs and dogs with CE. It was hypothesized that dogs with AE had significantly different concentrations of [25(OH)D_3] compared to healthy dogs while similar concentrations to dogs with CE.

The study was approved by the local ethics committee. Twenty-eight client owned adult dogs were recruited prospectively. Ten healthy, 10 diagnosed with AE (clinical signs <72 hours) and 8 diagnosed with CE (clinical signs >3 weeks). Serum 25-hydroxyvitamin D_3, ionized calcium, total calcium, phosphorus, magnesium and C- reactive protein (CRP) were measured in all dogs. Dietary vitamin D_3 was calculated. Comparisons between groups and correlations were performed using appropriate parametric and non-parametric statistics. $P < 0.05$ was significant.

Descriptive statistics revealed comparable groups apart from an overweight of small medium breed dogs ($P = 0.01$) in the CE group. The mean $\pm$ SD [25(OH)D_3] was 271.4 $\pm$ 95.2 nmol/L for healthy dogs, 205.9 $\pm$ 62.1 nmol/L for AE and 196.5 $\pm$ 76.7 nmol/L for CE and not statistically different between groups ($P = 0.1$). A negative correlation between [25(OH)D_3] and CRP ($P < 0.05$) in dogs with AE was observed. No significant difference in electrolytes were found between groups. There was no correlation between [25(OH)D_3] and dietary Vitamin D_3.

In conclusion, in this pilot study, no statistical significant difference in [25(OH)D_3] was detected between groups.

Disclosures

No disclosures to report.

ESCG-P-4

Evaluation of abdominal ultrasound features in relation with canine Spec cPL, the severity of disease and mortality in suspected canine acute pancreatitis

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In canine acute pancreatitis (AP) abdominal ultrasound (US) is a widely-used non-invasive diagnostic tool involved in the diagnosis. Although, no specific recent studies about the relationship between US and clinicopathological features and severity of canine AP are available.

The aim of the study was to evaluate abdominal ultrasound features in relation with canine Spec cPL, severity of disease and outcome in suspected canine AP.

Dogs with suspected AP hospitalized between 2017 and 2019 were prospectively enrolled. AP was suspected based on compatible clinical and laboratory parameters, abnormal SNAP cPL test (Idexx Laboratories) at admission. Data regarding abdominal pain were recorded and serum samples for Spec cPL were sent to a commercial laboratory (Idexx Laboratories). US was performed at presentation, and every 24 h until 2 days from hospitalization. US was considered consistent with AP if there were hypoechoic and enlarged pancreas, irregular shape and margins, surrounded by hyperechoic mesentery and/or abdominal effusion. Recently developed Canine Acute Pancreatitis Severity (CAPS) score was calculated and dogs were divided into groups (CAPS <11 and > 11. Mortality rate was assessed at hospital discharge. US positivity at presentation was compared with the presence of abdominal pain, mortality rate and CAPS using Fisher's exact test. OR was also calculated. Spec cPL values were compared in positive/negative US at presentation using Mann-Whitney *U*-test. Forty-seven client-owned dogs were prospectively enrolled with owners' informed consent. Seventeen dogs (36%) died during hospitalization. Twenty-four dogs (51%) had US suggestive of AP at presentation, while other 10 US became positive within 2 days from hospitalization (US+ group, n = 34). Thirteen dogs (27%) remained US negative (US- group). No association between mortality and US positivity was found. Twenty-two dogs (47%) presented with abdominal pain. Dogs showing abdominal pain had significantly higher prevalence (88%) of positive US than dogs without abdominal pain ($P = 0.0014$; OR 10.22). CAPS and US positivity were not associated, although CAPS was associated with mortality ($P = 0.0021$; OR 9.3). Spec cPL were not significantly different between positive or negative US at presentation, and 8 dogs had negative US at admission, which became positive afterwards with Spec cPL < 400 mg/L. Interestingly, 4 dogs were in US- group, despite a Spec cPL > 400 mg/L.

In dogs with AP, changes in US could occur later during hospitalization, although the presence of abdominal pain at presentation may suggest US positivity. Furthermore, US positivity seems to be related neither with Spec cPL nor to the prognosis or the severity of the disease.

Disclosures

No disclosures to report.

ESCG-P-5

Gastric mucosal pathology in Belgian Shepherd dogs with and without clinical signs of gastric disease

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Belgian Shepherd dogs (BSD) are considered at increased risk for gastric carcinoma (GC), mucous metaplasia and glandular dysplasia. In humans, gastric mucosal atrophy, metaplasia and dysplasia are all linked to GC. We performed a prospective clinical trial exploring early diagnosis of such conditions. Gastroscopy with histology of mucosal biopsies was conducted to investigate possible associations of clinical signs, atrophy, metaplasia, dysplasia and GC.

After an online survey for recruitment, anamnesis and laboratory minimal database were performed. Canine chronic enteropathy clinical activity index (CCECAI) and signs of gastroesophageal reflux (GER) were used to ascribe dogs in two groups: with or without signs of gastric disease (Group A: CCECAI >1 and/or GER; Group B: CCECAI = 0 and no GER). Gastric inflammation, including mucosal fibrosis and glandular atrophy, was histologically assessed according to standardization guidelines by the World Small Animal Veterinary Association. Mucous metaplasia and glandular dysplasia were rated as present or absent.

Group A included 20 dogs (median age 9 years [5.5-11.6], mean CCECAI = 3.4 $\pm$ 2.1), and Group B included 11 dogs (median age 9.8 years [7.6-11.3]). Logistic regression analysis detected no statistical difference in histological findings between Groups A and B concerning mucosal atrophy (A: 14/20; B: 10/11), metaplasia (A: 5/20; B: 3/11), dysplasia (A: 11/20; B: 3/11) or GC (A: 4/20; B: 2/11). Independent of

grouping, numerous BSD had atrophy (25/31), followed by dysplasia (15/31), metaplasia (8/31), and GC (6/31). Out of the six dogs with GC, five had also atrophy, four metaplasia, and six dysplasia. Fisher's exact test revealed a significant association of GC with metaplasia ($P = 0.026$) and dysplasia ($P = 0.004$), but not with atrophy ($P = 1$). All neoplasms were gastric adenocarcinomas: 1/6 tubular type and 5/6 partly or completely of the diffuse, non-cohesive type, including four signet-ring-cell carcinomas and one mucinous adenocarcinoma.

This study in BSD showed a high proportion of gastric mucosal pathological changes, regardless of clinical signs of gastric disease; even GC presented repeatedly as an occult entity. The significant association of both metaplasia and dysplasia with GC supports an indication for endoscopic follow-up of affected BSD. Extensive endoscopic screening seems, however, unrealistic for early diagnosis due to the occurrence of occult disease. To address this problem, research on serum biomarkers for GC should be pursued. Future studies should also revise the diagnostic criteria for atrophy.

Keywords: gastric, carcinoma, atrophic gastritis, metaplasia, dysplasia, dog.

Disclosures

Disclosures to report.

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ESCG-P-6

Effect of stem cell therapy on serum cobalamin levels in dogs diagnosed with chronic enteritis without cobalamin supplementation

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Cobalamin deficiency is a common finding in dogs diagnosed with chronic enteropathy (CE) due to low absorption and/or bacterial competition. It has been described the importance of hypocobalaminaemia in the long-term prognosis of these patients being cobalamin supplementation highly recommended. Treatment of CE constitutes a therapeutic challenge and new approaches include stem cell therapy.

Our aim was to evaluate serum concentration of cobalamin in dogs diagnosed with CE in which stem cells were used in the absence of cobalamin administration.

Twenty dogs diagnosed with CE were included in this study; all showed persistent gastrointestinal signs, no response to conventional treatment (diet, antibiotics and/or immunosupressors/immunomodulators administration) and histopathological evidence of intestinal inflammation. The study was approved by the Ethical Committee of the University of Extremadura. A single dose of canine mesenchymal stem cells (MSCs) of allogeneic adipose origin at 2-4 x 10^6 cells per kilogram of weight was parenterally administered. A blood sample was obtained prior MSCs administration and subsequent cobalamin checkups were performed at 1, 3, 6 and 12 months; blood samples were submitted to a reference laboratory (Laboklin, Madrid, Spain). Clinical improvement was assessed at the same time points by the Clinical Activity Index (CIBDAI; clinically normal <3). The data were analyzed using a Saphiro-Wilk test and a repeated measures One-Way ANOVA followed by a Dunns or a Holm Sidak post-hoc test; $P < 0.05$ was considered as significant and results are expressed as mean ± SE of the mean. At the beginning of the study, 75% of dogs (15/20) presented hypocobalaminaemia (<300 pg/ml; laboratorial reference value). Statistically significant differences were observed between pre-treatment cobalamin values (225.3 ± 26 pg/ml) and those analyzed at 3 (360.7 ± 33.5 pg/ml), 6 (423 ± 55.7 pg/ml) and 12 (602.6 ± 54 pg/ml) months of treatment ($P < 0.001$). No significant differences were observed between pretreatment values and those obtained after one month of MSCs administration (299.2 ± 41 pg/ml). The CIBDAI significantly improved at all the checkups ranging from 8.2 ± 0.6 (pre-treatment) to 0.7 ± 0.3 (twelve months). CIBDAI was <3 for all groups treated with MSCs.

Our results demonstrate that administration of MSCs leads to an increase in serum cobalamin in dogs diagnosed with CE. This increase is associated with an improvement of their clinical status and therefore, MSCs therapy should be considered for the treatment of dogs affected with CE due to their positive impact on the long-term prognosis. Funded by: IB16133 and FEDER/FSE.

Disclosures

Disclosures to report.

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ESCG-P-7

Neutrophil-to-lymphocyte ratio (NLR) as a biomarker in dogs with chronic inflammatory enteropathies

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Chronic inflammatory enteropathies (CIE) comprise an important group of diseases in dogs. Only few biomarkers that can be routinely measured (eg, serum cobalamin and albumin, fecal calprotectin) appear to be of clinical utility in dogs with CIE and can potentially aid in CIE subclassification based on the response to treatment (ie, diagnosis of food-responsive enteropathy [FRE] vs. steroid−/immunosuppressant-responsive enteropathy [IRE]). The neutrophil-to-lymphocyte ratio (NLR) was recently shown to have diagnostic and prognostic potential in humans with inflammatory bowel disease (IBD). NLR also appears to be useful in the diagnosis of dogs with hypoadrenocorticism, but the NLR has not been evaluated or reported in dogs with CIE.

Data from 91 dogs diagnosed with CIE (that had not received steroids for ≥2 weeks prior to diagnostic evaluation) at 2 veterinary centers were used for this study. NLR was calculated as [neutrophil count/lymphocyte count] and was evaluated for a potential relationship with the severity of clinical signs (CCECAI scoring system, n = 65), serum albumin and cobalamin concentrations (n = 65), histologic lesion severity (4-point semi-quantitative grading system, n = 36), serum and fecal concentrations of other inflammatory markers (n = 60), and the response to treatment (FRE vs. IRE, n = 39). Statistical significance was set at $P < 0.05$.

NLR ranged from 0.23-54.0 (median: 5.69) in all dogs with CIE. NLR was significantly higher in dogs with very severe clinical signs compared to dogs with mild ($P = 0.014$) or moderate ($P = 0.026$) clinical disease. NLR was not correlated with the overall histologic score ($P > 0.05$), but was significantly higher in dogs with histologic lesions compatible with protein-losing enteropathy ($P = 0.006$). Hypoalbuminemia ($P < 0.001$), but not hypocobalaminemia, was significantly associated with a higher NLR. NLR correlated significantly with serum S100A12 ($P = 0.032$), C-reactive protein ($P = 0.046$), decoy receptor for advanced glycation end products ($P = 0.034$), and fecal alpha$_1$-proteinase inhibitor ($P < 0.001$), but not with serum or fecal calprotectin concentrations. Dogs with IRE (n = 26) had significantly higher NLRs (median: 8.41) than dogs with FRE (median: 3.09; n = 13; $P = 0.008$), and an NLR ≥4.60 best distinguished dogs with IRE from those with FRE (sensitivity: 77%, specificity: 69%).

Our findings suggest that neutrophils play a role in the systemic inflammatory response associated with CIE in dogs. NLR (ie., leukogram changes) in canine CIE are of similar magnitude as in human IBD. NLR can be easily obtained during routine hematology, and can potentially aid in the subclassification of dogs with CIE. The potential utility of NLR in the monitoring of dogs with CIE requires further investigation.

Disclosures

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Not applicable.

ESCG-P-8

Calprotectin concentrations are increased in the intestinal mucosa of dogs with chronic inflammatory enteropathies

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Calprotectin, a Ca^{2+} – binding protein of the S100/calgranulin family, has potential as a marker of inflammation in dogs and mainly originating from granulocytes. Increased canine calprotectin concentrations have been detected in feces and serum samples from dogs with chronic inflammatory enteropathy (CIE). However, intestinal mucosal calprotectin concentrations have not been extensively investigated in canine CIE.

We evaluated the mucosal concentrations of calprotectin in dogs with CIE in comparison with healthy Beagle dogs using a particle-enhanced turbidimetric immunoassay (PETIA) method on a clinical chemistry analyzer. Additionally, we assessed the association of mucosal calprotectin levels with the canine clinical IBD activity index (CIBDAI), histopathologic findings, clinical outcome, and serum albumin concentrations. Intestinal mucosal biopsies were collected from 38 dogs with CIE (duodenum [n = 34], ileum [n = 10], colon [n = 14], and caecum [n = 7]). Archived intestinal tissue samples from 18 healthy Beagle dogs served as controls (duodenum [n = 17], ileum [n = 18], colon [n = 18], and caecum [n = 6]). Data are presented as medians (interquartile ranges).

In comparison to healthy Beagles, mucosal calprotectin concentrations of CIE-dogs were higher in the duodenum (332 [91-639] vs. 94 [24-137] μg/L; $P = 0.001$) and colon (380 [187-542] vs. 112 [36-196] μg/L; $P = 0.002$). Histologic severity was significantly associated with mucosal calprotectin levels ($P < 0.05$) for total histopathology score, lymphoplasmacytic infiltration in the duodenum, and epithelial injury in the colon. Duodenal calprotectin concentrations were higher in hypoalbuminemic dogs than normoalbuminemic dogs (1441 [1098-1748] μg/L vs. 227 [74-506] μg/L), but because of the small number of hypoalbuminemic dogs (n = 4) the results were only descriptively reported. There was no significant association of mucosal calprotectin levels with CIBDAI scores or with the clinical outcome.

This study showed that mucosal calprotectin concentrations are increased in the duodenum and colon of dogs with CIE. The results provide supporting evidence for the potential diagnostic value of mucosal (or fecal) calprotectin concentrations in dogs with CIE. Further prospective research is needed to assess the value of measuring mucosal calprotectin concentrations in clinical practice, the relationship between mucosal and fecal calprotectin, and other inflammatory markers in dogs with CIE.

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ESCG-P-9

Neutrophil-to-lymphocyte ratio (NLR) in canine patients with immunosuppressant-responsive enteropathy (IRE)

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In human IBD, neutrophil-to-lymphocyte ratio (NLR) was associated with active bowel inflammation and correlated with clinical and laboratory indices. So far, NLR in dogs has been only evaluated in oncologic patients and in septic peritonitis.

The aim of the study was to evaluate the NLR in canine immunosuppressant-responsive enteropathy (IRE).

Forty-one dogs presented to two veterinary facilities (Veterinary Teaching Hospital and Private Veterinary Center) with a final diagnosis of IRE were retrospectively included. The Canine Chronic Enteropathy Clinical Activity Index (CCECAI) score was assessed for each dog at presentation. The diagnosis of IRE was set on histopathology performed on endoscopic biopsies and classified using the current WSAVA guidelines. Lacteal dilatation (LD) and crypts abscesses (CD) were also recorded. NLR was calculated for each dog. Serum total protein, albumin, cholesterol and C-reactive protein (CRP) were also recorded. Kruskal-Wallis test was performed to evaluate NLR between different CCECAI category (0-3, 4-5, 6-8, 9-11 and > 12) and different histological grading. Spearman's correlation tests were performed between NLR and total protein, albumin, cholesterol and CRP. Mann-Whitney *U*-test was used to compare NLR in dogs with or without LD and CD. A receiving operator characteristic curve (ROC) was built to obtain an optimal cut-off value of NLR to differentiate dogs with or without LD. A Fisher's exact test was then performed between the presence of LD and NLR groups.

NLR was significantly different between CCECAI score categories ($P = 0.004$). NLR was negatively correlated with total protein ($P = 0.022$, $r = -0.35$), albumin ($P = 0.007$, $r = -0.41$) and cholesterol ($P = 0.03$, $r = -0.33$). No significant correlation between CRP and NLR was found. NLR was not different between histological grading and dogs with or without CD. Contrarily, NLR was higher in dogs with LD ($P = 0.004$). The cut-off value of NLR for the detection of LD was 3.96 (sensitivity 82.4% and specificity 58.3%).

So far, this is the first report evaluating the NLR in IRE dogs. Our results suggest that NLR could be an easy, feasible and economic additional tool to evaluate the disease severity in IRE dogs. Moreover, NLR seem to have a good correlation with other essential biochemistry parameters in the evaluation of dogs with protein-losing enteropathy. Furthermore, the most interesting data was the association between NLR and histologic lymphangectasia.

Disclosures

No disclosures to report.

ESCG-P-10

A Novel Canine-Specific Model System to Study Intestinal P-Glycoprotein-Mediated Drug Transport

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P-Glycoprotein (P-gp) modulates oral absorption of therapeutic drugs in the small intestine. Many drugs serve as substrates for P - gp

including chemotherapeutic agents (vincristine, doxorubicin), parasiticides (macrocyclic lactones) and antidiarrheal agents (loperamide). Due to its significant influence on drug disposition and the risk of severe adverse drug reactions seen with defective P - gp function, routine screening of drug candidates for P-gp transport is common practice in human drug discovery. However, there is currently no canine-specific *in vitro* system for evaluating P-gp-mediated drug transport in veterinary medicine available. Our laboratory has recently developed an *ex vivo* 3D canine enteroid (ENT) system, which accurately mimics the cellular and molecular features of the intestinal epithelium in vivo. The aim of this research was to evaluate P-gp functional activity and expression in canine ENT vs. intestinal tissues.

P-gp expression was assessed using qPCR of intestinal tissues and ENT from the ileum of 3 healthy and diseased dogs. Immunofluorescence (IF) staining of canine ileal ENT in transwell culture was performed to assess the localization of P-gp transporters. Functional assays were performed using 10 μM rhodamine123 (Rh123), a fluorescent dye substrate for P-gp, with or without 20 μM verapamil (P-gp inhibitor). An un-paired *t*-tests was used to compare the mean luminal fluorescence intensity obtained with ImageJ. Kruskal-Wallis test was used to compare the mean quantitation cycle obtained with qPCR, and $P = 0.05$ was considered as statistically significant.

P-gp gene expression was not significantly different between ENT and the epithelial layer of the intestinal mucosa (quantitation cycle values expressed in Mean ± S.D): Organoid: PLE M = 24.5 ± 1.0; RW M = 24.7 ± 0.3; HC M = 25.7 ± 0.3; Tissue: PLE M = 25.4 ± 0.6; RW M = 26.3 ± 0.7; HC M = 24.1 ± 0.5; $P = 0.06$), confirming that ENT express P-pg in comparable amounts as reported *in vivo* tissues from the same dogs. The culture of primary canine ENT in Transwell showed apical expression of P-gp, which is again consistent with *in vivo* observations. Co-incubation with verapamil significantly reduced Rh123 fluorescence in the lumen of ENT at 30 min ($P < 0.0001$) and 60 min ($P < 0.001$), indicating that P-gp-mediated transport was successfully blocked.

In summary, P-gp gene expression, localization, and function in canine ENT were similar to those of intestinal tissues they were originally derived from. Our novel ENT model can serve as a useful *ex vivo* system for oral drug transport and related safety studies in veterinary medicine.

Disclosures

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ESCG-P-11

Investigation of the efficacy of a novel diet in the management of chronic enteropathies in dogs

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Chronic Enteropathies (CE) are a common cause of morbidity in dogs. Chronic enteropathies are diagnosed in dogs with chronic gastrointestinal clinical signs (>3 weeks), inflammatory changes on intestinal biopsies and where no other underlying cause is determined based on a thorough, standardised diagnostic workup. Based on response to therapy, CE are sub-classified into food-responsive, antibiotic-responsive or steroid-responsive enteropathies. A significant proportion of dogs with a CE are food-responsive; however, there are limited peer-reviewed publications describing the clinical efficacy of the commercially available food used to treat CE.

In this study, we evaluated the response of 15 dogs with a CE to a commercially available dietetic food (Hill's Prescription Diet i/d Sensitive Canine Dry). The dogs underwent a standard diagnostic evaluation, and did not receive concurrent anthelmintics, antibiotic, glucocorticoid or gastroprotectant therapies. The clinical efficacy of the dietary treatment was assessed by comparing the Canine Inflammatory Bowel Disease Activity Index (CIBDAI) before and a median of 13 days after dietary therapy.

We found that the CIBDAI significantly decreased following the introduction of the dietetic food (median CIBDAI score pre treatment 9, post treatment 2 [$P < 0.0005$]). Our study demonstrates that this dietetic food can be used to successfully manage CE in dogs.

Disclosures

Disclosures to report.

Although the study was not funded, owners got the food from Hills for free during the trial.

ESCG-P-12

Effect of dietary fat content on mucosal microbiota and serum metabolome in healthy beagles

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Dietary fat composition has been shown to modulate fecal microbiota composition and impact host health. High-fat diets have been linked to reduced fecal microbial diversity, increased Firmicutes to Bacteroidetes ratio, and low-grade systemic (LPS) inflammation. While high-fat diets may modulate the fecal microbiota, there is no data available on the impact of high-fat diets on the canine mucosal microbiota. The aim of the study was to investigate changes in composition of the mucosal microbiota and serum metabolome in Beagle dogs fed two rations varying in their content of dietary fat.

Eight healthy adult Beagle dogs were fed a control diet (13% fat), followed by low-carbohydrate diet (1% starch) containing either 32% fat (T1) or 46.5% fat (T2) for 2 weeks each in randomized order. Endoscopic biopsies of the small and large intestines and sera were collected for analysis of mucosal microbiota and metabolomic profiles, respectively, before and during dietary intervention. Fluorescence *in situ* hybridization (FISH) using a 4-probe array (ie, total bacteria, Firmicutes [*Clostridium* cluster XIVa], Bacteroides-Prevotella and Enterobacteriaceae) quantified colonic mucosal bacteria into distinct compartments. Serum metabolomic profiles resulting from the different diets were quantified by a targeted approach to analyze samples using mass spectrometry and the Biocrates AbsoluteIDQ p400 HR Kit. Metabolite changes between diets were analyzed by PCA, PLS-DA, HCA and

univariate statistics. *P* values <0.05 were considered statistically significant.

Analysis by FISH showed that most mucosal bacteria (EUB-338) were located within the adherent mucus. There was no difference in the total number and spatial distribution of bacteria within the mucosa of dogs fed control diet versus T1. For the different bacterial groups, sub-populations of *Clostridium spp*. were significantly ($P < 0.05$) increased in adherent mucus of dogs fed T2 versus T1. Changes in mucosal bacteria were accompanied by altered serum metabolomes of dogs fed either T1 or T2. Perturbations in lipid metabolism predominated and primarily involved different glycerophospholipids (GPL), including the phosphatidylcholines (PC) and acylcarnitines (AC). Most GPLs were significantly ($P < 0.05$) reduced in dogs fed T2 but not T1 when compared to control ration. Conversely, T2 also resulted in lower ($P < 0.05$) concentrations of other lipid metabolites and select amino acids.

Our results indicate that enhanced dietary fat modified the mucosal microbiota and the serum metabolome of healthy dogs. Consumption of high fat diets has implications for canine health by modulating host immune responses in association with changes in gut microbial composition.

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ESCG-P-13

The erythrocyte membrane lipidome in dogs with chronic enteropathy

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Chronic enteropathies (CE) are common cause for persistent or recurrent gastrointestinal signs in dogs. Food-responsive enteropathy (FRE), antimicrobial-responsive enteropathy (ARE), and immunosuppressive-responsive enteropathy (IRE) have different etiologies however clinical signs overlap and distinguishing among these disorders may be challenging with the most reliable diagnostic tool represented by sequential treatment using diet, antimicrobials, and immunosuppressive drugs.

Analysis of erythrocyte membrane lipidome represents a powerful tool in humans for assessing the quantity and quality of fatty acids (FA) and the follow-up of the membrane FA remodeling under physiological and pathological conditions. The aim of this study was to compare the FA membrane profile of healthy dogs (HD, n = 68) with 29 dogs with CE (ie, >3 weeks). Dogs receiving dietary ω3 supplementation were excluded from the study.

Erythrocyte membranes were isolated from EDTA-treated blood and a cluster of 10 FA, that is, saturated [SFA (palmitic; stearic)], monounsaturated [MUFA (palmitoleic; oleic; vaccenic)], polyunsaturated [ω-6 (PUFA-ω6): linoleic, dihomo-gamma-linolenic, arachidonic and ω-3 (PUFA-ω3): eicosapentaenoic and docosahexaenoic] FA, was determined by Gas-Chromatography. Results are referred as % of one FA in the cluster. Relevant lipid parameters (SFA/MUFA, SFA/PUFA, ω6/ω3, PUFA balance, unsaturation and peroxidation indexes) were calculated.

HD dogs were 30 males (6 neutered) and 38 females (12 sterilized) with a median age of 41 months (2-156), while CE dogs were 20 males and 9 females (4 sterilized) with a median age of 43 months (10-114). Among CE dogs 11 were diagnosed with FRE, 1 ARE, 6 IRE, while 11 are undergoing diagnostic trials or were lost to follow-up. Diminished value of palmitic acid ($P < 0.0001$) and increased value of stearic acid ($P < 0.0001$), with decreased total SFA ($P < 0.05$) were observed in CE group. Among PUFA-ω6, CE dogs showed increased values of dihomo-gamma-linolenic ($P < 0.001$) and arachidonic ($P < 0.05$) acids, while no differences were observed in PUFA-ω3 levels between the two groups. Unsaturation ($P < 0.05$) and peroxidation ($P < 0.05$) indexes were found significantly increased in CE dogs. Interestingly, dogs with FRE and IRE dogs had similar erythrocyte membrane lipidome profiles; ARE was not object of statistical analysis, due to the low number of dogs in this group.

These results point out the importance of the balance between pro-inflammatory arachidonic acid and the anti-inflammatory dihomo-gamma-linolenic acid levels in the inflammatory conditions of CE.

The erythrocyte membrane lipidome of dogs may be successfully applied in dogs with CE, providing important information leading to personalized intervention targeted to decrease inflammation and increase protective components.

Disclosures

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ESCG-P-14

Hypercobalaminaemia and its possible association with disease severity in dogs: a retrospective study of 47 cases

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Serum cobalamin concentration is frequently assessed in companion animals, especially when facing a patient with chronic gastrointestinal signs. Although the clinician's attention is mainly focused on patients with hypocobalaminaemia, recent studies in humans and cats suggest that high serum cobalamin concentration could be associated with specific conditions such as neoplasia and liver disease and, in addition, may be a marker of severity. The aims of this retrospective, cross-sectional study were therefore to identify the conditions associated with hypercobalaminaemia in dogs and to determine whether it could be used as a marker of disease severity in these patients.

Medical records of dogs having serum cobalamin measured between November 2016 and December 2018 in 14 practices in the United Kingdom were reviewed. Dogs were excluded if they had received cobalamin supplementation at any time prior to analysis. Signalment, clinical signs, laboratory & imaging findings were recorded for each case. The cases were then classified into different disease categories by consensus depending on the final diagnosis. Values were expressed

as percentages and medians and variables were compared between groups using a Kruskal-Wallis, Chi-2 or Fischer's exact test.

One hundred-and-sixty dogs were included in the study and divided into three groups: hypocobalaminaemia (39 dogs), normal serum cobalamin concentration (74 dogs) and hypercobalaminaemia (47 dogs). The age distribution was significantly different between groups (P = 0.0214), with hypercobalaminaemic dogs being significantly younger (median age 79 months, range [2-207]). Dogs with hypercobalaminaemia presented with diarrhoea (49%), vomiting (47%), inappetence (38%), lethargy (40%) and/or weight loss (38%); this was not significantly different from the other groups. Conditions associated with hypercobalaminaemia included gastrointestinal (57%), hepatic (11%), neurological (11%), endocrine (9%), renal (4%), pancreatic (2%) and miscellaneous (6%) diseases. Among all, 11% had neoplasia. This distribution was not significantly different from hypocobalaminaemic and normocobalaminaemic dogs.

There were significantly more dogs with high serum folate concentration in the hypercobalaminaemia group, as compared to the other groups (P = 0.009). Dogs with hypoalbuminaemia, anaemia, high ALT activity and/or hypocholesterolaemia were also compared between the three cobalamin groups and no statistical difference was identified. There was no association found between hypercobalaminaemia and the parameters tested for disease severity.

Our results suggest that hypercobalaminaemia in dogs is most commonly seen with gastrointestinal and hepatic disease as with other species, but can also be seen with endocrine and neurological conditions. Interestingly, hyperfolataemia was most commonly seen with hypercobalaminaemia; it is unknown whether this reflects or not active dysbiosis.

Disclosures

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ESCG-P-15

In vitro model (SCIME) to study the intestinal microbiota in dog

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In vivo studies on the physiology of the gastrointestinal tract (GIT) in living animals meet with serious technical difficulties and ethical question. Therefore, much attention has been given recent years to the development of *in vitro* models which mimic metabolic process of the GIT.

The aim of the present work is the validation of the SCIME (Simulator of the Canine Intestinal Microbiome) above the SHIME model (Simulator of Human Intestinal Microbiome Ecosystem) considering the physiological parameters of the dog, the different diet and the different microbiological populations comparatively to the in vivo microbial population from the faecal samples of donor dogs.

Fresh faeces collected from four healthy dogs were inoculated in the SCIME system, the experiment lasted 14 days and was conducted in duplicate by giving two different types of feeds to each donor. Model validation was evaluated through analysis of microbial activity by the quantification of SCFA, lactate, and ammonium. The composition of the colonic microbiota was studied through qPCR using primers targeting the Firmicutes, Bacteroidetes, Bifidobacteria, Lactobacilli, and Enterobacteriaceae and through 16S-targeted Illuminates sequencing of the total bacterial population.

Different effects on the composition of the canine intestinal microflora during the experiment time were promoted. Among *Firmicutes*, the results obtained highlighted the ability to a significant increase (P < 0,005)in the amount of *Acidaminococcaceae (OTU 0022:0.8 ± 0.9% at lumen level)* and *Enterococcaceae (P < 0,0005)*. The *Bacteroidetes* displayed a decrease (P < 0,05) in species that belong to the family of *Bacteroidaceae (OTU 0002:5.2 ± 0.2% at lumen level)*. In addition, there is a significant growth (P < 0,05) in the family of *Coriobacteriacaee*. The *Akkermansiaceae* family *(OTU 0010:4.2 ± 0.4% at lumen level)* is absent in the inoculum but there is a little increase during the experiment. *Enterobacteriaceae (OTU 0008:23.2% ± 0.4)* recorded a significant increase (P < 0.05).

Interesting is the result regarding the SCFAs where is promoted a significant increase (P < 0.05) in their concentration; especially in the amount of butyrate *(average: 2.05 ± 2.19 mmoL/L)*, acetate (*average: 7.41 ± 5.34 mmoL/L*) and branched fatty acids *(average: 1.37 ± 0.58 mmoL/L)*.

Finally, the study highlighted the ability of the SCIME model to increase also the ammonium levels showing an average concentration of 461.37 ± 63.80 mg/L for the proximal colon and 580.79 ± 68.25 mg/L for the distal colon.

It followed that the novel model allowed the growth of the bacteria present in the original inoculum, offering a relevant technology platform to simulate the intestinal ecosystem for evaluation of pharmaceutical and nutraceutical effects on dog microbiota.

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ESCG-P-16

Expression and distribution of Toll-Like Receptor (TLR)2, TLR4, TLR5 and TLR9 in the colonic mucosa of dogs with Inflammatory Bowel Disease

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Inflammatory bowel disease (IBD) is a common cause of chronic gastrointestinal disease in dogs. The current paradigm of IBD involves complex interactions between environmental factors, such as the intestinal microbiota, and dysregulated host responses. Toll-Like receptor (TLR) 2, TLR4, TLR5 and TLR9 recognize bacterial Pathogen-Associated Molecular Patterns (PAMPs). In a previous study, TLR2, TLR4, and TLR9 mRNAs were reported to be up-regulated in the inflamed duodenal and colonic mucosa of IBD dogs compared to asymptomatic healthy Beagles. The aim of this study was to evaluate

the expression of these receptors by immunohistochemistry in colon biopsies isolated from dogs with IBD compared to asymptomatic controls.

Ten dogs with IBD were included in this study. Diagnosis was based on clinical signs of at least 3 weeks' duration, the presence of a lymphocytic and plasmacytic and/or eosinophilic inflammation on colon biopsies and exclusion of other causes of chronic gastrointestinal signs. Nine dogs were included in the control group. Colon paraffin-embedded biopsies were processed for immunohistochemistry using anti-human TLR2, TLR4, TLR5 and TLR9 antibodies. The expression of TLRs in the different samples was graded from 0 (no expression) to 3 (strong expression) by a board-certified pathologist in a blinded fashion.

TLR5 and TLR9 were detected both in the epithelial cells and in the lamina propria cells. In contrast, TLR2 was detected only in the epithelial cells while TLR4 staining was restricted to the leucocytes of the lamina propria. No statistical differences were found in TLRs expression when comparing IBD dogs with asymptomatic dogs although epithelial TLR9 expression tended to be higher in the IBD group (P = 0,054).

To our knowledge, our study describes for the first time the pattern of expression of TLR2, TLR4, TLR5 and TLR9 in the colon of dogs and suggests that their expression is poorly affected by IBD. Further experiments are warranted in a larger number of patients and in other intestinal segments (duodenum and ileum) to analyze whether epithelial TLR9 expression might be correlated with clinical activity or histopathology score.

Disclosures

Disclosures to report.

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ESCG-P-17

Serum Vitamin A and E concentrations in dogs with pancreatitis

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Acute and chronic pancreatitis in humans results in significantly lower levels of serum Vitamin A and Vitamin E due to antioxidative stress and as part of the inflammatory response. To our knowledge no studies about Vitamin A and E levels in serum of dogs with pancreatitis have been performed so far.

The aim of this retrospective study was to evaluate the level of serum vitamin A and E from routine diagnostic left-over samples in dogs with pancreatitis compared to a control group of dogs with other diseases.

40 dogs with pancreatitis were included in this study. These dogs all had serum pancreatic lipase concentrations >600 µg/L and clinical symptoms of pancreatitis like vomiting, abdominal pain or inappetence and had received no medical treatment so far. Additionally, in 3 dogs pancreatitis was diagnosed by histology of pancreatic tissue biopsies.

The 45 dogs of the control group had serum pancreatic lipase values <40 µg/l and had clinical symptoms concurrent with other diseases than pancreatitis.

Serum Vitamin A and E levels were measured by HPLC from cooled serum within 72 hours of sampling. Dogs with pancreatitis had significantly higher (P < 0.001) levels of serum Vitamin A (average 1701 µg/L, range: 405-4448 µg/L) than dogs of the control group (average: 978 µg/L, range: 139 µg/L - 1904 µg/L). The study dogs also had significantly higher (P < 0.001) serum Vitamin E levels (average: 36.0 µg/L, range: 15.3-83.7 µg/L) than the control group (average: 20.4 µg/L, range: 1,9-54.6 µg/L).

This study suggests that dogs with pancreatitis have a different metabolism of Vitamin A and E compared to dogs with other diseases and humans with pancreatitis. In the course of pancreatitis a release of Vitamin A and E into the blood stream due to necrotic processes of tissue may be discussed in this species.

Disclosures

Disclosures to report.

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ESCG-P-18

Water immersion vs gas insufflation in canine duodenal endoscopy: is the future underwater?

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The endoscopy of the gastrointestinal tract is often part of the diagnostic protocol for canine acute and chronic gastroenteropathy. To analyze properly the intestinal wall, it is fundamental to distend the lumen, usually inflating it with air. In human medicine, it is well known how the gas insufflation (GI) with air or carbonic dioxide during colonscopy can induce pain in the patient. More and more frequently it is used warm-to touch water, instead of air, to distend the lumen. Randomized controlled trials suggest that the introduction of water to distend the lumen of the colon decreases spasm of the musculature of the bowel and pain, and significantly increases the visualization of mucosal texture and the adenoma detection rate.

This study was the first in veterinary medicine to compare GI and water immersion (WI) during duodenoscopy in anesthetized dogs, in order to evaluate eventual differences in procedural nociception and in the quality of mucosal visualization.

Twenty-five dogs, subjected to endoscopy under general anesthesia, were included in the study. To evaluate differences in nociception during anesthesia, heart rate and arterial blood pressure (sistolic, diastolic and mean) were measured throughout the procedure and divided into four steps (baseline, water, air, outcome). A random sequence of GI or WI was applied to dilate duodenal lumen and, in every condition, the same mucosal image of the bowel was recorded. For every dog, two images (GI and WI) were recorded and subjected to a texture analysis by using image processing approaches like skeletonization and entropy evaluation, and to a subjective blind evaluation by three expert endoscopists, considering the architecture and the intestinal texture. No systematic significant differences were detected for the cardiovascular parameters and the texture analysis between GI and WI, except for the subjective evaluation by the endoscopists, who identified the WI images as qualitatively better.

The results of this study highlight how the algic answer does not change between the two methods, maybe influenced by the drugs used during the endoscopy, which well control nociception and give deep anesthesia. Based on the evaluation of the endoscopists, the WI allows to get better quality images, with a detailed visualization of the intestinal villi, while this is still not confirmed by objective texture analysis.

Disclosures

No disclosures to report.

ESVC-P-1

Heart rate variability of dogs in various stages of degenerative mitral valve disease

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Hemodynamic changes associated with mitral valve degeneration (MVD) activate neurohumoral mechanisms, to maintain adequate cardiac output and capillary perfusion pressure. A predominant sympathetic and/or a diminished vagal tone are linked to disease progression. Only scarce information about autonomic nervous system (ANS) dynamics during MVD progression is available in the veterinary literature. To our knowledge, the ANS imbalance in dogs with MVD prior to receiving cardiac pharmacotherapy has not been evaluated. We sought to analyze heart rate variability (HRV) for assessing the ANS activity in dogs with preclinical and yet untreated clinical MVD.

Seventy-four client-owned dogs with echocardiographically confirmed MVD were retrospectively divided into three groups according to the 2009 ACVIM Consensus Statement: B1 (n = 20), B2 (n = 20) and C (n = 34). A control group, labeled N (n = 21) included healthy normal dogs. All dogs received physical examination, five-minute-long six-lead electrocardiography (ECG), complete echocardiography, thoracic radiography and bloodwork. Short-term heart rate variability was evaluated using the time and frequency domains from the five-minute-long ECG recordings.

A significant decrease in time domain parameters was shown in Group C when compared to Groups N, B1 and B2. These parameters included the SD of all normal intervals (SDNN) *($P < 0.05$)*, the root-mean square of successive differences between normal heartbeats (rMSSD) *($P < 0.01$)*, and the percentage of adjacent normal intervals that differ from each other by more than 50 ms (pNN50) *($P < 0.01$)*.

In addition, significant increase in the low-frequency (LF, *$P < 0.01$)* and a decrease in high-frequency (HF, *$P < 0.01$)* band was observed in Group C, compared to Groups N, B1 and B2. The LF/HF ratio was significantly increased in Group C *($P < 0.01$)* relative to all other groups. When Group B2 was compared to B1, only SDNN showed significantly lower values *($P < 0.05$)*.

This study shows that in dogs with aclinical MVD, both sympathetic and parasympathetic tones might remain unchanged until only after clinical signs have developed, despite the presence of structural remodeling. The onset of clinical signs is likely to trigger an ANS imbalance due to both sympathetic tone activation and vagal tone withdrawal. Moreover, SDNN, which is an independent marker for risk of mortality, may be the only HRV-related parameter able to distinguish between aclinical dogs with and without cardiomegaly.

These findings may have future implications regarding choices made around the best timing of pharmacotherapy onset, as well as around the most effective order of adding specific medications to an on-going pharmacotherapy regimen, administered to dogs with MVD.

Disclosures

No disclosures to report.

ESVC-P-2

Comparison of serum digoxin concentrations from blood collected in Vacutainer® tubes with or without gel

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Therapeutic monitoring of serum digoxin levels to avoid digoxin intoxication is performed at least 3 to 5 days after beginning therapy. Some labs discourage the use of blood collection tubes containing gel because of potential absorption of digoxin by the gel resulting in an underestimation of the serum digoxin concentration. The aim of this study was to compare serum digoxin concentrations in blood collected in serum tubes with and without gel. Blood samples were taken with a syringe and needle from the jugular vein of 48 dogs treated with digoxin (Lanoxin®). For each dog, half of the blood sample was put in a tube containing no gel (NGC) (Vacutainer Z) while the other half of the sample was put in a gel containing (GC) tube (BD Vacutainer® STT II Advance). NGC tubes were sent to the referral lab (Synlab, Belgium) for immediate analysis. GC tubes were kept uncentrifuged and refrigerated for 5 days before analysis was performed. All analyses were carried out by the same lab using the ARCHITECT ci System (Abbott Diagnostics, US). Median digoxin concentration (quartile 1-3) was 0.90 μg/L (0.70-1.10) for the NGC tubes and 1 μg/L (0.80-1.10) for the GC tubes. Agreement between the two methods was shown by a mean bias of 0.05 and a narrow range of agreement (lower level of agreement: −0.09; upper level of agreement: 0.18). Mean bias (%) was 5.1% and did not exceed the method quality specifications for total allowable error (14%). Bland-Altman plot of data did not show significant differences in data sets. Serum digoxin concentrations do not decrease when collected in tubes containing gel and kept refrigerated for 5 days before analysis. Blood collection serum tubes containing gel can be used for therapeutic monitoring of serum digoxin levels.

Disclosures

No disclosures to report.

ESVC-P-3

Transverse right ventricle strain and strain rate assessed by 2-dimensional speckle tracking echocardiography in dogs with pulmonary hypertension

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Right ventricular (RV) strain analysis using 2-dimensional speckle tracking echocardiography has focused on assessing longitudinal strain and strain rate variables in dogs. However, RV contraction is also characterized by transverse deformation; this strain component has not been investigated in dogs. Therefore, we evaluated the ability of transverse RV strain and strain rate, obtained by 2-dimensional speckle tracking echocardiography in healthy dogs and dogs with pulmonary hypertension, to identify dogs with pulmonary hypertension. Additionally, we examined relationships of transverse strain and strain rate variables with heart rate, age and bodyweight in healthy dogs, and with tricuspid regurgitation (TR) velocity and left atrial size in dogs with pulmonary hypertension.

We acquired 2D echocardiographic cineloops from the left apical 4-chamber view optimized for the right ventricle and analyzed transverse RV free wall strain and strain rate in 74 dogs (40 healthy dogs and 34 dogs with pulmonary hypertension) using Xstrain® software. Dogs were classified as having pulmonary hypertension based on the TR jet velocity (> 3 m/sec). We classified dogs as having moderate pulmonary hypertension if TR velocity > 3.5 m/sec, and severe pulmonary hypertension if TR velocity > 4.5 m/sec.

Seven dogs (3 healthy and 4 dogs with pulmonary hypertension) were excluded during the analysis for low quality images. In healthy dogs, strain and strain rate showed no relationship with heart rate, body weight or age. In dogs with pulmonary hypertension, strain and strain rate showed weak negative relationships with TR velocity ($r2$ = 0.25), but no relationship with left atrial size ($r2$ = 0.05). Although transverse RV strain (but not strain rate) showed a negative relationship with class of pulmonary hypertension, it was not useful in identifying dogs with pulmonary hypertension.

Transverse RV strain and strain rate using 2-dimensional speckle tracking echocardiography can be obtained in most dogs, but does not help in identifying dogs with pulmonary hypertension.

Disclosures

No disclosures to report.

ESVC-P-4

Potential renoprotective effect of angiotensin-receptor antagonists in dogs with myxomatous mitral valve disease

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Blockade of renin-angiotensin-aldosterone system is a pivotal strategy to manage congestive heart failure (CHF). Angiotensin-receptor blockers (ARBs) are perceived to offer more complete neurohormonal suppression in the treatment of CHF by directly occupying angiotensin II receptor sites. However, whether the combination therapy of ARBs with current CHF treatments has clinically incremental benefits, is unknown in dogs with myxomatous mitral valve disease (MMVD).

This retrospective cohort study was conducted with client-owned dogs with symptomatic MMVD, which have been treated with a standard protocol (Controls, n = 43) and with the addition of ARBs (Cases, n = 29; irbesartan 6~16 mg/kg/day, n = 20; telmisartan 2~4 mg/kg/day, n = 9) to the conventional treatment. For the initial analysis of homogeneity between two groups, physical examination, comprehensive blood test profile (CBC, serum biochemistry, NT-proBNP, SDMA), radiographic, and echocardiographic data were collected. Subsequently, the same clinical indices were compared at the time points when ARBs were given for three to six months. In addition, survival analysis was also performed using Kaplan-Meier curves.

After 3~6 months treatment, the degree of increases in clinical parameters related to renal function was significantly higher in the control than the case group (P < 0.01): BUN (+11.3 ± 9.9 vs. -3.3 ± 7.9), creatinine (+0.5 ± 0.27 vs. +0.1 ± 0.19), SDMA (+5.1 ± 2.25 vs. +2.0 ± 5.78), sodium level (+3.5 ± 3.31 vs. -0.8 ± 3.81). The differences of heart rate (+7.9 ± 12.2 vs. -1.5 ± 9.8) and blood pressure (+14.2 ± 14.6 vs. -3.8 ± 15.9) between baseline and follow-up were significantly lower in the case group (P < 0.01). Furthermore, the increased level of NT-proBNP (1621.4 ± 1274.2 vs. 444.3 ± 619.5) was also greater in the controls (P = 0.04). However, no statistical differences were found in thoracic radiographic and echocardiographic indices between the groups. Comparing survival curves of two groups did not reveal an overall survival advantage with ARBs treatment.

These results suggest that ARBs in combination with ongoing conventional CHF treatments may provide an additional renoprotective effect in dogs with MMVD.

Disclosures

No disclosures to report.

ESVC-P-5

Perioperative management with peripheral arteries in dogs undergoing open heart surgery

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Insertion of an arterial catheter is essential in open heart surgery. The femoral artery is one of the most commonly used arteries in open heart surgery in dogs. Although peripheral arteries, such as the dorsalis pedis, have been used in veterinary medicine in recent years, little is known about their use for open heart surgery in dogs. Therefore, we compared it with conventional management via the femoral artery.

We retrospectively reviewed cases involving mitral valve repair with cardiopulmonary bypass between September 2018 and November 2018. We compared successful catheter placement, catheter insertion, removal time, adverse events, and operation time in the femoral and peripheral arteries.

The study included 104 dogs. Five (4.8%) underwent catheter insertion into the femoral artery (group F), and 99 (95.2%), into either the dorsalis pedis (n = 96) or caudalis median (n = 3) peripheral arteries (group P). Group F required a median 8 min (range 6-13 min) for catheter insertion and a median 18 min (range 10-29 min) for catheter removal. Three dogs in group F bled after heparin infusion during surgery. There was no bleeding in group P, and the pulse was palpable immediately after surgery. Good pressure waveforms were obtained

during all perioperative periods, and blood collection was uneventful. There were no adverse events after surgery in both groups. Operation time did not differ significantly between the two groups.

These results suggest that perioperative management with peripheral arteries in open heart surgery is effective and safe.

Disclosures

No disclosures to report.

ESVC-P-6

Effects of in-hospital diuretic therapy on electrolytes concentration, renal function and survival in 85 dogs with acute congestive heart failure

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Critically hill patients with acute congestive heart failure (CHF) may often show haemoconcentration, dysnatremia, dyskalemia and increased azotemia, due to aggressive diuretic therapy. Haemoconcentration is associated with lower risk of mortality, while dysnatremia and dyskalemia are associated with higher mortality in human medicine. The aim of this study was to retrospectively evaluate the impact of in-hospital diuretic therapy for CHF on selected laboratory parameters and long-term mortality.

Dogs with clinical and radiological evidence of CHF confirmed by echocardiography were included. Blood samples collected throughout the hospitalization at presentation (T0) and discharge (T1) were: venous blood gas analysis (VBGA), serum creatinine (sCr), blood urea nitrogen (BUN), microhematocrit (Htc) and total proteins (TP). Length of hospital stay, ACVIM class and other clinical indices were recorded. Haemoconcentration was defined as a simultaneous increase in Htc and total protein.

A total of 85 dogs (45 male and 40 female; mean age 11.07 $\pm$ 2.54 years; mean weight 8.86 $\pm$ 6.92 kg) were included. Thirty-six dogs had previous episodes of CHF. Mean length of in-hospital stay was 31.15 $\pm$ 17.35 hours. Treatment protocol included a single furosemide endovenous bolus at 2 mg/kg followed by multiple 1 mg/kg bolus/hour until respiratory rate reach 40respiratory rate. Each dog received 8.6 $\pm$ 2.8 mg/kg and 11.1 $\pm$ 2.9 mg/kg furosemide in 24 and 48 hours respectively. Ten dogs received higher furosemide doses or torasemide bolus.

Haemoconcentration was reached in the 33% of dogs. Considering the VBGA and biochemistry results, the number of dogs showing extra-range values (T0-T1) were respectively: hyponatremia (10-23), hypernatremia (13-17), hypokalemia (18-30), hyperkalemia (10-10), hypocloremia (46-61), increased BUN (26-34), increased sCr (3-8). Fourty-one dogs experienced cardiac death, 12 during hospitalization, the remaining dogs between 3 and 721 days after admission.

Stepwise backward regression demonstrated haemoconcentration (HR 0.33) and disnatremia (HR 2.85) influence over outcome. Statistically significant correlation (Pearson) was seen between furosemide dose and kalemia (r = –0.32, P = 0.014) and between BUN and sCr (r = 0.27, P = 0.021). No correlation was seen between furosemide dose and the variables sCr, BUN, Htc and between sCr and Htc.

In conclusion, haemoconcentration and disnatremia affected the outcome in dogs with CHF. Haemoconcentration was associated with lower risk of mortality and had to be considered a target in CHF therapy. In-hospital diuretic therapy increased electrolyte disorder due to loop diuretics inhibition of the renal Na, K, Cl cotransporter in the Henle's loop and disnatremia was a risk factor for adverse outcome. Diuretics doses and haemoconcentration didn't play a direct role in inducing renal disfunction.

Disclosures

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ESVC-P-7

Platelet proteomic profile in dogs with heart failure

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Heart failure can cause haemostatic complications due to platelet over- activation in human and veterinary medicine, especially in cats with hypertrophic cardiomyopathy. There is not enough information on the molecular relationship between hemostasis and cardiovascular diseases in dogs. Thus, in this study, it was aimed to elaborate hemostasis (coagulation) changes with the platelet proteomic profile in dogs with naturally occurred heart failure.

The material of this study consisted of totally 20 dogs with different breed, age, and sexes. Two different group were designed; control (n = 10) and test groups (n = 10). Based on the physical, laboratory and cardiologic examination results, dogs were included into control (healthy) group, or dogs with stage C of heart failure according to ACVIM classification were enrolled into test group. Platelet isolations were performed from each dog, and platelet pellets were stored –80 C until analysed. Platelet proteomes were identified by use of UPLC-ESI/QTOF/MS method. Global clotting times (PT and aPTT) were measured, as well.

Platelet proteomes (n = 107) were matched with the previously described proteins for *Canis lupus familialis*, and compared to the control, at least P < 0.05 level and 1.2-fold change of proteomes (n = 10) was considered statistically significant. Compared to control group, of platelet proteomes, guanine nucleotide-binding protein subunit alpha-11, apolipoprotein C-III, apolipoprotein A-II and clusterin levels increased whereas CXC-motif chemokine-10, cytochrome-C-oxidase subunit-2, cathepsin-D, serine/threonine - protein phosphatase PP1-gamma catalytic subunit, creatine kinase B-type and myotrophin levels decreased in testgroup. PT and aPTT values in the test group were longer than the control values (P < 0.05).

As a result, each platelet protein identified in the present study could be a potential biomarker in the diagnosis of heart failure and therefore these proteins need validation with field studies. Also this proteomes provide detailed information of heart failure process.

Disclosures

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ESVC-P-8

Vitamin D Status in Cats with Cardiomyopathy compared to Normal Cats

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Low serum 25-hydroxyvitamin D ($25(OH)D_3$) has been associated with cardiovascular (CV) disease in people and dogs. We aimed to determine if $25(OH)D_3$ and its metabolite, 3-epi-$25(OH)D_3$ (3-epi), are lower in cats with cardiomyopathy (CM) vs. normal cats, or if associations with certain clinical variables exist. CM cats (n = 44) were enrolled from patients at the Veterinary Medical Centers of Iowa State (ISU) and Tufts Universities; 38 CM cats had congestive heart failure. Exclusion criteria included hypertension, hyperthyroidism, and clinically relevant systemic (including kidney) disease. Normal cats (N) were recruited at ISU; 44 were age- and sex-matched to CM cats, although 56 normal cats ultimately were enrolled. All cats were eating commercial cat foods. CV exam, blood pressure and echocardiographic data were collected; collected serum was frozen until analyzed.

$25(OH)D_3$ and 3-epi were positively correlated (correlation coefficient, 0.35; $P = 0.004$). However, neither was significantly different between CM and matched-N groups (paired Wilcoxson rank-sum). Linear regression analysis to assess impact of CM status, age, and sex among all enrolled cats showed age was significant to $25(OH)D_3$ ($P = 0.0028$). Statistical modeling including all variables was confounded by multicollinearity; therefore, one-by-one simple linear regression tested for relationships between vitamin D status and 18 clinical variables. Significant relationships were found between $25(OH)D_3$ and age, survival time, azotemia category, left atrial enlargement, and left ventricular fractional shortening (FS). After accounting for age, only FS and survival time remained significant with $25(OH)D_3$. We conclude that vitamin D status in cats is not significantly associated with CM.

Disclosures

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ESVC-P-9

Identification of increased desmin aggregates consistent with intermediate filament dysfunction in feline hypertrophic cardiomyopathy

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Desmin, the intermediate filament (IF) in cardiomyocytes is critical for maintaining architecture and mechanical integrity of the contracting cell. Aggregation of desmin and impairment of protein quality control systems including heat shock proteins (HSP) which governs correct IF protein folding are reported in humans and rodent models with hypertrophic cardiomyopathy (HCM). Incorrect folding of desmin leads to its aberrant accumulation within cardiomyocytes. This study aimed to characterise the expression and localisation of desmin, and its chaperone the HSP αB-crystallin in feline HCM.

Residual left ventricular (LV) tissues from necropsy (5 normal and 5 HCM cats) were used for immunoblotting of desmin and αB-crystallin normalised to GAPDH. Fluorescent immunohistochemistry was performed on transverse sections of formalin-fixed and paraffin-embedded LV from another 4 normal and 8 HCM cats. A minimum of 10 images were captured under 40x magnification (5 longitudinal and 5 transverse) were used for fluorescence quantification and protein localisation. Diagnoses were made by histopathology ± clinical assessment. Mann-Whitney's *U* test, or Spearman's rho test were used and the results were expressed as median (range).

On immunoblotting, only one control had a recorded age (5.8 yrs), the other controls were documented as young adult. The median age of HCM cats was 8 yrs (1.7-17). The protein level of desmin was 0.54 (0.27-0.69) in the controls and 1.41 (0.59-2.19) in the HCM group ($P = 0.0159$). The protein level of αB-crystallin was 0.56 (0.18-0.66) in the controls and was 1.46 (0.63-2.39) in the HCM group ($P = 0.0317$). The protein expression of αB-crystallin and desmin was highly correlated ($R = 0.8788$, $P = 0.0016$). On immunohistochemistry, the median age was 3 yrs (1.6-5.8) in control cats and 8.5 (2-17.9) in the HCM group ($P > 0.05$). In controls, desmin appeared as stripes at Z-bands and intercalated discs and a scarce amount of perinuclear aggregates were observed in the cross-section of cardiomyocytes close to epicardium. In HCM cats, dissociation of desmin from the intercalated discs was a frequent finding and clumps of aggregates were seen in the transversely sectioned cardiomyocytes not limited to the periphery of epicardium. Averaged mean intensity of fluorescence was 95.2 (76.6-116.3) in the HCM group and 70.7 (64.7-81.1) in the control group ($P = 0.0056$).

In HCM cats, the IF desmin is structurally disorganised which might compromise the integrity of contractile apparatus. The presence of desmin aggregates suggests that the protein quality control measures failed to restore proteostasis in the HCM affected heart despite the corresponding increase in the chaperone protein αB-crystallin.

Disclosures

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ESVC-P-10

Prevalence and risk factors for atrial fibrillation in dogs with myxomatous mitral valve disease

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Atrial fibrillation (AF) is a common canine supraventricular arrhythmia usually observed in large breed dogs with cardiac disease associated with left atrial enlargement. Although some epidemiologic data and risk factors for AF have been reported, no study has thoroughly

examined the prevalence and risk factors for this arrhythmia in dogs with myxomatous mitral valve disease (MMVD). The aims of this study were to estimate the prevalence of AF in a large population of dogs with MMVD and to identify the risk factors for AF development in these animals.

The medical databases of three Veterinary Teaching Hospitals were retrospectively reviewed. Inclusion criteria were a diagnosis of MMVD after complete cardiovascular assessment (ie., physical examination, thoracic radiography and trans-thoracic echocardiography) and cardiac rhythm assessment via routine 2 minutes ECG and/or good quality ECG tracing during echocardiographic examination of at least 20 minutes' duration. For dogs with multiple examinations during the observing period only data of the most recent exam were considered. Selected clinical and echocardiographic parameters were compared using univariable and different multivariable logistic regression models.

A total of 2194 dogs were enrolled, including 1280, 588, 290, and 36 dogs in ACVIM stage B1, B2, C, and D, respectively. Pulmonary hypertension (PH) was diagnosed in 526 (23.9%) dogs. Atrial fibrillation was diagnosed in 59 dogs with a prevalence of 2.7%. Univariate analyses showed that mixed breed, male gender, decompensated ACVIM stage, left atrial diameter-to-aortic ratio (LA/Ao) > 1.6, normalized left ventricular diastolic diameter > 1.7, and presence of PH were significantly associated with the development of AF. Also the continuous variables LA, body weight (BW), fractional shortening (FS), and velocity of trans-mitral E wave (E-max) were significantly different ($P < 0.01$) for AF cases. After evaluation of autocorrelation and/or interaction between predictors, two multivariable models were obtained. LA/Ao (odds ratio [OR] 14.011, 7.463-26.304), E-max (OR 2.204, 1.192-4.076), BW (OR 1.094, 1.058-1.130), and FS (OR 0.899, 0.865-0.934); and LA (OR 5.28, 3.377-8.092), decompensated ACVIM stage (OR 4.922, 1.481-16.353), and FS (OR 0.919, 0.881-0.959) were significant predictors of AF for model 1 and 2, respectively. An LA > 3.45 cm had sensitivity and specificity of 98.3% and 89.8% to predict development of AF.

Atrial fibrillation is an uncommon complication of canine MMVD and is significantly associated with the more advanced stages of the disease. Increased LA dimension and BW, and decreased systolic function are associated with development of AF in dogs with MMVD.

Disclosures

No disclosures to report.

ESVC-P-11

Left atrial volume assessment in 160 Cavalier King Charles Spaniels with and without degenerative mitral valve disease (2017-2019)

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Degenerative mitral valve disease (DMVD) is the most common acquired heart disease in small-sized dogs with a high predisposition of the Cavalier King Charles Spaniel (CKC) breed. Echocardiographic assessment of the disease is based on the evaluation of atrial and ventricular dimensions, with left atrial (LA) diameter measurement being one of the strongest predictors of clinical outcome. The LA diameter is usually evaluated on the right parasternal transaortic short-axis view and compared to the aortic (Ao) diameter (LA:Ao ratio). However, LA dilation can develop in medio-lateral, cranio-caudal, or ventro-dorsal directions. Therefore, the LA:Ao ratio may not be reliable for the early detection of LA dilation. Measurements of LA volumes have been recently recommended in the dog using monoplane and biplane Simpson's modified methods of discs (SMOD) and area-length methods (ALM). The objectives of this prospective study were therefore to 1) compare different echocardiographic methods in evaluating LA volume in a large population of CKCs and 2) to assess LA volume according to DMVD severity.

The study population consisted in 160 CKCs either healthy or affected by DMVD (median weight = 9.15 kg [interquartile range (IQR) = 7.8-10.3], male-to-female ratio = 0.95). According to the ACVIM classification, 28/160 dogs (17.5%) had no identifiable DMVD lesions (stage A), 86/160 (53.8%) and 22/160 (13.8%) were in stages B1 and B2, respectively, 23/160 (14.3%) had past or current congestive heart failure (CHF, stage C), and 1/160 (0.6%) had refractory CHF (stage D). Dogs for which mitral regurgitation (MR) was adequate for quantification by the Proximal Isovelocity Surface Area method (n = 107) had a median regurgitation fraction (RF) of 38% [23-55].

The monoplane SMOD and ALM using the left apical 4-chamber view overestimated LA volume by comparison with the biplane ALM (+2.6% and + 10.8%, respectively), and the monoplane ALM overestimated LA volume in comparison with the monoplane SMOD (+8.3%). Left atrial volumes significantly increased with ACVIM stages. Among B1 dogs, end-systolic LA volume assessed by the biplane ALM was significantly lower in dogs with mild MR (RF < 30%, median volume = 0.75 mL/kg [IQR = 0.59-0.90]) than for dogs with higher MR (RF≥30%, median volume = 1.39 mL/kg [IQR = 0.96-1.56]; $P < 0.01$).

In conclusion, this study demonstrates the importance of LA volume measurement in CKCs with DMVD, especially among B1 DMVD dogs. These results suggest that a category of B1 dogs (with RF > 30%) actually show LA dilation, which is however not apparent using the LA:Ao ratio calculation, thus potentially leading to a misdiagnosis between DMVD B1 and B2 stages.

Disclosures

Disclosures to report.

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ESVC-P-12

Hematological abnormalities in dogs with congenital arterial stenosis: a prospective study of 56 cases (2017-2019)

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Intravascular hemolysis has been identified in human patients with moderate to severe aortic stenosis. The underlying mechanism implies shear stress exerted on erythrocytes by high velocity flows through the stenotic orifice. No similar data are currently available in veterinary medicine. The aim of this prospective study was therefore to 1) document hematological abnormalities in dogs with arterial stenosis (ie, aortic stenosis [AS], pulmonic stenosis [PS]) between 2017 and 2019 and 2) evaluate if maximal and mean Doppler-derived trans-stenotic pressure gradients (ΔP), as well as features of stenotic lesions (number and location), were associated with erythrocyte abnormalities.

The study sample consisted of 56 dogs (median age = 1.6 year [interquartile range (IQR) = 0.7-4.0 years], male-to-female ratio = 1.5). The most commonly recruited breeds were French Bulldogs (n = 13), White Swiss Shepherds (n = 6), English Bulldogs (n = 4), Boxers (n = 4), Golden retriever (n = 3), Chihuahua (n = 2), and 24 other breeds (n = 1 for each). Among the study population, 44 dogs (79%) had PS and 12 (21%) had AS. The median maximal ΔP values were 161 mmHg [110-215 mmHg] and 144 mmHg [IQR = 125-176 mmHg] for PS and AS, respectively. Included dogs showed 1 (34/56), 2 (16/56) or 3 (6/56) obstructive lesions: subvalvular stenosis for 24/56 dogs (13/44 with PS, 11/12 with AS), valvular stenosis for 42/56 dogs (37/44 with PS, 5/12 with AS), and supravalvular stenosis for 16/56 dog (only PS).

Hematological abnormalities were detected in most dogs (n = 49; 88%), with schizocytes found in 28/56 (50%) dogs (median proportion = 1‰ cells [IQR = 0-3‰]), acanthocytes in 46/56 (82%) dogs (median proportion = 45‰ cells [IQR = 10-226‰]), and hemolytic anemia in 4 dogs with PS (hemoglobinemia <12.4 g/dL). No significant association was identified between these abnormalities and the above-mentioned echocardiographic parameters. Three out of the 4 dogs with hemolytic anemia had a maximal ΔP > 200 mmHg (242 to 412 mmHg). Interestingly, the dog with the highest maximal ΔP also had the most severe anemia and schizocytosis, and both abnormalities decreased, and then resolved, after balloon valvuloplasty.

In conclusion, these results show that red blood cell abnormalities (acanthocytosis and schizocytosis) are very common in dogs with congenital arterial stenosis, thus suggesting that turbulent blood flows through stenotic orifices induce mechanical erythrocyte damage. Further prospective studies are needed to better document these findings and elucidate the precise mechanisms of red cell damage using other hematolytic markers (eg, lactate dehydrogenase, hemosiderinuria, etc.), as performed in human patients.

Disclosures

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ESVC-P-13

Use of torasemide in cats with congestive heart failure: 17 cases (2016-2019)

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Torasemide is a loop diuretic whose safety and efficacy have been demonstrated in dogs with congestive heart failure (CHF). Torasemide is characterized by a longer duration of action, a more potent diuretic action, and a higher bioavailability than furosemide. However, to the best of our knowledge, no study has focused on the efficacy and safety of torasemide in cats with CHF. The objectives of this retrospective study were therefore to 1) describe the clinical and echocardiographic characteristics and document the clinical outcome of cats with CHF treated with oral torasemide, and 2) identify potential adverse events related to torasemide administration in this feline population.

The case records of cats treated with torasemide were reviewed. The study population consisted of 17 cats (median age = 10.6 years [interquartile range (IQR) = 6.4-11.0], male-to-female ratio = 2.4), with a majority of Domestic shorthair cats (13/17). All cats presented dyspnea related to CHF (pleural effusion [4/17], pulmonary edema [6/17] or both [7/17]), associated with ascites in 2/17 cats. The cause of CHF was determined in all cats by echocardiography: hypertrophic (8/17,47%), restrictive (3/17,18%), dilated (3/17,18%) and arrhythmogenic right ventricular (2/17,12%) cardiomyopathy, and aortic valve congenital abnormality (1/17,5%). The left atrium (LA) was dilated in all cats, with a median end-diastolic LA-to-aorta-ratio of 1.97 [IQR = 1.80-2.11, normal values <1.2]. Median torasemide dosage at initiation was 0.20 mg/kg/day [IQR = 0.17-0.23].

Follow-up was available for all cats. Torasemide dosage was increased for 41% (7/17) cats due to the persistence of CHF signs (median time from treatment initiation to dosage change = 7 days [7-32]; median dosage = 0.26 mg/kg/day [0.19-0.34]). Additionally, furosemide was added 12 hours after torasemide intake for 29% (5/17) of cats (median time from treatment initiation to furosemide initiation = 15 days [1-44]; median dosage = 1.13 mg/kg/day [1.09-1.35]). Other treatments included benazepril (4/17), pimobendane (4/17), clopidogrel (12/17), aspirin (3/17) and spironolactone (3/17). Regression of clinical signs was observed in most cats (16/17), with no remarkable adverse events. Death was reported in 8/17 cats, with 6/8 euthanized as a result of CHF worsening (n = 4) or aortic thromboembolism (n = 2), and death related to CHF for 2/8 cats. Median survival time after torasemide prescription was 96 days [19-330].

In conclusion, to the best of our knowledge, this is the first description of torasemide use in cats with CHF. This drug was well tolerated in all cases. This case series illustrates the therapeutic interest of torasemide in cats, which needs to be confirmed by further prospective clinical trials.

Disclosures

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ESVC-P-14

Use of torasemide as a second line diuretic in dogs with congestive heart failure

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Torasemide is a loop diuretic used in dogs with congestive heart failure (CHF) as an alternative to furosemide. Studies comparing furosemide and torasemide in dogs with refractory CHF are sparse.

Medical database of our Teaching Hospital has been reviewed searching for dogs with myxomatous mitral valve disease who experienced CHF and received torasemide as a second line diuretic after treatment failure with furosemide (study group: 25 dogs). Clinical, echocardiographic, radiographic and laboratory findings have been annotated. Survival time has also been reported and calculated from the first episode of CHF. A control group (23 dogs) of dogs with similar cardiac disease, that never received torasemide has also been selected. Data were collected at the time of first CHF (first examination) and in concomitance with the last examination available.

Sex distribution was equal between the two groups ($P = 0.250$). In both groups the mixed breed was overrepresented. Besides furosemide and torasemide, other medications used to control the cardiac disease were comparable. At admission there was no difference between groups regarding age ($P = 0.956$), body weight ($P = 0.543$), left ventricular diastolic internal diameter normalized for body weight (LVIDDn) ($P = 0.331$), left atrial to aortic ratio (LA:Ao) ($P = 0.959$), tricuspid regurgitation peak velocity ($P = 0.838$), creatinine ($P = 0.433$) and potassium ($P = 0.230$) serum levels. There was no difference in prevalence of atrial fibrillation between groups ($P = 1.000$). The overall number of decompensations experienced by each dog was higher for the study group (4.4 ± 1.9) compared to the control group (2.9 ± 1) ($P = 0.001$). At last examination only LA:Ao was higher in the study group compared to controls ($P = 0.035$). The total maximal dose of furosemide was 6.1 ± 3 mg/kg/day for the study group and 5.6 ± 2.1 mg/kg/day for the control group ($P = 0.475$). While the initial dose of torasemide used was 0.7 ± 0.4 mg/kg/day. When comparing the first and last examinations within the two groups, body weight reduced in the study group ($P < 0.001$), while LA:Ao ($P < 0.001$), and creatinine ($P < 0.001$) increased. The number of decompensations recorded before (2.9 ± 1.1) and after (1.5 ± 1.5) torasemide initiation reduced significantly ($P = 0.001$). Within the control group only creatinine increased ($P = 0.013$). Thirty-six dogs died of cardiac related causes. There was no difference in survival between the two groups ($P = 0.413$), with a median survival of 523 days for the study group and of 383 days for the control group.

Torasemide appears effective as a second line diuretic for CHF control in dogs with myxomatous mitral valve disease, offering a life expectance comparable if not superior to furosemide alone.

Disclosures

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ESVC-P-15

Echocardiographic predictors of first onset of atrial fibrillation in dogs with myxomatous mitral valve disease

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Atrial fibrillation (AF) occurs in dogs with myxomatous mitral valve disease (MMVD) as a consequence of left atrial (LA) dilatation. Predicting its occurrence might have beneficial consequences. This is a retrospective study evaluating the usefulness of echocardiography in predicting the first occurrence of AF in dogs with MMVD.

The medical databases of two Veterinary Teaching Hospitals were reviewed searching for dogs with MMVD that developed AF during the following year (study group). The last echocardiographic examination obtained during sinus rhythm was used to derive selected variables used for successive statistical comparison. For each dog with AF a control dog, matched for body weight (BW), class of heart failure, and LA dimension as expressed by the ratio between LA and aortic diameters (LA:Ao), but never developing AF over a comparable follow up period was selected. All echocardiographic exams were reviewed by a single board certified cardiologist and several variables of LV and LA dimension and function, and LA volumes were obtained. In particular LV internal diameters in diastole and systole were obtained as absolute values and indexed to BW. LA dimensions were expressed as absolute value and LA:Ao, moreover LA volumes were measured during maximal and minimal LA expansion and at the peak of the P wave, and then indexed to BW. Several indexes of LA function were then calculated from these volumes. Peak trans-mitral E and A waves velocities, E:A ratio, and A wave duration were also measured. LA speckle tracking echocardiography (STE) was carried out and peak atrial longitudinal strain (PALS), peak atrial contraction strain, and contraction strain index were measured.

Forty-four dogs with MMVD were included, 22 dogs developing AF and 22 dogs maintaining a sinus rhythm. There was no difference in terms of BW ($P = 0.803$), sex distribution ($P = 0.393$), and class of heart failure ($P = 0.550$). Among the tested echocardiographic variables only LA diameter ($P = 0.034$), and LV internal diameter in diastole not indexed for BW ($P = 0.031$) differed significantly between groups. There was no difference in terms of LA:Ao ($P = 0.097$), LV internal diameters in diastole and systole indexed for BW ($P = 0.131$ and $P = 0.406$, respectively), and LA volumes and volume-derived functional parameters. Among the STE-derived variables, PALS values differed significantly between AF group (mean value 23.8 ± 8.6) and control group (mean value 30.5 ± 9.6) ($P = 0.027$).

Absolute cardiac dimensions as well as LA STE, and in particular PALS, are useful echocardiographic predictors for the development of AF in dogs with MMVD.

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ESVC-P-16

Usefulness of Holter-derived Lorenz plots analysis to discriminate different cardiac rhythms in dogs

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Lorenz plot (LP) is a representation of heart rate variability that summarizes graphically the beat-to-beat intervals recorded during a Holter

monitoring (HM). The use of LP patterns (LPPs) to diagnose rhythm disturbances has been studied in people, but little is known in animals. The aim of this study was to analyze the graphic features of LPs and to evaluate the diagnostic value of LPPs for identifying arrhythmias in dogs.

HMs with >20 hours of valid data obtained from dogs with sinus rhythm (SR) or different types of tachyarrhythmias were used. One operator blinded to the underlying rhythm diagnosis reviewed the automated analyses in order to: assess HMs quality, manually correct any software misinterpretation, and make the rhythm diagnosis. Cardiac rhythms were classified as SR; SR with frequent (>100) premature ectopic complexes (SR + PEC), either supraventricular or ventricular; atrial fibrillation (AF); and AF with frequent ventricular premature complexes (AF + VPC). For each HM, a LP was generated by the software using all RR intervals. LPs were studied qualitatively and quantitatively and distinct LPPs were created by adapting previously recognized patterns in humans. The diagnostic accuracy of LP analysis in predicting the underlying cardiac rhythm was evaluated by calculating the corresponding sensitivity (Se) and specificity (Sp).

One hundred and nineteen HMs were analyzed including 48 SR, 49 SR + PEC, 4 AF, and 18 AF + VPC. Ten distinct LPPs were identified: comet (10 cases); torpedo (3 cases); Y-shaped (6 cases); diamond (12 cases); diamond with central silent zone (15 cases); a combination of one of the above 5 patterns with a double side-lobe (DSL) (46 cases), a triple side-lobe (2 cases), or a quadruple side-lobe (3 cases); fan (19 cases); and fan with DSL (3 cases). When pooled together as a single normal pattern, the comet, torpedo, Y-shaped, and diamonds, predicted presence of SR with Se and Sp of 91.7% and 97.2%, respectively. Two LPs with a diamond pattern had a Holter diagnosis of SR + PEC. The DSL pattern indicated presence of SR + PEC with Se and Sp of 85.7% and 94.3%, respectively. Triple and quadruple side lobe patterns were exclusively associated to SR + VPC. The fan configuration (considering together the fan and fan with DSL pattern) indicated AF with both Se and Sp of 100%. The three cases of fan with DSL were associated with AF + VPC.

In conclusion, different cardiac rhythms are associated with peculiar LPPs and their analysis holds relevant diagnostic value in dogs with SR and/or tachyarrhythmias.

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ESVC-P-17

Visual Representations of Cardiac Arrhythmias in Dogs using Lorenz Plots

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Lorenz plots (LPs) can be generated from continuous electrocardiographic (ECG) recordings to provide visual representations of rhythm patterning. This study sought to characterize LP patterns for common rhythms of dogs.

Twenty Holter recordings free from pathologic arrhythmias and ten recordings each of supraventricular premature complexes (SPVCs), complex supraventricular ectopy, ventricular premature complexes (VPCs), complex ventricular ectopy, atrial fibrillation (AF), high-grade second degree atrioventricular block (AVB) and paced rhythms were retrospectively evaluated and utilized for one-hour LP generation. Beat origin was color coded. Patterns found in each arrhythmia group were described, and arrhythmia numbers and LP shape measurements were reported.

Normal Holter recordings uniformly showed a torch morphology with variable silent zones. Premature beats were associated with double and triple side lobe patterns, with variations resulting from multiple coupling intervals and variable post-ectopic pauses. Complex ectopic rhythms were evidenced by a small data point concentration in the lower left corner of the LP. Recordings with AF uniformly showed a fan pattern consistent with random atrioventricular nodal conduction, and recordings with AVB showed island patterns consistent with variable atrioventricular nodal conduction. Paced rhythms were torpedo shaped when the rhythm was completely controlled by the pacemaker.

Specific LP patterns were identified for common cardiac rhythms in dogs which supports non-random mechanisms for most rhythms. Incorporation of LPs in arrhythmia interpretation may aid mechanistic understanding, which in turn may advance the understanding of, and approach to diagnosis and treatment of arrhythmias in dogs.

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ESVC-P-18

First case of successful transcatheter pulmonary valve implantation in a dog with severe pulmonary regurgitation

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Transcatheter pulmonary valve implantation is a therapeutic approach, approved by the US Food and Drugs Administration in 2010 for human patients with failing pulmonary conduits and for failing bioprosthetic surgical pulmonary valves in 2017. We report here the first case of successful transcatheter implantation of a stented valve in a pulmonary position in a dog with congenital pulmonary valve disease. A 3-year-old, 10.9 kg, client-owned Beagle dog was referred for a follow-up visit after a percutaneous balloon valvuloplasty performed 22 months before for correcting a severe type A valvular pulmonic stenosis (Doppler-derived peak trans-stenotic pressure gradient, DP = 348 mmHg before the procedure, 66 mmHg 24 hours later). At time of presentation the dog was lethargic, and echocardiography revealed a mild pulmonic stenosis (DP = 43 mmHg) associated with severe pulmonary regurgitation (proximal width ratio assessed by color-flow Doppler mode of 100%), and secondary major right

ventricular and right atrial dilation. Despite medical therapy, worsening of right heart dilation was observed two months later, and a transcatheter pulmonary valve implantation using a Melody™ valve with a pre-stenting system was decided. A contrast-enhanced cardiac-gated computed tomography 3D scan was performed one week before surgery, to accurately assess the pulmonary and coronary artery morphology, and perform measurements of the pulmonary annulus and the right ventricular outflow tract maximal diameters. Aneurysm of the pulmonary trunk with plications of the arterial wall was confirmed. The initial minimally invasive approach via the left jugular vein with a Seldinger technique was unsuccessful due to the size of the outer diameter of the delivery system and therefore was converted to a left 3rd intercostal thoracotomy and a trans-ventricular delivery (the approach had been prepped at the same time as the neck area in case of conversion). The dog recovered uneventfully and was discharged 10 days after the procedure. Right heart dilation disappeared within 15 days. The dog is still doing well three months after valve implantation. This case illustrates that pulmonary transcatheter stented valve implantation is technically feasible in the dog with severe pulmonary valve disease. Stented valves as an alternative to open-heart surgery is the source of tremendous development in human medicine and it can be foreseen that veterinary cardiology will, in some way, benefit from this revolution.

Disclosures

Disclosures to report.

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ESVC-P-19

Normal aortic annulus dimensions in Boxer dogs according to sex and body weight

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Boxer dogs breeding in Italy is regulated by Boxer Clubs. The Boxer Club of Italy (BCI) drafted the guidelines for the "selection" of dogs with morphological and attitudinal standards. Given the high prevalence of congenital heart diseases in Boxer dogs the BCI established a cardiovascular screening mandatory for the admission to the selection process. Screening data have been collected by either board-certified cardiologist or operators, selected through a practical exam after a training. During this exam different operators examined the same Boxer and their findings were compared with those of the board-certified (inter-observer variability) and were compared also three measurements of the same dogs performed by each operator (intra-observer variability). Current reference intervals for aortic annulus dimensions do not account for body weight (BW).

The objective of this study is to analyse the aortic annulus dimensions based on sex and BW of dogs.

Four thousand two hundred one Boxer dogs free from cardiovascular diseases were included in the study.

Cardiovascular screening conducted between 12/11/1999 and 09/03/2018 were included. Two-dimensional, M-Mode, spectral and color flow Doppler transthoracic echocardiography (TTE) is performed following the published recommendations. Sixteen dogs were excluded from the analysis due to the lack of data on sex and 119 for the annulus less than 15 mm, remaining for the analysis a sample of 4066 dogs. These dogs have been divided into males and females and into weight quartiles. Subsequently the normal values and confidence intervals for annulus were analysed for the entire sample, by gender, by weight quartiles and by sex and weight interaction.

Regarding the results we have objectified that aortic annulus dimensions increased with increasing BW. The values obtained were tested by sex using a *t*-test while for weight quartiles or by sex and weight interaction with the ANOVA test (with the Bonferroni correction for multiple comparisons): in all cases the significance level p is lower than 0.001.

As for the cases excluded from the analysis by measure of the annulus less than 15 mm, they are mostly females (84.9%, $P < 0.001$) and an average weight lower than 4.1 kg (SE 0.40, $P < 0.001$) compared to cases analysed.

In conclusion we can say that BW based 95% confidence interval may help in screening dogs for heart disease, discriminating normal aortic annulus dimension.

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ESVC-P-20

Inflammatory and oxidative stress markers are associated with survival in canine cardiovascular patients

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Various factors can influence survival of dogs with mitral valve disease (MVD) and dilated cardiomyopathy (DCM). In canine cardiovascular patients, inflammatory and antioxidant markers have not been investigated in terms of association with survival, yet. Thus, we investigated the association of inflammatory (white blood cell (WBC), neutrophil (NEUT) and monocyte (MONO) counts, C-reactive protein (CRP), tumour necrosis factor-alpha (TNF-α) and interleukin-6 (Il-6)) and oxidative stress (malondialdehyde), vitamin E and glutathione peroxidase (GPX)) markers with survival in dogs with MVD and DCM.

Thirty-seven dogs with MVD (21) and DCM (16) (11 dogs in International Cardiac Health Council Class [ISACHC] I, 7 dogs in ISACHC II and 19 dogs in ISACHC III) were included in the study (2 dogs were censored). Survival time was counted from the day of admission when the blood samples were collected to the day of death or euthanasia. Markers were analysed using Cox proportional-hazards models. Hazard ratios (HR), 95% confidence intervals (CI) and corresponding P values were calculated. A value of $P < 0.05$ was considered significant.

A log-rank test was performed on survival time with respect to disease type (MVD or DCM). No significance was found ($P = 0.37$), although dogs with MVD (median = 452 days) had on average longer survival time than those with DCM (median = 184 days). Univariate Cox proportional-hazards models were performed for every marker.

The following markers were significantly associated with survival (HR; 95% CI; P value): WBC (1.139; 1.049, 1.237; 0.002), NEUT (1.184; 1.074, 1.305; 0.001), MONO (7.502; 2.044, 27.532; 0.002), CRP (1.000; 1-000, 1.000; 0.008), TNF-α (1.078; 1.018, 1.142; 0.010), Il-6 (1.009; 1.000, 1.017; 0.039) and GPX (0.992; 0.986,0.999; 0.025). If we interpret some of the results: an increase of NEUT for 1 unit ($x10^9$/L) increased the risk of death by 18.4%, while an increase of GPX (U/g of haemoglobin) for 50 units decreased the risk of death by 33%. Furthermore, Cox models were fitted, with ISACHC and an additional marker as covariates, since survival was significant with respect to ISACHC class (log-rank test, $P = 0.0004$). In these models, the effect of the above markers decreased; however, GPX (0.992; 0.985,0.999; 0.025) and NEUT (1.122; 1.007,1.249; 0.036) remained significantly associated with survival.

These results suggest that increased inflammation and decreased activity of antioxidant enzyme GPX are associated with decreased survival in canine MVD and DCM patients, even when ISACHC is considered in the model. We may conclude that selected inflammatory and oxidative stress markers predict survival.

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ESVC-P-21

Right heart remodelling in brachycephalic obstructive airway syndrome

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Brachycephalic Obstructive Airway Syndrome (BOAS) is characterised by various upper airway abnormalities that could potentially trigger remodelling of the right heart. The aim of this study was to evaluate echocardiographic characteristics in symptomatic (BOAS) and asymptomatic French Bulldogs (FB) and Pugs. Dogs were diagnosed as BOAS according to clinical signs and anatomical abnormalities.

Fifteen FB (7 BOAS, 3F/4M, 8 asymptomatic, 8F) and 10 Pugs (7 BOAS, 2F/5M, 3 asymptomatic, 3F) underwent complete echocardiographic examination of the left and right heart according to guidelines. In each breed, echocardiographic parameters of dogs with clinical signs of BOAS were compared to asymptomatic dogs. Furthermore, BOAS FBs were compared to BOAS Pugs. Weight-depended variables were indexed (variable/weight$^{1/3}$). Normally distributed variables were compared with independent *t*-test and for not normally distributed data Mann-Whitney test was used. Statistical significance was defined as $P \leq 0.1$.

There were no significant differences in age and weight between BOAS FB and asymptomatic FB. BOAS FB had significantly lower vena cava collapsibility index ($P = 0.013$), larger right ventricular internal diameter in mid cavity (RVIDmid) ($P = 0.056$), larger indexed RVIDmid ($P = 0.098$), higher tricuspid valve (TV) E wave velocity ($P = 0.083$), and lower peak systolic tricuspid annular velocity (St) ($P = 0.037$) compared to asymptomatic FB. There were no statistical differences between asymptomatic and BOAS Pugs in age, weight and echocardiographic parameters. No significant difference in weight between FB and Pugs was found; however BOAS FBs were younger ($P = 0.067$). BOAS FBs comparing to BOAS Pugs showed lower collapsibility index ($P = 0.002$), larger indexed right ventricular longitudinal internal diameter (RVIDlong) ($P = 0.084$), larger indexed right ventricular area (RVA) in diastole ($P = 0.022$), larger indexed RVA in systole ($P = 0.027$), larger indexed right atrial area ($P = 0.023$), larger indexed LVIDd ($P = 0.048$), higher mitral annular plane excursion (MAPSE) ($P = 0.011$), higher mitral valve (MV) E velocity ($P = 0.013$), higher MV A wave ($P = 0.084$), higher TV A ($P = 0.099$) and higher peak systolic annular velocity of left free wall Em ($P = 0.047$).

In BOAS FB right ventricle was larger compared to asymptomatic FB, which suggests remodelling of the right heart. Higher TV E velocity might suggest higher right atrial pressure and lower St might suggest decreased right ventricular systolic function in BOAS FB. Lower vena cava collapsibility index in BOAS FB compared to asymptomatic FB, might be due to higher right atrial pressure in BOAS dogs. Breed specific reference ranges are recommended for evaluation of the right heart. Echocardiographic differentiation of BOAS and asymptomatic brachycephalic dogs seems challenging.

Disclosures

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ESVC-P-22

Effect of a single dose of Pimobendan on right ventricular and atrial function in healthy cats

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Pimobendan is an inodilator widely used in canine cardiology. It increases life expectance in dogs with heart failure. Moreover it is able to enhanced right ventricular (RV) systolic function in healthy and diseased canine patients. Only a few studies are focused in evaluating the effect of pimobendan in cats, and no report exist regarding its effect on the right heart. The aim of the present study was to analyze the changes of RV and right atrial (RA) echocardiographic parameters in healthy cats after a single oral dose of pimobendan.

Eleven apparently healthy cats were used for this study. Cats were deemed to be healthy on the basis of clinical examination, cardiac auscultation, non-invasive blood pressure measurement, blood work, and conventional echocardiography (including RV wall thickness in diastole, RV internal diameters in diastole and systole from a short axis view, RV fractional shortening (FS), RV areas in diastole and systole from an apical view, fractional area change, tricuspid annular plane systolic excursion, tissue Doppler imaging derived tricuspid annulus systolic wave velocity, RA maximal and minimal diameters, and RA FS). Cats were scanned following standard techniques while gently manually restrained. Each cat was scanned a total of eight times, at different time points in two subsequent days. The first day cats were scanned at time 0, and after 1, 3, and 6 hours. The following day, each cat received an echocardiography before (time 0), and 1, 3, and 6 hours after a single dose of 1.25 mg of pimobendan administered orally.

None of the cats showed any adverse reaction to the drug. Data obtained from different time points before (day 1 at time 0, 1, 3, and

6, and day 2 at time 0) and after (day 2 at time 1, 3 and 6) pimobendan administration were pooled together. The statistical comparison was then performed between two global time points (before and after pimobendan administration).

Among the tested variables, some differed before and after pimobendan administration. In particular heart rate (P = 0.002), RV FS (P = 0.011), RV fractional area change (P = 0.010), and tissue Doppler imaging derived tricuspid annulus systolic wave velocity (P = 0.014) increased significantly. On the other side, RV internal diameter in diastole (P = 0.019) and systole (P = 0.002), and RA maximal (P = 0.004) and minimal (P = 0.002) diameters were reduced after pimobendan administration.

Pimobendan appears safe when administered to healthy cats and it is able to induce a significant increase in RV and RA systolic properties.

Disclosures

No disclosures to report.

ESVC-P-23

Ambulatory electrocardiography and serial cardiac specific troponin I measurement in twenty-two dogs envenomated by the European Adder (Vipera berus)

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Envenomation by the European adder (*vipera berus*), is a common seasonal presentation in small animal practice in Norway. Cardiac arrhythmias are observed in patients but studies describing their time course and severity are limited. Cardiac auscultation and short in-hospital electrocardiograms (ECGs) are insensitive for the detection of arrhythmias and as such, many may go undetected.

The aim of this study was to describe the prevalence and nature of arrhythmias detected in dogs during the first forty-eight hours post envenomation and to investigate associations between arrhythmia grade, serum cardiac specific troponin I (cTnI, a marker of myocardial cell injury) and snakebite severity score (SS score) at presentation.

Twenty-two dogs bitten by *vipera berus* were included in this prospective cohort study. An ambulatory ECG (AECG) was placed on each dog at presentation, for 24-54 hours. Arrhythmia grades of 0-3 were assigned based on frequency and severity of arrhythmia during AECG recording (modified from previously described grading systems). Serum was obtained at presentation, 12 h, 24 h, 36 h and 14 days post bite, for cTnI analysis. An SS score of 1-3 was recorded upon admission using a previously described grading system.

Four dogs (18%) had an SS score of 1 (mild) while SS scores of 2 (moderate) and 3 (severe) were observed in fourteen (64%) and four dogs (18%), respectively. Seventeen dogs (77%) had raised cTnI concentrations at a minimum of one time point. Ten dogs (45%) had elevated cTnI at presentation, 12 h, 24 h and 36 h.

Nine dogs had arrhythmia grades of 0 (non-pathological). Thirteen dogs (59%) developed pathological arrhythmias (grades 1-3). All arrhythmias were ventricular in origin. Severe complex ventricular arrhythmias (grade 3) were observed in 6 dogs (27%). All grade 3 arrhythmias persisted into day two.

Concentrations of cTnI at presentation were significantly higher in dogs that developed pathological arrhythmias compared to those that did not (P < 0.05). One dog with a pathological arrhythmia had normal cTnI concentrations at all time points. SS score was not significantly associated with arrhythmia grade, initial cTnI concentrations or peak cTnI on day one.

This study shows that raised cTnI concentration and ventricular arrhythmias are common following *vipera berus* envenomation in dogs and highlights the value of prolonged ECG monitoring of these patients. Dogs that developed pathological arrhythmias could not be differentiated from those that did not, based on SS score. Normal cTnI concentrations did not rule out the development of pathological arrhythmias, in this study.

Disclosures

Disclosures to report.

Unsure if this is relevant: Joanne Harris is a director at Heartvets who provide ECG interpretation and Holter rental services.

ESVC-P-24

E point to septal separation (EPSS): difference of measurement from the right parasternal long axis and short axis view in dogs

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E point to septal separation (EPSS) is the distance of the maximal early diastolic motion of the septal mitral valve leaflet (E-point) to the interventricular septum measured using the M-mode recordings. EPSS is an important index in the diagnosis of occult and symptomatic dilated cardiomyopathy (DCM) as recently reported in the European screening guidelines for DCM in Doberman Pinschers. EPSS can be measured from the right parasternal long axis view (RPLA) or from the right parasternal short axis view (RPSA). However, no previous studies assessed if the EPSS values are different using different echocardiographic views. Therefore, the aim of this study was to compare EPSS values obtained from the RPLA and RPSA views in different canine breeds.

This was a prospective observational study. Dogs were presented to the Istituto Veterinario di Novara for routine screening purposes. All dogs underwent a complete clinical and echocardiographic examination performed in right and left lateral recumbency with a simultaneous ECG tracing. The measurement of the EPSS was obtained from both the RPLA and RPSA views. All measurements were performed offline by the same operator (OD) evaluating 3 cardiac cycles, and the mean values were calculated.

A total of 33 healthy dogs were included: 20 Golden Retriever, 7 Doberman Pinschers, 3 English Bulldogs, 2 Labrador Retriever and 1 Czechoslovakian Wolf. This study included 27 females and 12 males, with a median age of 3 years (range 1-8 years) and a median body weight of 29.5 kg (range 22-42 kg). The mean EPSS measured from the RPLA and RPSA view were 4.0 $\pm$ 0.92 mm and 4.8 $\pm$ 1.2 mm

respectively. EPSS was significantly higher when measured from RPSA than RPLA view ($P < 0.0001$) with a mean difference of 0.85 ± 1 mm. The EPSS measurement obtained from the RPLA and RPSA view might not be used interchangeably. The greater lateral and torsional movements of the heart obtained from the short axis view during respiratory phases and cardiac cycle, might be the reason for the higher values of EPSS obtained from the RPSA view. This finding could be taken into account for an adequate echocardiographic evaluation and diagnosis. Further study with a larger canine population is warranted to confirm this result.

Disclosures

No disclosures to report.

ESVC-P-25

Investigation report of the effect of long flightprolonged air travel on dogs with heart mitral valve disease

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The effect of long air travel on animals with heart disease has not been investigated. In this study, we assessed the changes in physical condition of animals with mitral valve disease before and after flight.
Target case profile: 38 dogs with mitral valve disease who underwent air travel to undergo mitral valve repair at the JASMINE Veterinary Cardiovascular Medical Center between September 2017 and March 2019. Before and after flight events: changes in exercise intolerance, appetite, respiratory rate and frequency of coughing, gastrointestinal signs and presence of syncope were evaluated. Also presence or absence of events related to heart disease post-flying and post-operative discharge rate were studied.
All cases boarded the cabin. Thirty-five patients were able to return home (2 dogs without surgery). Two dogs developed events related to mitral valve disease (1 left atrium rupture and 1 pulmonary edema, respectively). The dog with left atrial rupture died before surgery and she did not receive cardiac medications during the flight. The dog that developed pulmonary edema post-flight also missed a dose during the journey, but responded to medical therapy after the landing. Both of the dogs were in ACVIM classification stage D. About 20% of cases showed changes in physical condition (exercise intolerance, appetite) before and after flight. None of the changes required treatment. Two dogs showed gastrointestinal symptoms (vomiting, diarrhea) before flight. Three patients developed diarrhea after the flight. Among patients who showed respiratory symptoms before and after flight, 38% showed respiratory distress and 19% showed worseing in coughing. There were 5 dogs with syncopal episodes before the flight; however, none of the dogs fainted after the flight. One dog had post-flight syncope..
Although dogs with mitral valve disease may show clinical changes related to air travel, these are only transient and can be alleviated with symptomatic treatment. Post-flight cardiac disease related symptoms can be prevented by managing the medication time with alarm clock or other devices when travelling across timezones. Avoidance of medication errors that may be caused by inexperienced while moving and misreading of the clock due to time difference may be sufficient to avert mitral valve disease related events with sufficient alertness. In conclusion, long distance air travel of patients with mitral valve disease can be safely performed by carrying out routine care and medication adherence.

Disclosures

No disclosures to report.

ESVC-P-26

Clinical, ECG and echocardiographic findings in a canine case series of presumptive myocardial infarction

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Acute myocardial infarction (AMI) is the most common cause of human ischemic heart disease. Its pathogenesis involves atherosclerosis of coronary arteries, platelet activation, thrombosis and vasospasms. Only anecdotal information on canine AMI exists. Postmortem descriptions of arteriosclerosis with myocardial ischemic injury and congestive heart failure are available, but clinical data regarding ante-mortem diagnosis of AMI is lacking. This case series describes possible predisposing factors, clinical, electrocardiographic and echocardiographic findings in dogs with a presumptive diagnosis of myocardial infarction.
The database of 4 veterinary clinics were retrospectively screened for dogs with a presumptive diagnosis of AMI. Suspicion was based on echocardiographic regional wall motion abnormalities (hypokinesia, akinesia or dyskinesia at initial presentation or follow-up), and/or elevated serum cardiac troponin I (cTnI), without any other obvious cause. Thirteen dogs with a presumptive diagnosis of AMI were identified. History, clinical data, cTnI, electrocardiographic and echocardiographic findings were reviewed. Data are expressed as median and range.
Three out of thirteen cases were West Higland White Terriers. Median age was 9 years (1-12) and body weight was 21,8 kg (6,1-40,8). Factors predisposing to thrombosis were identified in 9 dogs (4 postoperative, 3 neoplastic, 1 immune-mediated hemolytic anemia and 1 hypothyroidism). Common clinical findings were arrhythmia (13/13), 1 with normal heart rate, 9 tachycardic and 3 bradycardic; weakness (11/13); and syncope (6/13). cTnI was severely elevated (50.000 ng/L, range: 8.960-221.961) in all tested cases (10/10).
Identified ECG abnormalities were ventricular arrhythmias (9/13), 3rd degree atrioventricular block (4/13) or sinus rhythm with right axis deviation (1/13). ST segment abnormalities were present in 6 dogs.
Regional wall motion abnormalities affecting various myocardial segments (left ventricular free wall (4/11), apex (4/11) and interventricular septum (3/11)) were observed at the first exam in 8/13 or at follow-up visits in 9/10 dogs. Abnormal segments appeared thin and hyperechoic in 7/10 dogs during follow-up echocardiograms. Systolic

dysfunction was observed in 9/13 dogs (5/13 at presentation and 4/10 at follow-up). Three dogs did not survive to discharge. Postmortem exam was performed in 2 dogs, showing severe extensive myocardial necrosis and hemorrhage and/or replacement of myocardial tissue by fibrous tissue on histopathology.

This is the largest case series of dogs with a presumptive diagnosis of AMI. Although AMI remains a rare condition, it should be considered a differential diagnosis in dogs with a predisposing condition, arrhythmia, elevated cTnI and/or segmental wall motion abnormalities. However, Findings may only become apparent after the acute phase.

Disclosures

No disclosures to report.

ESVCN-P-1

Studies on estimation of ideal body weight by morphometry in dogs

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Accurate clinical nutritional assessment by body condition scoring in dogs is not easy because of subjective method. Therefore, to make an objective nutritional assessment, we examined a morphometric method. As a result, a high correlation coefficient (r = 0.945) occurred between the length from the sternal process to the sciatic process and the ideal body weight (IBW). In this studies, we present a caliper that can estimate the IBW by measuring the length by applying this finding.

A caliper was prepared on which the length and the IBW appeared simultaneously. The ideal body weight on the caliper was the weight corresponding to either a body fat percentage of 20% or 25%. The length between the sternal process and the sciatic process was measured by clinical veterinarians using caliper. Ten dogs that visited the animal hospital were used. The veterinarians were asked to assess how the IBW determined by the callipers differed from that of the IBW based on their experience.

The correlation coefficient between body length and IBW was r = 0.208, when the IBW corresponded to a body fat percentage of 20%. On the other hand, the correlation coefficient between the two was r = 0.333 when the IBW corresponded to a body fat percentage of 25%. Clinical veterinarians also replied in the questionnaire survey that a body weight that corresponded to a body fat percentage of 25% was closer to the IBW assessed by their experience. Body fat percentage at a BCS of 3 on a 5-point scale corresponds to 15 to 25%, with a median of 20%. It is suggested that Japanese clinical veterinarians are evaluating their nutritional assessments by setting their IBW higher. It is found that life span can be extended by dietary restriction in dogs. From this point of view, Veterinarians should set IBM lower than their assessment results. Also, there was a large difference in the correlation coefficient between our measured body length and that measured by the clinical veterinarians. The difference was caused by the large variation in body length measurements by veterinarians. In this regard, it may be necessary to create a manual for body length measurement in dogs to improve measurement accuracy.

Disclosures

No disclosures to report.

ESVE-P-1

Hypothyroidism and its association with extra hepatic biliary diseases in dogs: a retrospective case-control study

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Hypothyroidism may have a major implication in delayed gallbladder emptying. Its role in the pathogenesis of gallbladder mucocele and other extrahepatic biliary diseases merits investigation.

The aim of this study is to evaluate the incidence of extrahepatic biliary diseases in hypothyroid dogs.

Records of 63 dogs diagnosed with hypothyroidism have been examined for the presence or absence of extrahepatic biliary disease (mucocele, colecistitis, colelitiasis and gallblader masses).

Forty nine dogs (77.7%) had serum elevation of gamma glutamyl transferase, alkaline phosphatase, alanine aminotransferase and total bilirubine, enlarged gallbladders, finely striated or immobile stellate bile patterns, thickened gallbladder walls, biliary sludge or hyperechogenic gallbladder masses. Older dogs and small breeds were overrepresented. Most dogs presented non-specific clinical signs such as vomiting, anorexia and lethargy, diarrhea and/or fever. Thirty four dogs (53.9%) were diagnosed with extrahepatic biliary disease: gallbladder mucocele (n = 21 dogs), cholelitiasis (n = 8 dogs) and colecystitis (n = 5 dogs) upon ultrasound examination, histologic/macroscopic evaluation and culture and sensitivity tests. The incidence of gallbladder mucocele was found to be the highest (33,3%) followed by cholelitiasis (12.6%) and colecystitis (7.9%).

This study suggests that hyporthiroid dogs that are presented for acute illness with laboratory evidence of hepatobiliary disease should undergo evaluation for the presence of extrahepatic biliary diseases such as biliary mucocele, cholelitiasis and colecystitis.

Disclosures

No disclosures to report.

ESVE-P-2

Planar and SPECT imaging of canine thyroid tumors: 68 cases

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Thyroid scintigraphy is indispensable for the diagnosis, staging and treatment planning of thyroid carcinoma. However, literature on pertechnetate (^{99m}TcO4) and iodine-123 (^{123}I) scintigram findings in dogs is scarce, and nearly absent on single-photon emission computed tomography (SPECT). Similar to human medicine, the use of SPECT could increase the sensitivity in detection of metastases.

The aim of this retrospective study was to describe planar and SPECT imaging results in canine thyroid tumors, and to compare it with thoracic radiography for detection of thoracic metastases.

Thyroid scintigraphy was available from 68 dogs presented at our clinic between 2008 and 2018, of which 6 presented after surgical resection. All dogs had a confirmed diagnosis of thyroid neoplasia based on histopathology, cytology, and/or abnormal radionuclide accumulation.

Thirty-nine dogs had unilateral tumors, 14 bilateral, and 10 ectopic tumors. One dog had an ectopic and unilateral tumor, and 1 dog had bilateral masses and an ectopic tumor. For 3 dogs, tumor extent hindered accurate localization.

Uptake relative to the parotid salivary glands (thyroid/salivary (T/S) ratio) was increased in 35 of 62 dogs (median T/S ratio 3.2 (1.1-12)), decreased in 15 dogs (median T/S ratio 0.73 (0.36-0.90)), and comparable to that of the salivary glands (median T/S ratio 0.96 (0.85-1.0)) in 8 dogs. In 4 dogs with multiple masses, uptake extent was different at the level of the various masses. A homogeneous, uniform uptake pattern was present in 10 dogs and a heterogeneous uptake pattern in 45 dogs. In 7 dogs (all with multiple masses), various uptake patterns were present.

Thirty-one dogs were euthyroid, 12 were hyperthyroid, and 9 were hypothyroid. The majority of hyperthyroid dogs (10 of 12 dogs) had increased radionuclide uptake.

SPECT imaging was available in 41 dogs. In 16 dogs, SPECT revealed tracer uptake at the level of the thorax, suggestive of distant metastases. In 13 of these dogs, thoracic radiographs were performed, and metastases were detected in only 2 of 13 dogs.

In the majority of dogs, scintigraphy was performed using ^{99m}TcO4. In 5 dogs, both ^{99m}TcO4 and ^{123}I scintigrams were performed. In 1 of these dogs, ^{123}I SPECT imaging revealed a thoracic metastasis that was not clearly identified with ^{99m}TcO4.

This study is the first to describe planar and SPECT imaging in a large number of dogs with thyroid tumors. Our results suggest that SPECT imaging is superior to thoracic radiography for detection of metastases.

Disclosures

No disclosures to report.

ESVE-P-4

Ultrasonographic evaluation of adrenal gland thickness in healthy dogs and in dogs with hyperadrenocorticism

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Ultrasonography is commonly used in the diagnostic workup of dogs with hyperadrenocorticism (HAC) to support its diagnosis, to rule out non-adrenal diseases and to help differentiate the type of HAC. Some studies have established reference ranges for ultrasonographic adrenal gland thickness based on weight categories. However, the use of those ranges in dogs with HAC, have not been evaluated. Therefore, four weight categories were used in order to avoid large variations of weight within each group. The aims of this study were to establish new reference ranges for adrenal gland thickness in healthy dogs, to evaluate the sensitivity of those ranges to detect adrenomegaly in dogs with HAC and to describe ultrasonographic adrenal findings in dogs with HAC.

A total of 86 clinically healthy dogs were prospectively included. Adrenal thickness in a sagittal plane were measured in dogs using the following weight categories: 21 dogs ≥2.5-5 kg, 22 dogs >5-10 kg, 22 dogs >10-20 kg, and 21 dogs >20-40 kg. Reference ranges (5-95th percentile) for left adrenal gland maximum thickness were as follows: 3.4-4.8 mm (dogs ≥2.5-5 kg), 3.4-5.6 mm (dogs >5-10 kg), 3.9-6.2 mm (dogs >10-20 kg), and 5.2-7.4 mm (dogs >20-40 kg); whereas for right adrenal gland maximum thickness were as follows: 3.2-5.5 mm (dogs ≥2.5-5 kg), 3.8-6.0 mm (dogs >5-10 kg), 4.2-7.7 mm (dogs >10-20 kg), and 5.4-9.4 mm (dogs >20-40 kg).

In addition, other 85 dogs with HAC were retrospectively included. These dogs were classified into the same weight categories used for healthy dogs: 23 dogs ≥2.5-5 kg; 37 dogs >5-10 kg; 17 dogs >10-20 kg, and 8 dogs >20-40 kg. The overall sensitivity for detection of unilateral or bilateral adrenomegaly on ultrasound examination was 97% (82/85 dogs). Dogs with HAC were ultrasonographically classified as: 41 (48%) dogs with symmetrical adrenomegaly (consistent with pituitary-dependent HAC), 11 (13%) dogs with unilateral adrenomegaly and atrophy of the contralateral adrenal gland or unilateral or bilateral adrenomegaly with malignancy features on the ultrasound examination (consistent with adrenal-dependent HAC), 30 (35%) dogs with equivocal adrenal asymmetry, and 3 (4%) dogs with normal adrenal gland thickness.

The overall sensitivity of ultrasonography to detect adrenomegaly using four weight categories for left and right adrenal gland thickness was 97%. Although, most dogs with HAC (61%) had ultrasonographic findings consistent with either pituitary- or adrenal-dependent HAC, equivocal adrenal asymmetry was a common finding in dogs with HAC, occurring in 30 of 85 dogs (35%).

Disclosures

No disclosures to report.

ESVE-P-5

Ultrasonographic accuracy in primary adrenal insufficiency: a retrospective cohort study of 182 dogs

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The clinical relevance of ultrasonography in canine primary adrenal insufficiency has been evaluated in very few studies. This retrospective cohort-study evaluated in a large canine population referred to our facility, the diagnostic performance of ultrasonographically assessed adrenal gland dimensions in distinguishing dogs with primary adrenal insufficiency (Exposed or Group 1) from dogs without primary adrenal insufficiency (Non -exposed or Group 2).

Group 1 included 91 consecutive client-owned dogs diagnosed with primary adrenal insufficiency on the base of signalment, history, clinicopathologic tests (CBCs, serum biochemistry, urinalyses) and ACTH stimulation test results (including basal plasma ACTH and serum aldosterone pre- and post-ACTH determination). Group 2 included 91 dogs without primary adrenal insufficiency, individually matched with Group 1 dogs for breed, sex, sexual status and age (± 6 months). To reduce technological bias, Group 2 dogs selection took place in contiguous time periods with each Group 1 dog recruitment. Dogs that had received mitotane, trilostane or steroid medications and ace-inhibitors before our clinical evaluation, were excluded from both groups.

Ultrasonographic adrenal lengths (AL), caudal and cranial pole thicknesses (CdT and CrT, respectively) were determined on longitudinal images in both Groups and compared.

Median (IQR) right and left CdTs in Group 1 were 0.30 (0.16) cm and 0.29 (0.10) cm, respectively; in Group 2 were 0.50 (0.17) cm and 0.51 (0.16) cm, respectively (p value <0.0001 for both comparisons). Median (IQR) right and left CrTs in Group 1 were 0.32 (0.18) cm and 0.28 (0.10) cm, respectively; in Group 2 were 0.53 (0.12) cm and 0.49 (0.16) cm, respectively (p value <0.0001 for both comparisons). Median right and left ALs in Group 1 were 1.40 (0.55) cm and 1.42 (0.76) cm, respectively; in Group 2 were 1.92 (0.75) cm and 2.08 (0.66) cm, respectively (p value <0.0001 for both comparisons). From the analysis of ROC curves, global diagnostic accuracy separating Group 1 from Group 2 resulted 0.899 and 0.900 for the right and left CdT, respectively; 0.861 and 0.911 for the right and left CrT, respectively; 0.762 and 0.760 for the right and left AL, respectively.

This study shows that Group 1 sonographic adrenal gland measures were significantly lower compared to Group 2 and strengthens the role of adrenal ultrasonography as a reliable screening test for dogs suspected of primary adrenal insufficiency.

Disclosures

No disclosures to report.

ESVE-P-6

Effect of sample dilution on free T4 depends on physiological state and analytical method

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The binding capacity of human sera for thyroxine is such that, in the absence of conditions that would reduce thyroid binding capacity (TBC), e.g., non-thyroidal illness (NTI), pregnancy or free fatty acids, sera may be diluted more than 100-fold without affecting the measured result for Free T4 using an appropriate analytical technique such as equilibrium dialysis. However, in conditions associated with reduced TBC, results may decrease at dilutions as low as 1:10. Free T4 dilution profiles have been used to determine the validity of free T4 methods for humans.

In dogs and cats, TBC is much lower and it is not clear what the equivalent dilution would be that would cause a decrease in measured Free T4. The analytical impact of physiological states including NTI have not been investigated. Analytical methods which are less resistant to the effects of sample dilution may be more susceptible to interference from lowered TBC (eg., NTI) and consequently may generate diagnostically misleading results.

Surplus serum samples from dogs and cats in a selection of physiological states were analysed by an equilibrium dialysis method (FT4 by equilibrium dialysis, Antech Laboratories (FT4d)) and an analogue chemiluminescent method (Immulite 2000 Veterinary Free T4, Siemens (VF4)) with and without dilution in HEPES buffer at 1:2, 1:10, 1:20, 1:40, 1:80. Samples included: normal cat (TT4 = 11.8 nmoL/L), hyperthyroid cat (TT4 = 104), suspected canine NTI (TT4 = 12.8, normal TSH), suspected feline NTI (TT4 = 40, FT4d = 112), feline lipaemia (TT4 = 52).

Percentage decreases could not be calculated for all samples because of the reporting limits of the assays. However, in all samples, there was a decrease in FT4d between 20 and 32% at the 1:10 dilution which compared to decrease of 59 to 79% by VF4 at the same dilution for samples in which percentage decrease could be calculated. The steepest decline in concentration beyond 1:10 was in the suspected feline NTI sample and the least decline in the normal cat.

Veterinary samples were more susceptible to the effects of sample dilution (and therefore conditions of reduced TBC) than reported for human sera. The VF4 method was more susceptible to dilution than the FT4d method. As in humans, some physiological states demonstrate a greater decline in FT4 measurements in serial dilution. Further studies could determine whether comparisons between undiluted and diluted FT4d results within an individual sample, could differentiate NTI from thyroid dysfunction in diagnostically challenging cases.

Disclosures

Disclosures to report.

NationWIde Laboratories (Consultancy) Dechra Veterinary Products (Consultancy).

ESVE-P-7

Survival in cats with diabetes mellitus and chronic pancreatitis: a preliminary study of 36 cases

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Previous studies suggest that pancreatitis could be a significant comorbidity in diabetic cats, even in absence of digestive signs. Long-term studies of diabetic cats with pancreatitis are lacking and potential survival factors in such population are unknown.

The main objectives were 1/to review epidemiological, biological and ultrasonographic findings in cats with concurrent chronic pancreatitis (CP) and diabetes mellitus (DM) and 2/to document chronology of both diseases, survival time (ST) and potential prognostic factors.

Thirty-six cats diagnosed with CP and DM were retrospectively recruited at Veterinary Teaching Hospital of National Veterinary School of Alfort between January 2008 and June 2017. For all cats, signalment, medical history, physical findings, biochemistry panel, complete blood count, standard urine analysis, and abdominal ultrasonography (US) were available. ST was known for all cases. Potential

prognostic factors including elevation of hepatic enzymes, hyperbilirubinemia, hypokalemia, episode of ketoacidosis, history of hospitalization and US features suggesting biliary tract disease were evaluated. Association of each criteria and ST was tested with univariate analysis (Log rank test); significance was set at $P < 0.05$.

Median age of study population was 12 years. Recruited cats were previously overweight (71%) with weight loss (80%). The most frequent biochemical abnormalities included elevation of liver enzymes (63%), hyperproteinemia (61%), hyperbilirubinemia (47%) and hypokalemia (54%). Median survival time (MST) was 955 days. Mortality rate of cats having at least one US finding compatible with biliary tract disease was significantly higher and MST was also 2,57 times shorter ($P = 0.05$). The other studied criteria were not associated with ST.

Long-term outcome of cats suffering from CP and DM may be favorable. However, MST of cats diagnosed with those concurrent diseases and having at least one ultrasonographic finding compatible with a biliary tract disease is shorter than cats without any biliary lesion on US.

Disclosures

No disclosures to report.

ESVE-P-8

Efficacy of once daily Protamine Zinc Recombinant Human Insulin (ProZinc®) in canine diabetes mellitus

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Quality of life research among diabetic dog owners emphasizes the negative impact of diabetes mellitus (DM) treatment on owner lifestyle, even leading to euthanasia. Once-daily (SID) insulin injection regimens reduce such impact.

This prospective, baseline-controlled, multi-centre clinical field study evaluated the efficacy of SID administered ProZinc insulin in dogs diagnosed with DM in line with the ALIVE-criteria. Seven follow-up visits occurred over an 84-day period. Between days 28-42, the veterinarian was permitted to switch to twice-daily (BID) treatment on the basis of clinical signs, maximum blood glucose (BG) or minimum BG obtained from a 9-hour BG-curve. Satisfactory diabetic control was defined as an improvement in ≥1 glycemic laboratory parameter and in ≥1 clinical sign.

Thirty dogs were enrolled; 5 withdrew early (n = 2 consent withdrawn, n = 2 compliance, n = 1 lack of improvement). Overall, 76% (19/25) of dogs showed satisfactory diabetic control at day 84; 77% (10/13) of SID treated dogs and 75% (9/12) of BID treated dogs. In treatment-naïve dogs, satisfactory control was achieved in 91% (10/11; 7 SID, 3 BID) and in insulin pre-treated dogs in 64% (9/14; 3 SID, 6 BID). At study conclusion, 80% (20/25) showed improvement in ≥1 one clinical sign (PU/PD, body weight). Mean BG decreased from 482 ± 125 to 295 ± 65 mg/dL, minimum BG from 437 ± 140 to 218 ± 84 mg/dL, and fructosamine from 566 ± 119 to 394 ± 83 µmol/L. Clinical hypoglycemia was observed once in two SID treated dogs, which recovered after feeding.

SID ProZinc was effective and safe in controlling DM in the majority of dogs, particularly in naïve dogs.

Disclosures

Disclosures to report.

S.J.M. Niessen - consultancy work for Dechra, Purina, Boehringer Ingelheim C. Kroh - employee Boehringer Ingelheim S. Maruyama - employee Boehringer Ingelheim K. Jerrentrup - employee Boehringer Ingelheim A. Keller - employee Boehringer Ingelheim R. Klee - employee Boehringer Ingelheim A. Mori - consultancy work Boehringer Ingelheim T. Sako - consultancy Boehringer Ingelheim.

ESVE-P-9

Brachycephalic morphotype and pituitary tumor size in dogs with Cushing's disease

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Cushing's disease (CD) is a common canine endocrinopathy, due to a deregulated secretion of ACTH by a pituitary microadenoma or macroadenoma. A recent study focusing on histopathologic findings in canine pituitary gland identified an overrepresentation of brachycephalic dogs (BD) among macroadenomas, disregarding their secretory status.

We therefore hypothesised that pituitary tumor size and brachycephalic morphotype were associated in dogs with CD.

Medical records of dogs with CD presented at the Internal Medicine Units of 2 referrals hospitals were retrospectively evaluated. Inclusion criteria were: 1/ clinical signs suggestive of hyperadrenocorticism; 2/ hyperadrenocorticism confirmation by at least 1 endocrine test; 3/ a brain and abdominal CT scan indicative of a pituitary origin.

Macroadenomas were diagnosed when the pituitary height/brain area ratio (P/B) was $\geq 0.40 \times 10^{-2}$ mm^{-1}. A skull index (SI: skull width/lengthx100) was calculated to differentiate BD from non-brachycephalic dogs (NBD).

The SI accuracy to distinguish between BD and NBD breeds was tested after calculation of the area under the curve (AUC) of the Receiver Operator Characteristic (ROC) curve. The optimal cut-off was selected based on the value of the Youden's index. The correlation between SI and P/B was evaluated using the Spearman's test.

Hundred and twenty dogs were included. The median [range] age at first clinical signs was of 9 [5-15] years. According to their breeds, 52 were BD, 61 were NBD, and 5 were unclassified.

The AUC (95% confidence interval) of the ROC curve of SI for distinguishing BD from NBD was 0.92 (0.85-0.98). Using a cut-off at 71.03, the sensibility and the specificity of SI for morphotype distinction were 0.90 (0.77-0.96) and 0.87 (0.70-0.95) respectively.

The median [range] P/B were 0.3×10^{-2} mm^{-1} [$0.2\text{-}1.4 \times 10^{-2}$ mm^{-1}] and 0.4×10^{-2} mm^{-1}[$0.1\text{-}1.7 \times 10^{-2}$ mm^{-1}] in BD and NBD respectively. No correlation could be established between SI and P/B ($\rho = 0.02$, $P = 0.80$).

Brachycephalic morphotype quantitatively evaluated was not associated with pituitary tumor size in dogs with CD. Our results are in contrast with a previous study focusing on pituitary samples collected mainly by necropsy and embracing dog with and without CD. The sole inclusion of dogs with CD in our study may explain this discrepancy. A possible predisposition of BD to undifferentiated, rapidly growing non-functioning pituitary tumors remains to be assessed.

Disclosures

No disclosures to report.

ESVE-P-10

Critical illness-related corticosteroid insufficiency (CIRCI) in dogs with systemic inflammatory response syndrome (SIRS)

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Critical illness-related corticosteroid insufficiency (CIRCI) is an inadequate corticosteroid activity in relation to the patient's current degree of stress or illness. CIRCI occurs in 30-60% of critically-ill human patients and up to 48% of dogs with sepsis.

This study investigated the frequency of CIRCI in systemic inflammatory response syndrome (SIRS) dogs and associations between CIRCI and hypotension and mortality.

A single-center prospective study was performed between December 2016 and May 2017 (ethical approval n°63 711/2016). SIRS was diagnosed if dogs presented at least two of the following criteria at the admission in ICU: 1) rectal temperature > 39.0°C or < 38.0°C; 2) heart rate > 120 bpm; 3) respiratory rate > 20 bpm; 4) white blood cells <6 x 10^3/µL or > 16 x 10^3/µL or > 3% of band neutrophils. Dogs were excluded if they have a history of or suspected adrenal illness or if they received glucocorticoids within the previous 72 hours or long acting formulations within the previous month or other drugs known to affect the hypothalamic-pituitary-adrenal axis.

ACTH stimulation test was performed in all dogs immediately after inclusion in the study and dogs with a Δ cortisol (difference between post-ACTH stimulation and basal cortisol) ≤3 µg/dL supported diagnosis of CIRCI. Non-invasive blood pressure (petMAP™graphic II, Ramsey Medical) was measured in all dogs and hypotension was defined as a mean arterial pressure (MAP) <60 mmHg. Information about survival at 28 days after admission were collected. Dogs were divided into survivors and non-survivors. Dogs that were euthanized for financial reasons were excluded. D'Agostino-Pearson's test tested data for normality. Age, basal cortisol and MAP were compared between dogs with or without CIRCI. Δ cortisol, basal cortisol and MAP were compared between survivors and non-survivors using *t*-test. Association between CIRCI, hypotension and mortality were evaluated with Fisher's exact test. For all analyses, a P-value ≤0.05 was considered significant.

Twenty-one dogs met the inclusion criteria and were enrolled in the study. CIRCI and hypotension was detected in 10/21 (48%) and 7/21 (33%) dogs, respectively. Age, MAP, basal cortisol and hypotension were similar between dogs with or without CIRCI. 14/21 dogs (67%) died within 28 days from admission. As independent factor, hypotension and basal hypercortisolemia were associated with higher risk of death (P = 0.04 and P = 0.0251, respectively).

CIRCI seems to occur frequently in SIRS dogs. However, only presence of hypotension or basal hypercortisolemia was associated with increased mortality risk.

Disclosures

No disclosures to report.

ESVE-P-11

Prednisolone induced hyperglycaemia and diabetes mellitus in cats

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Prednisolone is a commonly used drug in cats. Potential adverse effects include glucocorticoid-induced hyperglycaemia (GIH) and diabetes mellitus (GIDM) but predisposing factors for the development and the overall incidence rate of GIH and GIDM are currently unknown. The aims of this study were: (1) to evaluate the incidence rate of GIH and GIDM, and (2) evaluate for predisposing risk factors of GIH and GIDM in cats receiving prednisolone.

The electronic records of a tertiary referral centre were searched for cats receiving prednisolone at a dose of >1.9 mg/kg/day of >3 weeks duration, and that had follow-up data available of >6 weeks during a study period between January 2010 and June 2017. In total 143 cats were included in the study. Of these cats, 14 cats (9.8%) were diagnosed with GIH or GIDM. Nine cats (6.3%) developed GIDM, with 8 requiring insulin therapy and 1 was euthanized due to diabetic ketoacidosis. The remaining 5 cats (3.4%) were identified as GIH with none requiring insulin treatment. Twelve out of 14 cats (85.7%) developed GIH or GIDM within 3 months after the initiation of prednisolone therapy, the remaining two cats developed GIDM after 27 and 32 months. Four of the cats developing GIDM/GIH were < 2 years of age. Comparison between cats developing GIH or GIDM to those that did not, showed no statistical difference in the baseline (pre-prednisolone) blood glucose, presence of glucosuria, body weight and body condition score. A trend towards cats developing GIDM/GIH if receiving >3.0 mg/kg starting dose (OR 2.7) was seen, however this was not statistically significant (P 0.097).

We conclude that in a tertiary referral population, approximately 10% of cats receiving prednisolone develop GIH or GIDM. There was atrend towards cats receiving higher doses of prednisolone being more likely to develop GIH/GIDM.

Disclosures

No disclosures to report.

ESVE-P-12

Accuracy and precision of insulin administration using human and veterinary pen-injectors and syringes

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Many diabetic dogs and cats require small doses of insulin, which may be administered with syringes or pen-injector devices. It is important that these small doses are administered accurately and that the magnitude of potential dosage error is appreciated. The aim of this study was to compare the accuracy and precision of insulin syringes and pen devices.

To determine how accurately and precisely insulin doses are delivered, 0.5, 1, 2, 4, 8 and 16 U doses were dispensed 25 times from five SoloSTAR® containing insulin glargine, five FlexPen® containing insulin detemir, five KwikPen® containing insulin lispro, five JuniorSTAR® containing insulin glargine, five VetPen® 0.5-8 U and five VetPen® 1-16 U containing insulin Caninsulin®, and by five veterinarians using 30 U/0.3 mL and 40 U/mL insulin syringes. Each dose was weighed immediately using a precision balance (resolution of 0.00001 g), and the intended and delivered doses were compared.

All pen-injectors tended to deliver less insulin than the intended dose, underdosage being inversely proportional to dose (accuracy from −6.86% to −0.84%). The differences between intended and delivered dose were not significant only using JuniorSTAR and VetPen 0.5-8 U at insulin dosage of 0.5, 1, 2 and 4 U. Using 30 U/0.3 mL insulin syringes the intended dose was significantly overdosed when attempting to deliver 0.5, 1 and 2 U (+26.51%, +10.32% and + 3.26%, respectively), and significantly underdosed at 8 and 16 U (−3.44% and −4.46%, respectively). Using 40 U/mL syringes the intended dose was significantly overdosed when attempting to deliver 0.5, 1 and 2 U (+30.77%, +5.63% and + 2.84%, respectively). With all six pen-injectors and with both 30 U/0.3 mL and 40 U/mL insulin syringes, the coefficient of variation (precision) diminished with increasing doses of insulin. Precision was <8% for all six pen-injectors (from 7.67% to 0.69%). Conversely, precision using 30 U/0.3 mL and 40 U/mL syringes at insulin dosage of 0.5 U was 12.08% and 9.39%, respectively; precision improves at insulin dosages ≥1 U (from 5.80% to 0.46%).

All devices, with the exception of JuniorSTAR and VetPen 0.5-8 U, are unacceptably inaccurate when delivering 0.5, 1, and 2 U doses of insulin. The accuracy improves when higher doses are dispensed, but the delivery of 8 and 16 U doses resulted sufficiently accurate compared to intended doses only using 40 U/mL syringes. In conclusion, if pen-injectors tend to be more accurate at lower dosages, syringes tend to be more accurate at higher dosages.

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ESVE-P-13

Fractional excretion of electrolytes in dogs with primary hypoadrenocorticism before and after treatment

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Electrolytic abnormalities in dogs with primary hypoadrenocorticism (PH) have been widely described, while fractional excretion (FE) of urinary electrolytes (FEe) has not yet been evaluated. Furthermore, mineralocorticoid supplementation's monitoring is based on blood sodium ([Na]) and potassium ([K]) concentrations, and it is possible that FEe could add useful information regarding the monitoring of the treatment.

The aim of this study were to evaluate FEe in dogs with PH. Study-population was grouped as follow: 1) dogs with acute adrenal insufficiency (AAI), dogs treated for PH (TD) [both with fludrocortisone and desoxycorticosterone pivalate (DOCP)] 2) dogs with PH treated with DOCP classified as well controlled ([Na] and [K] in the RI), under-controlled (hyponatremia and/or hyperkalemia) and over-controlled (hypernatremia and/or hypokalemia), respectively 3) dogs well controlled classified based on drug administration's timing: 9-15, 23-27 and 28-33 days after DOCP injection, respectively.

Only dogs with "typical" PH (hyponatremia and/or hyperkalemia) at the time of diagnosis were included. Healthy dogs (HD) were used as controls. Serum and urine chemistry were performed on combined samples using an automated analyser, and FEe was calculated. Non-parametric tests were used to compare FEe among groups. Data are expressed as median and (range). $P < 0.05$ was considered significant.

Seven dogs with AAI, 18 TD and 115 HD were enrolled; 76 follow-up from TD (13 DOCP, 5 fludrocortisone) dogs were evaluated.

1) FE of sodium (FENa), cloride (FECl) and calcium (FECa) were elevated in AAI [FENa% 2.64(1.48-7.77); FECl% 3.76(1.87-8.15); FECa% 1.68(0.68-8.11)]; they were significantly lower in TD [FENa% 0.41 (0.04-1.91); FECl% 0.73(0.07-21.85); FECa% 0.35(0.07-1.82)], but still significantly higher compared to HD [FENa% 0.25(0.01-1.55); FECl% 0.54(0.05-2.28); FECa% 0.14(0.03-0.66)]. AAI and TD had a significantly higher FE of K (FEK%) [18.53(8.34-62.74); 16.01(4.33-44.19)] compared to HD [10.54(2.23-45.20)].

2) FENa and FEK were not significantly different among the 3 groups; nevertheless, despite not significant, over-controlled dogs had lower FENa and higher FEK if compared to controlled dogs [FENa% 0.43 (0.04-0.95) vs 0.40(0.04-1.91); FEK% 16.14(4.33-42.95) vs 17.10 (5.76-44.19)]. FECa was significantly lower in under-controlled compared to well controlled and over-controlled dogs [FECa% 0.11 (0.07-0.14); 0.33(0.07-1.60); 0.55(0.17-1.82)].

3) FENa, FECl and FEK did not differ significantly among 3 groups; nonetheless, despite not significant, all showed an increasing trend over time. Although not significant, FECa was lower at 23-27 days after DOCP.

Dogs with AAI have high FENa, FECl, FECa and they decrease after treatment. Further studies are necessary to clarify the clinical utility of the FEe in dogs treated for PH.

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ESVE-P-14

Effects of bodyweight, age and pituitary hyperadrenocorticism on the adrenal gland size of dogs, measured by ultrasonography

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Ultrasonography is a sensitive method to measure adrenal gland size. The most reliable indicator of adrenal gland size is the maximum diameter of the caudal pole (thickness). However pituitary hyperadrenocorticism has an increased prevalence in small-breed dogs, there are only few informations in the scientific literature about the correlation between the bodyweight, age and size of adrenal glands in dogs (general upper limit is 7,4 mm). Recently one retrospective study suggested 6 mm cut-off value for dogs below 10 kg and another found an age dependency of the size.

We aimed to examine adrenal gland's thickness of dogs in a retrospective study. Results of 67 dogs (healthy and diagnosed with pituitary hyperadrenocorticism) were used. Diagnosis of pituitary hyperadrenocorticism was based on clinical symptoms and dexamethason supression test. Bodyweight ranged from 3,0 to 42,0 kg (12,7+/−9,6), while age ranged from 1,2 to 14,4 years (7,9+/−3,6). The correlation of bodyweight, age and adrenal gland thickness were analysed in healthy dogs (n = 31). The subgroup below 10 kg (n = 17) was compared to dogs diagnosed with pituitary hyperadrenocorticism below 10 kg (n = 17). To data analysis descriptive statistics, correlation- and regression-analysis, hypothesis-tests and ROC-analysis were used ($P < 0,05$).

There was a significant correlation between adrenal gland size and bodyweight in the entire range of 3-42 kg and no significant correlation in the subgroup of dogs below 10 kg. There was no age dependency. We found significant difference between adrenal gland sizes of dogs below 10 kg and those of weighting more. Adrenal gland sizes were normally distributed. In the group of dogs weighting less than 10 kg the thickness of the left adrenal gland's caudal pole was 4,5 +/−0,7 mm, while the right adrenal gland's size was 4,7+/−0,8 mm. Normal range for the left adrenal gland's size was 3,0-6,0 mm and 3,1-6,1 mm for the right one (95% CI). Comparing data of healthy dogs and dogs with pituitary hyperadrenocorticism resulted 5,5 mm to be the optimal upper cut-off value for dogs below 10 kg. This value provided 82% sensitivity and 94% specificity for the left adrenal gland and 76% sensitivity and 88% specificity for the right one to diagnose pituitary hyperadrenocorticism using ultrasonography.

Different results between the left and right sides are caused by the more difficult imaging of the right sided organ. Our results strenghten the findings of the available few literature regarding the size-range and bodyweight correlation, but we could not find any age dependency.

Disclosures

No disclosures to report.

ESVIM-P-2

Retrospective study of 23 cases of canine nasal polyposis, of which 10 were treated endoscopically

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Nasal polyposis is a pathology of unknown etiology, rarely described in canine species, that can provoke chronic clinical symptoms including discharge, sneezing and stertor.

The diagnosis is based on diagnostic imaging, endoscopic aspect and histologic exam of the nasal biopsy. Rhinotomy, despite possible surgical complications and the possibility of relapse, is to date the first choice in treatment[1,2].

In our retrospective study we looked at 23 cases of nasal polyps over 8 years. The dogs were principally mixed breeds; males (60%) and females (40%); and adult or elderly, but 39% of the patients were less than 8 years old. They presented with sneezing, stertor and nasal discharge. The clinical exam showed 6 cases of frontonasal deformation with a clear presence of newly formed polypoid tissue in the nostrils and in the opening of the nasolacrimal ducts. X-rays and computerized tomography reveal abnormalities compatible with newly formed endonasal tissue in 84.6% and 100% of the cases, respectively. Anterograde rhinoscopy reveals newly formed tissue with a smooth translucent surface, pink in color, and with an elastic consistency, completely occupying the nasal meatus and in 12 dogs the nasopharynx as well. The histological exam shows an exophytic structure bounded by the respiratory epithelium that are often hyperplastic or with squamous metaplasia. The stroma is, as a rule, myxoid, fibrillary and surrounds serous or mucus glands, which are present in highly variable quantities. In the corium the inflammatory infiltrate is polymorphous. The stroma can show areas of hyperplasia of the mesenchyme and/or hyperplasia and dilation of the vascular system. Serious dysplasia was not observed in any of the cases.

Twelve of the subjects underwent only medical treatment, 10 underwent medical treatment and endoscopic debulking with mixed techniques (external grasping forceps plus diode laser 5 W, continuous rhythm, 600 μm fiber), and 1 patient underwent surgical rhinotomy. In all the dogs treated only with Prednisolone, despite the clinical improvement, there were constant relapses. All of the subjects that underwent endoscopic debulking showed clinical improvement of the symptoms and absence of relapse, for 6 months in 77.8% of the cases and for 24 months in 44.5% of the cases. In conclusion, nasal polyposis is an infrequent pathology that can present serious clinical symptoms, even in young subjects, and that tends to relapse. Therapy with endoscopic debulking is a less invasive alternative than traditional surgery, even though it does not prevent relapse in the medium and long term.

Disclosures

No disclosures to report.

ESVIM-P-3

Diagnostic utility of reticulocyte haemoglobin content (RETIC-HGB) to detect iron-limited erythropoiesis in cats

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Reticulocyte haemoglobin content (CHr) (Siemens ADVIA 2120) is a diagnostic marker of iron deficiency in humans and dogs. RETIC-HGB (IDEXX ProCyte Dx) is a new parameter for analysis of iron deficiency

for veterinary use. Aim of this prospective study was to evaluate the clinical and diagnostic utility of RETIC-HGB compared to CHr in the diagnosis of feline iron-limited erythropoiesis (ILE).

First, reference intervals (RIs) for RETIC-HGB and CHr were established analysing 59 healthy non-anaemic cats. Second, 275 cats were classified as having ILE or not. Low plasma iron or low transferrin saturation (%TfS) in combination with either anaemia and/or altered red blood cell (RBC) indices was required for diagnosis of ILE. Haematologic variables, parameters of iron metabolism as well as serum amyloid A were compared between both groups and correlation between RETIC-HGB and CHr was assessed.

RIs for RETIC-HGB and CHr were 12.5-18.0 pg and 14.0-19.9 pg, respectively. In respect of iron and haematologic variables, 20/275 cats (7.3%) were classified as ILE cats. Compared to non-ILE cats, ILE cats had significantly lower median values of RETIC-HGB and CHr. Not unexpectedly, HCT, HGB, MCH, and RBC were significantly decreased in ILE cats. Additionally, ILE cats had significantly increased median SAA values. Correlation between RETIC-HGB and CHr was moderate (r_s = 0.59) with a small bias of −1.2 pg.

CHr and RETIC-HGB may be suitable early indicators of ILE, especially when used in addition to parameters of iron metabolism. The moderate correlation between RETIC-HGB and CHr is likely due to species and different methodology.

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ESVIM-P-4

A comparison of the diagnostic utility of the classic model, the value of the Anion Gap (AG), corrected Anion Gap (AGcorr) and the chloride/sodium ratio in the diagnosis of acid-base basalnce disturbances in cats with chronic kidney disease (CKD)

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In addition to hypophosphatemia, metabolic acidosis is the most common complication of chronic kidney disease in cats. Using the standard approach, metabolic acidosis is diagnosed based on the concentration of HCO_3^- and pCO_2 in arterial blood.

The aim of the study was to assess the possibility of using the value of the anion gap (AG), corrected anion gap (AG_{corr}) and the chloride/sodium ratio (Cl^-/Na^+) in the diagnosis of metabolic acidosis. The study was carried out on 100 cats (both sexes, 7-9 years old). The control (C) group consisted of 20 healthy cats, while 80 cats that were diagnosed with CKD based on their blood creatinin, SDMA and urea levels were included in the study group. The cats with CKD were divided into four groups: I, II, III and IV, depending on the IRIS stage. Arterial and venous blood was collected from all the animals. Parameters of the acid-base balance (ABB): pH, pCO_2 and HCO_3^- were measured in arterial blood. The concentration of Na^+, K^+, Cl^-, HCO_3^-, albumin were measured in venous blood. Based on the obtained results, the values of the AG, AG_{corr} and Cl^-/Na^+ were calculated using the following formulae: $AG = (Na^+ + K^+) - (Cl^- - HCO_3^-)$, $AG_{corr} = AG + (39\ g/L - alb_{akt}\ g/l)/4$, where 39 g/L was the upper reference limit of serum albumin in cats, alb_{akt} was the measured albumin concentration in the studied cats and $Cl^-/Na^+ = (Cl^-)$: (Na^+).

The analysis of the arterial blood revealed the presence of metabolic acidosis in cats from group IV. In the remaining groups, the ABB parameters were within the reference range. The values of AG and AG_{corr}, in cats from group II, III and IV were significantly lower than those in group C, while the Cl^-/Na^+ value in cats from group II, III and IV was significantly higher than in cats from group C.

The following conclusions were drawn based on the obtained results: 1) the ABB analysis based on the classic model enables detection of ABB disturbances in cats with stage IV kidney disease, 2) the analysis of the AG, AG_{corr},and Cl/Na^+ values enable the diagnosis of ABB disturbances in cats with stage II, III and IV kidney disease, 3) The analysis of the changes in the ion concentrations in CKD are a more accurate tool to diagnose ABB disturbances than the classic model.

Disclosures

No disclosures to report.

ESVIM-P-5

Evaluation of different cleaning methods for bacterial decontamination of feline aerosol chambers

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For aerosol therapy of cats with chronic respiratory conditions commonly aerosol chambers specifically designed for use in cats are utilized. Depending on the type of chamber, certain cleaning procedures are recommended by the different manufacturers to prevent bacterial contamination. Aim of the study was to investigate, if chambers can be adequately decontaminated using different recommended cleaning or sterilizing procedures after standardized bacterial contamination.

For that purpose the chambers “RC Chamber” (RC) (Cegla Medizintechnik) (n = 10) and “AeroKat” (AK) (Trudell Medical International) (n = 5) were evaluated. Standardized bacterial contamination was performed using a *Pseudomonas-aeruginosa*-suspension that was previously established by serial dilution. Aliquots of 50 mL were applied with a pipette on three pre-defined locations of the chamber. After 24 hours chambers were cleaned/sterilized according to manufacturers` instructions. For RC this included a cleaning procedure using a special bag in the microwave for 3 minutes at 800 watt (n = 5) or placement in boiling water for 5 minutes (n = 5). AK was placed in lukewarm water with cleaning detergent for 15 minutes (n = 5). After air drying of all chambers swabs were taken from three defined areas of each chamber (mask, valve, chamber), applied on Mueller-Hinton-agar, and incubated for 24 hours.

With all three cleaning protocols no bacterial growth could be detected in any of the 15 chambers tested. The study shows that with manufacturer recommended cleaning procedures adequate bacterial decontamination of feline inhalation chambers can be achieved.

Disclosures

No disclosures to report.

ESVIM-P-6

Evaluation of clinico-pathological alterations including some leukocyte ratios and survival rate in dogs with IMHA transfused and not transfused: a retrospective study

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Immune-Mediated Hemolytic Anaemia (IMHA) is a common hematological disorder in dogs. It can be primary or secondary and it is characterized by anti-RBC antibodies production. IMHA requires a detailed diagnostic pathway as well as a complex therapeutic approach that can include blood transfusion. Unfortunately, IMHA presents a high mortality rate, especially within 15 days after onset.

This retrospective study evaluated: a) the clinical and clinico-pathological alterations that influenced the choice to perform a blood transfusion in an IMHA patient; b) if blood transfusion could be an additional therapeutic approach; c) application of leukocyte ratios in the prognosis.

Sixty-seven cases of IMHA, both primary and secondary, admitted to the Veterinary Teaching Hospital between May 2010 and July 2018, were included. Signalment, history, clinical signs, clinico-pathological parameters and survival rate were collected. Patients were divided in two groups: 44 patients (IMHAnt) treated with immunosuppressive therapy alone (primary n = 36, secondary n = 8) and 23 patients (IMHAt), which received also a blood transfusion (primarily packed RBC) (primary n = 16, secondary n = 6). For all collected parameters, both groups were statistically compared.

The IMHAt patients compared to IMHAnt patients (un-regarding to primary or secondary cause) presented: worse marks according to Tokyo Score System (TSS) (Chi Squared, $P = 0.003$); a lower erythrocyte count (T-test, $P = 0.039$), hemoglobin concentration (T-test, $P = 0.029$) and platelet count (Mann-Withney, M-W, $P = 0.008$); a higher value of band neutrophils (M-W, $P = 0.022$), band neutrophil to lymphocyte ratio (M-W, $P = 0.005$), (band neutrophil/neutrophil) to lymphocyte ratio (M-W, $P = 0.006$) and a lower value of lymphocyte to monocyte ratio (M-W, $P = 0.013$); a higher value of C-reactive protein (M-W, $P = 0.011$) and activated partial thromboplastin time (M-W, $P = 0.014$); and a lower survival rate at day 120 (Kaplan-Meyer, logrank, $P = 0.004$) and not at 7, 15 and 30 days. Blood transfusions were performed based on the severity of clinical and clinico-pathological signs.

IMHAt patients showed a more severe disease (according to TSS), a greater acute inflammatory condition and more coagulative defects. The high death rate among IMHAt patients at 120 days was related to their critical condition, which is probably why the desired benefit of blood transfusion wasn't reached. However, a link between blood transfusions and the related worse clinical signs in IMHAt patients could not be ruled-out. Finally, the leukocyte ratios in dogs affected by IMHA were assessed for the first time so far and they were proven to be useful markers of acute inflammation and could have a prognostic value.

Disclosures

No disclosures to report.

ESVIM-P-7

Expression of serum exosomal miRNA 122 in dogs naturally infected by Leishmania infantum

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Leishmaniasis a zoonosis caused by *Leishmania* spp., is a chronic and often fatal disease for humans and dogs if left untreated. In recent years, microRNAs (miRNAs), a group of small, single-stranded non-coding RNAs able to regulate gene expression have been shown to play a critical role in the development and function of immune responses. While in circulation, free-serum miRNAs are highly degradable, when transported in mycelial vesicles (exosomes) they become stable (protected from RNAse degradation) and reliable diagnostic biomarker in diseased patients. In 2013, using murine animal the role played by exosomes and miRNAs was explored during Leishmania infection; a reduction in the activity of miR-122, the most abundant miRNA present in the liver tissue, was obtained. Very little is known about the role of exosomal miRNA in canine leishmaniasis (CL); in particular, the interaction between exosomal miR-122 and lipid alterations. The aim of this study was 3fold: 1) isolate/characterize exosomes in canine serum obtained from 6 healthy dogs; 2) evaluate their quality/quantity of exosomal miRNAs and proteins; 3) evaluate the expression of serum exosomal miR-122 in 10 healthy dogs and 10 leishmaniotic dogs.

Blood samples were collected for routine hematological/biochemical analyses on healthy dogs or before anti-*Leishmania* therapy. Biochemical panel was completed with a serum cholesterol profile (HDL, LDL). Serum exosomes were isolated using a polymer-based kit and characterized by flow cytometry and electron microscopy. miR-122-5p expression was analyzed via quantitative RT-PCR. Differences between the two groups were statistical analyzed. A P value of <0.05 was considered significant.

This is the first study showing the detection of circulating serum exosomes content of miR122 in in dogs affected by CL. Serum exosomes of 30-130 nm in diameter containing miR-122 and RNU6-2 miRNAs were isolated. A concentration of 12 ng/μL of miRNAs and 10 μg/μL of proteins were recovered. Albumin and HDL were decreased whereas total proteins and LDL were significantly increased in

affected compared to healthy dogs. As shown in an experimental study performed in mice, a significant decreased expression of miR-122-5p was seen in leishmania infected dogs compared with healthy ones.

This study suggests that alterations of circulating lipoproteins associated with a low expression of exosomal miR-122 indicate a liver dysfunction in dogs naturally affected by *Leishmania infantum*.

Disclosures

No disclosures to report.

ESVIM-P-8

Anemia and hypoferremia in cats with hepato-pancreatic and intestinal involvement

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In veterinary medicine, although red blood cells (RBC) and iron serum levels seem to be influenced by inflammation, specific investigations regarding red blood cell parameters and iron serum levels in enteropathic cats are lacking.

The aim of this study was to investigate which type of anemia and how are the serum iron levels in cats with hepato-pancreatic and intestinal involvement.

A retrospective review was conducted on ten-year medical records of cats presented to the University Veterinary Teaching Hospital, looking for ultrasonographic signs of concurrent inflammation of at least two organs among liver, pancreas and intestine. Cats were included if information about clinical signs and laboratory tests (CBC and serum iron level) were available.

Sixty-three cats met the inclusion criteria and were enrolled in the study. Patients were divided into two groups according to ultrasonographic signs: cats with concurrent involvement of pancreas, liver and intestine (Group A, n = 19) and cats with concurrent involvement of only two organs between pancreas, liver and intestine (Group B, n = 44). Differences between groups were statistically investigated by Mann-Whitney test for iron, and Unpaired *t*-test for anemia parameters. Categorical data were analyzed with Fisher's exact test.

Twenty-nine cats (46%) showed anemia which was more frequently mild (Hct < 26%; 62.1%) or moderate (13% < Hct < 19%; 31%), normocytic-normochromic (72.4%), and non-regenerative (86.2%). Microcytosis was an infrequent finding (6.3%), and only two cats had microcytosis, anemia and hypoferremia concurrently. Hypoferremia (serum iron <90 mcg/dL) was present in 34 cats and concurrent anemia was observed in 15 cats (without any association between these two parameters). Both hypoferremia and anemia were more severe in group A (median serum iron 60 mcg/dL; mean RBC 6.06 M/μL; $P = 0.0321$) than group B (median serum iron 90.5 mcg/dL; mean RBC 7.00 M/μL). Moreover, anemia was more frequently present in group A (63%; $P = 0.0321$). A ROC curve was used to determine the optimal cut-off of serum iron to identify cats with hepato-pancreatic and intestinal involvement. Cats with serum iron lower than 61.5 mcg/dL were more frequently belonging to group A (sensitivity 82.2%; specificity 52.6%; $P = 0.0048$).

The most plausible hypothesis for the origin of anemia was the presence of a chronic disease. Decreased serum iron levels may be considered as a marker of inflammation in enteropathic cats. Hepato-pancreatic and intestinal inflammation may cause more severe hypoferremia, erythropoiesis suppression, and anemia.

Disclosures

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ESVIM-P-10

Lung ultrasound findings in dogs using a regionally based protocol (Vet BLUE) versus entire thorax scanning

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Lung ultrasound (LUS) can be used to detect comet-tail artifacts (B lines) in animals with trauma, pulmonary edema or alveolar-interstitial syndrome. The scanning protocols used in previous studies include four anatomic sites on each hemithorax (Vet BLUE) and scanning for all intercostal spaces. The regionally based protocol has the advantage of quickly assessing critically ill patients in respiratory distress; however, it is unclear how the results from Vet BLUE protocol correlate with the findings from entire thorax scanning (ETS). The present study aimed to compare the frequency and numbers of B lines, as well as other parenchymal abnormalities, between the Vet BLUE and ETS protocols. We hypothesized that B lines would be more frequently detected by ETS but not significantly affect the final conclusion.

Thirty-four dogs with various clinical problems (16 with cardiac disease, 12 with cardiorespiratory comorbidities, 5 with respiratory disease, and 1 with non-cardiorespiratory problem) were recruited in this prospective study. Dogs that were uncooperative or too critically ill to tolerate the two scanning protocols were excluded. All scans were performed by a single clinician who had completed an LUS training session. The thorax of each dog was first scanned by the Vet BLUE protocol and then by ETS without hair clipping.

Compared with the Vet BLUE protocol, B lines were significantly more frequent in ETS (26.5% vs. 50.0%, $P = 0.004$). Assessment of the severity of B lines (absent, rare, numerous, or confluent) showed more severity in ETS than that of B lines by the Vet BLUE protocol ($P < 0.001$). Nevertheless, when a final conclusion was drawn based on ≥2 positive sites (>3 B lines) per hemithorax as used in the previous studies, the final conclusions of the two protocols were not significantly different ($P = 0.25$), and agreement between the two protocols was substantial (kappa = 0.72). The detection of other parenchymal lesions by the two protocols was not significantly different ($P = 0.5$).

In conclusion, these results suggest that LUS findings from the Vet BLUE protocol substantially agree with those from ETS. However, it should be brought in mind that the most severe lesions on LUS may be underestimated using a regionally based protocol.

Disclosures

Disclosures to report.

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ESVIM-P-11

Comparison of plasma metabolomic profiles of healthy adult cats with low or high plasma homocysteine concentration

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Several prospective observational studies providing data on plasma homocysteine concentration in healthy cats revealed a larger-than-expected inter-individual variability. Investigations conducted on healthy laboratory cats suggested impaired homocysteine metabolism in some individuals, using a methionine loading test. The aim of our study was to determine differences in the metabolomic profile of healthy cats exhibiting low or high plasma homocysteine concentration, respectively.

We used left-over frozen plasma samples from client-owned healthy adult cats previously enrolled in a prospective observational study that aimed at determining a reference interval for plasma homocysteine concentration in the feline species. We extracted 10 individuals with high plasma homocysteine concentration, and we selected 10 epidemiologically-matched individuals with low plasma homocysteine concentration. Plasma samples were subjected to a targeted metabolomics analysis using an AbsoluteIDQ p180 kit (Biocrates Life Sciences AG), assessing six biochemical classes: acylcarnitines, amino acids, biogenic amines, glycerophospholipids, sphingolipids and hexoses.

The orthogonal partial least-squares discriminant analysis identified a model discriminating plasma samples from the two populations of healthy cats (R^2X = 0.665, R^2Y = 0.959, Q^2 = 0.715). The cross-validation performance of the model was confirmed by analysis of variance (CV-ANOVA P-value = 0.013). Based on this analysis, 56 differentiating metabolites were identified. Cats with high plasma homocysteine concentration exhibited higher concentrations of 29/90 glycerophospholipids, 9/15 sphingolipids, 7/21 biogenic amines, 3/21 AMino acids, and 2/40 acylcarnitines, and showed lower concentrations of 3/21 AMino acids, 2/90 glycerophospholipids and 1/21 biogenic amine, compared to cats with low homocysteine concentration.

This is the first study to investigate the feline metabolome, in relation to the plasma homocysteine concentration. Our results suggest that the inter-individual variability of plasma homocysteine concentration in healthy cats may be associated with metabolic peculiarities. Several discriminating amino acids and biogenic amines are involved in pathways of methionine metabolism. Additionally, higher concentrations of plasmalogens in cats with high plasma homocysteine concentration may indicate enhanced oxidative stress or increased activity of protective mechanisms against oxidative stress.

Disclosures

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ESVIM-P-12

Association between immune-mediated hemolytic anemia (IMHA) and acute pancreatitis in dogs

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Evidences in humans, rats and cats suggest that acute pancreatitis is a complication of hemolysis, including hemolysis from IMHA. This study investigates the association between IMHA and pancreatitis in dogs.

Case control study with nested retrospective cohort study, including 3 groups of 95 dogs matched for age, breed, and sexual status: dogs with IMHA (group-1), clinically healthy dogs (group-2), sick dogs without IMHA (group-3). Dogs in group-1 had a HCT < 30% (reference interval [RI], 38.6%" — 54.5%) and positive anti-erythrocyte membrane antibodies test. Acute pancreatitis was diagnosed if dogs had: amylase and lipase concentrations above RI (>1101 mg/dL and > 725 mg/dL, respectively), CRP concentration > 3.0 mg/dL (RI, 0.01 — 0.22), and anorexia and/or vomiting. Frequency of pancreatitis was compared among the 3 groups (fisher exact test). After admission dogs with IMHA with and without pancreatitis had a 7-day follow-up. If some of the dogs with IMHA without pancreatitis developed pancreatitis, they switched to the IMHA with pancreatitis group and were followed-up for 7 days after the pancreatitis event. Free plasma hemoglobin (Hb_{fp}) for dogs with IMHA with pancreatitis (at the time of pancreatitis diagnosis) and without pancreatitis (at presentation) was calculated (Hb_{fp} = Hb_{tot} -[RBC x CHCM x MCV]/1000) and compared (Mann-Whitney test). ROC curve analysis was used to identify the best Hb_{fp} cutoff value (Youden index) discriminating IMHA dogs with and without pancreatitis. Relative Risk (RR) of developing pancreatitis in dogs with IMHA and Hb_{fp} concentration ≥ the Youden Index, was calculated. Finally, 7-day mortality between IMHA dogs with and without pancreatitis was evaluated (chi square test).

At presentation, frequency of pancreatitis was significantly (P = 0.0013) higher in dogs with IMHA (12/95) compared to the controls (0/95 group-2 and 5/95 group-3, respectively). During the 7-day follow-up period a further 9 dogs with IMHA developed pancreatitis. IMHA dogs with pancreatitis (n = 21) had a significantly (P = 0.0067) higher Hb_{pf} (median = 0.17 g/dL, range, 0.0 — 1.55) compared to the IMHA dogs without pancreatitis (n = 74; median = 0.0 g/dL, range, 0.0 — 2.89). The Youden index for Hb_{pf} was 0.08 g/dL (sensitivity = 61.9%, specificity = 75.7%; AUC = 0.672, 95%CI, 0.569 — 0.765; P = 0.0096). In dogs with IMHA, a Hb_{pf} concentration ≥ 0.08 g/dL resulted in an increased risk of pancreatitis (RR = 2.54, 95%CI, 1.51 — 4.29). There was no difference in mortality (P = 0.211) between IMHA dogs with (8/21) and without (18/74) pancreatitis.

Dogs with IMHA are at increased risk of pancreatitis. Several dogs developed pancreatitis after being diagnosis with IMHA, this may suggest that IMHA may cause pancreatitis and that Hb_{fp} is a trigger for it.

Disclosures

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ESVIM-P-13

Evaluation of Serum Procalcitonin in Dogs with Induced Endotoxemia as a Biomarker for Sepsis

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Sepsis is a leading cause of death in veterinary patients and remains challenging to diagnosis in a prompt and accurate manner. Procalcitonin (PCT) has been studied extensively in human medicine and is commonly used as a biomarker for sepsis. Little information regarding PCT as a biomarker for sepsis in dogs exists, with no controlled studies evaluating the kinetics of PCT during sepsis.

The aim of this study was to evaluate the response of serum PCT in dogs with experimentally induced endotoxemia. We hypothesized that PCT would be rapidly detectable in serum after injection of LPS and would remain elevated for at least 24 hours.

Six healthy mixed breed dogs were included in this study. Experimental endotoxemia was induced by injecting healthy dogs with lipopolysaccharide (LPS; 2 ug/kg, IV, once). Vital signs were monitored and serial blood samples were collected for measurement of serum PCT for 72 hours following LPS injection. Difference in mean serum PCT between serial time points was assessed using a mixed effects model.

All dogs developed lethargy and gastrointestinal signs within 30 minutes of LPS administration. Within 1 hour of LPS administration all dogs experienced a decrease in mean arterial pressure (MAP) and within 2 hours of LPS administration all dogs developed a fever. All dogs had serum PCT concentrations above baseline by 2 hours post LPS administration ($P = 0.0002$) with an average peak at 4 hours. Average serum PCT concentrations remained significantly elevated at each two-hour time point through 12 hours post LPS administration, and were non-significantly elevated at 24 hours but remained above baseline for 48 hours.

The results of the current study suggest that PCT was expressed in response to experimentally induced endotoxemia and the kinetics of the PCT expression were favorable and support its use as a biomarker for sepsis in dogs. Additionally, as changes in serial measurements were in agreement with clinical evaluation of improvement, PCT may have an additional role in prognostication and therapeutic decision-making.

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Disclosures to report.

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ESVIM-P-14

Effect of a weight loss program on metabolic and immunological profile, blood leptin level and cardiovascular parameters in obese dogs

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In these last years, the increasing obesity incidence in canine species has enshrined its relevance as an important worldwide disease. Recently, obesity has been associated with impaired immunity and chronic low-grade inflammation in humans as well as mouse models. Increased concentration of leptin and other pro-inflammatory cytokines have been described in obese dogs. A reduced number of T regulatory cells (Treg) has been reported in visceral adipose tissue and blood of obese humans, and recently also in Labrador retriever obese dogs. Moreover, some evidences addressed the possible impact of obesity on cardiovascular apparatus in dogs. The aim of this study was to investigate the effect of a weight loss program on metabolic and immunological profile, blood leptin level and cardiovascular parameters in obese dogs. Ten overweight dogs (OB) (BCS > 7/9) were recruited into the study, and they underwent blood testing (complete blood count, serum biochemistry, blood level assay of CD3 + CD4+, CD3 + CD8+ T cells, CD4/CD8 ratio, CD21+ B cells, Treg cells by immune-fluorescence and flow cytometry and measurement of serum leptin by species-specific ELISA kit) and assessment of cardiovascular function (blood pressure measurement, electrocardiography and echocardiography) before (T0) and after five months (T1) of commercially available weight loss diet. Ten normal weight (BCS 4-5) healthy dogs represented a control group (CTR). Regarding metabolic profile, a mild no significant decrease in total cholesterol but a significant decrease in triglycerides serum levels ($P < 0.05$) were observed in the obese dogs at T1. There were no significant differences in the other biochemical parameters as well as in haematological values between the two observation times. Concerning the cardiovascular parameters, no significant differences were observed at T1, and particularly systolic arterial blood pressure values were in the reference range in both times. OB had elevated serum leptin concentrations that decrease significantly ($P < 0.005$) after weight loss, however remaining higher compared to CTR. OB dogs showed significant low levels ($P < 0.005$) of Treg compared to CTR but they did not increase after weight loss. Our data suggested that a deranged immune-regulation, combined with high leptin levels, might characterize obese dogs in the absence of cardiovascular alterations. Furthermore, on the basis of our results we may suggest that probably in dogs a greater reduction in fat mass, and long-term weight loss programs, are necessary in order to restore immunological balance.

Disclosures

No disclosures to report.

ESVIM-P-15

Life expectancy and causes of mortality of dogs at the National Veterinary School of Toulouse between September 2007 and September 2017: retrospective study

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Data on canine life expectancy are important for the owners, veterinarians and assurance companies, but they are sparse in Europe (United Kingdom excepted), especially in France.

The aim of this retrospective cohort study was to determine the life expectancy and causes of death of dogs seen at the Toulouse Veterinary Teaching Hospital (TVTH, France) and the effect of various factors (breed, gender, size) on these data.

The database of the TVTH was searched for dogs recorded dead between September 2007 and 2017. Their records were screened to determine the age at death and classify its cause between neoplastic, cardiovascular, behavioral, dermatological, endocrinological, gastroenterological, hematopoietic, infectious, musculoskeletal, neurological, ophthalmological, reproductive, respiratory, trauma, age-related and uronephrological causes. Breed and weight category (< 10, 10-25, 25-45 and > 45 kg) were recorded if specified. Results were analyzed using commercial software (R® and Excel®). Log rank test was used for life expectancy; the effect of breed, gender, reproductive status and weight category was assessed by Khi2 test and multivariate analysis.

A total of 3256 dogs belonging to 151 breeds were included; the number of dogs in each breed varied from 1 to 224. Cause of death and weight could be identified in 2164 and 3054 dogs respectively.

The median life expectancy of dogs was 10.7 years (range [0-24]). Weight had a significant effect on longevity, dogs <10 kg (n = 848) and dogs >45 kg (n = 107) living respectively longer (median 12.7 years) and shorter (median 6.1 years) than dogs weighting 10-25 (n = 723) and 25-45 kg (n = 1254) (median longevity 9.9 and 10.4 years respectively, $P < 0.05$). Other factors significantly associated with increased longevity in dogs were being female, spayed, and mixed breed ($P < 0,01$).

Neoplasia was the most common cause of death (30%), especially in Boxers and Bernese. Breed's influence on the cause of death was significant ($P < 0,05$): Brittany Spaniels died mostly after dermatological disorders, Cavalier King Charles and Bulldogs from respiratory causes and Cavalier King Charles and Spitz from cardiovascular diseases. Under 7 years, the most common causes were trauma (15%) and infections (7%). Compared to the whole population, intact females were more likely to die from reproductive-related and endocrinological causes, neutered females from neoplastic disorders ($P < 0,01$).

This study suggests that canine longevity is highly influenced by weight. The dominant causes of death in French dogs varies with breed, gender and reproductive status. This study has to be enlarged to first opinion practice.

Disclosures

No disclosures to report.

ESVIM-P-16

Comparison of habitual physical activity levels in French Bulldogs and normocephalic dogs - a pilot study

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In addition to respiratory difficulties, brachycephalic obstructive airway syndrome (BOAS) causes exercise intolerance. Daily habitual physical activity can be quantified as counts by accelerometers measuring frequency, duration and intensity of activity. The first aim of this study was to establish the cut points for sedentary, moderate and high activity and the second to compare the activity levels in French Bulldogs with moderate or severe signs of BOAS (BOAS+), French bulldogs with none or mild signs of BOAS (BOAS-) and normocephalic dogs.

Cut off points for Actical accelometer readings for sedentary (lying, slight movement of trunk), moderate and high (trotting at speed of 8 km/h or more) were collected with one-minute epoch lengths for sedentary (100 epochs) and high (117 epochs) activities. The upper limit for sedentary and lower limit for high activity was defined as mean +/−2 x SD (SD) and moderate as values between these. BOAS + (n = 10) dogs, BOAS - (n = 9) dogs and normocephalic (n = 11) dogs of comparable body sizes wore the Actical collar over seven consecutive days. Statistical comparisons in activity levels between groups were performed with an ANOVA method, Tukey's correction was used in comparisons.

For sedentary activity, established cut point was 347 and for high activity 1343. All dogs spent most of the time at sedentary activity (91% ± 3% for BOAS+ dogs, 89% ± 3% BOAS- dogs and 85% ± 5% controls; mean, SD). Significant differences were found only between BOAS+ and normocephalic dogs at sedentary and high activity percentages ($P = 0.005$ and 4% ± 1%, 7% ± 3% $P = 0.01$; respectively).

In conclusion, dogs with marked BOAS signs spend less time at high activity and more time at sedentary activity than normocephalic dogs.

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ESVIM-P-17

Normal or mild increased C-reactive protein values in 16 dogs with bronchial and pulmonary infection with Bordetella bronchiseptica

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C-reactive protein (CRP) is a well-known acute phase protein in dogs. It has been recently shown to be particularly useful in discriminating bacterial bronchopneumonia from other pulmonary diseases and promising in guiding antibiotic therapy duration. However, in those publications, dogs infected by *Bordetella bronchiseptica*(Bb) have not been specifically distinguished from others. In an experimental study, high elevations of CRP have been identified in dogs inoculated with Bb. Nevertheless, the amplitude of increase of CRP in naturally-infected dogs with Bb has not been described.

The aim of this study was to describe the values of CRP in dogs with lower airways Bb infection with or without radiographical pulmonary involvement. Magnitude of CRP elevation was also compared with dogs diagnosed with bacterial aspiration bronchopneumonia (ABP).

Sixteen dogs with lower airways Bb infection and 36 dogs with ABP were selected. For each included dog, a CRP value and thoracic radiographs at diagnosis were available. Bb infection was confirmed by

bacterial culture and/or quantitative PCR on bronchoalveolar lavage fluid. ABP was diagnosed based on compatible history, physical examination and radiographic findings and favorable evolution on empiric antimicrobial therapy.

Median age of dogs with Bb infection and ABP were 0.6 and 5 years respectively ($P < 0.001$). Eleven dogs with Bb infection had alveolar lesions on radiographs with only one out of 11 dogs having another bacterial coinfection. CRP value was mildly elevated in 11/11 dogs and 1/5 dogs with and without alveolar lesions, respectively ($P = 0.002$) and the median CRP value was significantly higher in dogs with alveolar lesions compared with dogs without alveolar lesions (20 mg/L, [14-38], versus 5 mg/L, [5-11], $P = 0.002$). Duration of clinical signs was longer than 2 weeks for all Bb dogs; duration was not different between dogs with normal or elevated value as well as between dogs with or without alveolar lesions. In dogs with Bb infection and alveolar lesions, median CRP value was significantly lower than in dogs with ABP (17 versus 118 mg/L, $P < 0.001$) and ranges of elevation of CRP did not overlap between these two groups ([15-38 mg/L] and [55-270 mg/L] respectively).

In conclusion, regardless of the presence of alveolar lesions, Bb may be suspected in coughing dogs with normal to slightly elevated CRP suggesting that CRP is not a good marker to hep guiding type and duration of treatment in bordetellosis.

Disclosures

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ESVIM-P-18

Polycythemia is uncommon in dogs with chronic hypoxic pulmonary disease

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Prolonged tissue hypoxia caused by chronic pulmonary disease is commonly cited as an important mechanism in the development of secondary physiologically appropriate polycythemia. However, the prevalence and severity of polycythemia has not been described in detail in dogs with chronic hypoxic pulmonary disease.

44 dogs (median age 11.9, interquartile range 10.0-13.3 years) with chronic pulmonary disease and documented chronic hypoxia (partial pressure of arterial oxygen [PaO_2] <80 mmHg on at least two separate arterial blood gas measurements minimum of 1 month apart) were retrospectively identified from patient records. Dogs of sight hound breeds were excluded. The diagnosis was based on thorough clinical examinations including bronchoscopy and brochoalveolar lavage, thoracic computed tomography, lung histopathology or a combination of the aforementioned. Statistical correlation between PaO_2 and red blood cell parameters was analyzed using Pearson's correlation coefficients.

The group comprised of 23 West Highland white terriers with canine idiopathic pulmonary fibrosis and 21 dogs of other breeds with the following lung diseases; chronic bronchitis 13/21, eosinophilic bronchopneumopathy 2/21 and interstitial lung disease 2/21. In 4/21 dogs with chronic respiratory signs (cough and/or tachypnea) and chronic diffuse radiographic lung changes, a final diagnosis was not established.

Median duration of hypoxia was 8 months (IQR 4-18 months) and the mean PaO_2 at the end of the period was 62.2 mmHg (SD [STD] $\pm$ 9.9 mmHg). Red blood cell parameters measured at the end of hypoxemia period were within the laboratory reference range in majority of dogs; Erythrocyte count (Erytr) was normal in 35/44 dogs (mean 7.4 $\pm$ STD 0.9 $x10^{12}$/L, range 4.9-9.3 $x10^{12}$/L, laboratory reference range 5.3-8.0 $x10^{12}$/L), hemoglobin concentration (Hb) was normal in 42/44 dogs (mean 173 $\pm$ STD 21 g/L, range 121-221 g/L, laboratory reference range 140-203 g/L) and hematocrit (Hkr) was normal in 39/44 dogs (mean 51.0 $\pm$ STD 5.7%, range 35-64%, laboratory reference range 38-57%). Marked polycythemia (hematocrit ≥65%) was not noted in any of the dogs. Red blood cell parameters were not correlated with the severity of hypoxia (correlation to PaO_2: Erytr $r = -0.054$, $P = 0.728$; Hb $r = -0.141$, $P = 0.360$; Hkr $r = -0.99$, $P = 0.521$).

These results indicate that polycythemia is uncommonly encountered in dogs with chronic hypoxic pulmonary disease and when encountered, only mild increases in red blood cell parameters are noted.

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ESVIM-P-19

A statistical analysis to predict persistence of canine sinonasal aspergillosis at endoscopic follow-up by comparing three different scoring systems: a retrospective study of 47 cases treated with one hour 1% clotrimazole per-endoscopic infusion and undergoing endoscopic follow-up

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The aim of this study was to investigate three different scoring system for Canine Sinonasal Aspergillosis (CSA) in order to predict whether they can indicate persistence of CSA at endoscopic follow-up.

Medical records of dogs treated for CSA were reviewed. Inclusion criteria were the presence of a full medical history, radiological investigation, after meticulous debridement of the fungal plaques and endoscopic follow-up between 30 and 90 days after treatment. Forty-seven dogs fulfilled the inclusion criteria and were included in this study.

To each dog a CSA grade score was given according to Sharp (1989) at the time of diagnosis; a rhinoscopic scoring according to Zonderland (2002) was given after endoscopic examination. The score was modified by the authors by creating three categories of severity: mild CSA (score between 0 and 5), moderate CSA (score between 6 and 10) and severe CSA (score between 11 and 16). The amount of fungal plaques was considered mild when countable, moderate when

partially occupying the sinonasal cavity and abundant when obliterating the sinonasal cavity. A clinical scoring proposed by Schuller (2007) was modified and used to assess the patients prior to endoscopic follow-up: dogs were classified as having no (no clinical signs reported), mild (occasional sneezing, occasional reverse sneeze, serous to mucoid discharge), moderate (mucopurulent discharge, frequent sneezing, frequent reverse sneezing) or severe (severe mucopurulent discharge, stertor, persistent reverse sneezing, persistent sneezing, epistaxis, generalized malaise) clinical signs.

The three scoring system were statistically analised and explicitly, we addressed the question: is there any chance to predict the persistence of fungal infection based on the score system outcome? If yes, than that scoring method was considered predictive for fungal infection persistence. A contingency table for each scoring system was computed and the null hypothesis of independence between the scoring methods and persistence of fungal infection was statistically tested by Fisher-Freeman-Halton Exact Test.

According to the statistical results, the Sharp grade score system can be considered significantly associated with persistence, or not persistence, of fungal infection at the endoscopic follow-up, and therefore it can be considered as a predictor of the fungal infection persistence (Sharp score system Fisher P-value = 0.021).

Endoscopic follow-up is strongly recommended to assess CSA treatment effectiveness. Sharp grade score can be used to predict treatment outcome in dogs treated with one hour 1% clotrimazole per-endoscopic infusion. Sharp grade score might apply also to other different methods for treating CSA; further studies are warranted.

Disclosures

No disclosures to report.

ESVIM-P-20

Evaluation of feline packed red blood cell units obtained by blood sedimentation and stored for 42 days for transfusion purposes

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Component therapy involves separation of whole blood (WB) into its components (packed red blood cells -PRBCs- and plasma), for specific replacement therapy and to reduce transfusion reactions. In cats, blood for transfusion is commonly collected using an open system and administered as WB, in part because of the challenge of preparing components from a small blood volume. Feline blood has a high erythrocyte sedimentation rate; therefore, if the syringe containing collected blood is placed upright, plasma can be removed from the red cells shortly after collection for separate storage of plasma and PRBCs. The aim of this study was to assess the characteristics of feline PRBC units obtained by blood sedimentation both at collection and after storage for 42 days.

Blood was collected from fourteen feline blood donors into three 20 mL syringes pre-charged with CPDA-1:blood ratio of 1:7 using an open system. A pre-donation CBC was performed in each donor. The three syringes were allowed to sediment for approx. 1 hour at room temperature. Then plasma was aseptically expressed into plain transfer bags and RBC expressed into another transfer bag pre-charged with 10 mL of SAG-M. PRBCs units were stored in a blood-dedicated refrigerator and sampled using blood bag segments at preparation time (D0) and after 42 days storage (D42). On pre-donation blood and on PRBC units at D0 and D42 the following parameters were evaluated: I) hematological parameters (RBC, Hb, Hct, WBC, PLT); II) percentage hemolysis; III) morphological index (only for PRBC units), scored of 0 to 3 based on echinocyte transformation of the normal discocyte; IV) aerobic and anaerobic blood culture (only for PRBC units).

From donor to PRBC units there was a significant increase in RBC count (mean increase +1886 $\pm$ SD1399 $\mu L/10^3$), Hb concentration (+2.8 $\pm$ 2.2 g/dL), Hct percentage (+8.3 $\pm$ 5.5%). Significant reduction was found in PLT count (–249 $\pm$ 189 $\mu L/10^3$). Comparing PRBC at D0 and D42 a significant increase was found in percentage hemolysis (+ 1.2%), morphological index (+ 0.9) and a significant reduction in RBC count (–460 $\pm$ 679 $\mu L/10^3$), Hct percentage (–3.2 $\pm$ 3.5%), WBC count (median – 2589 $\mu L/10^3$), and PLT count (median – 43 $\mu L/10$). All blood cultures were negative for bacterial growth.

PRBC units obtained by sedimentation of donated blood appear to be a suitable blood component for treatment of normovolemic anemia. However storage for 42 days, as suggested for canine and feline PRBC units, resulted in significant hematological changes that could reduce oxygen delivery after transfusion.

Disclosures

No disclosures to report.

ESVIM-P-21

Bronchoscopic findings in dogs with bronchial vegetal foreign bodies: a retrospective study of 52 cases (2010-2019)

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Vegetal foreign bodies are a frequent cause of cough in dogs during spring and summer in France. They can result in a variety of other clinical signs and endoscopic abnormalities. The aim of this study was to describe the foreign body location, rate of retrieval success, complications, and macroscopic endoscopic findings in dogs with vegetal foreign bodies. Fifty two dogs were included in this retrospective evaluation from the medical records of dogs admitted between 2010 and 2019 for a bronchial vegetal foreign body. Diagnosis was based on direct visualisation during the bronchoscopic exam or after surgical retrieval.

27 males and 25 females were included in this study. More than half were hunting dogs (29/52). Duration of clinical signs ranged from 1 to 1400 days. Cough was the main clinical sign (49/52). Foreign bodies were removed from a right-sided bronchus in 35/52 (67%) cases, from a left-sided bronchus in 11/52 (21%) cases, from both right and left bronchi in 6/52 (12%) cases. Endoscopic retrieval was successful in 43 of 52 cases (83%). Nine dogs needed a surgical treatment. The survival rate was 100%.

Endoscopic images were reviewed in 44 dogs. They showed purulent exsudate in the ventral larynx region (29/44), the trachea (38/44) and the bronchi where the foreign body is located (43/44).The presence of large bronchial nodule or an irregular mucosal surface were other frequent observations (36/44). A mild bleeding was the main complication (30/44).

This retrospective study confirm the safety and usefulness of bronchoscopy in diagnosis and treatment of bronchial vegetal foreign bodies in dogs. Mucosal nodules associated with purulent material within the airways are frequent endoscopic findings.

Disclosures

No disclosures to report.

ESVIM-P-22

Assessment of nasal microbiota in healthy dogs of different breeds

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Dolichocephalic breeds are predisposed to sinonasal aspergillosis while brachycephalic dogs are not affected. Since disruptions in the resident microbiome may contribute to disease pathogenesis by modulating immune responses and since microbiota dictates the type of host-fungus relationship, we hypothetized that differences in predisposition to nasal disease in dogs could be associated with differences in core nasal microbiota between breeds.

Fourty-six healthy dogs were recruited, including 22 medium to large dogs from dolichocephalic breeds (DC), 12 brachycephalic dogs (BC) and 12 terrier dogs (T). All dogs were older than 14 months. Dogs were living either in rural (n = 21) or urban domestic conditions (n = 25). Nasal swabs were obtained under anesthesia and banked at −80°C until batched analysis. After DNA extraction, a PCR targeting the V1-V3 region of the 16S rDNA was performed. Amplicons were then sequenced on a MiSeq Illumina sequencer. Taxonomical assignation and microbiota community analysis were done with MOTHUR V1.41.0 with an OTU clustering distance of 0.03.

Analysis of ecological indexes showed that bacterial richness (P = 0.01) and α-diversity (P < 0.01) were significantly higher in BC group compared to the two other groups. The AMOVA analysis indicated that the BC group was different compared to DC and T groups (BC vs DC P = 0.02; BC vs T P < 0.01). There was no difference in bacterial load between groups and no effect of the living conditions.

The nasal microbial population was predominantly composed of the phyla *Proteobacteria*(mainly represented by the family *Moraxellaceae*), *Actinobacteria*, *Firmicutes* and *Bacteroidetes*, in agreement with previous studies. However, within each breed group, the relative abundance in phyla was highly variable and no significant differences were found between groups while at the family level, the relative abundance in *Pasteurellaceae* was significantly higher in the BC group. Distinct species and genera were found as indicators of discrimination (P < 0.05) among which 8 species and 9 genera in the BC group and 1 species in the T group.

Our study mainly demonstrated significant differences in the nasal microbiota in the BC group compared with the two other groups. Such differences might be associated to a particular facial morphology and/or breathing pattern in brachycephalic dogs. We did not identify nasal microbiota breed-differences that would be in favor of a breed susceptibility of dolichocephalic dogs for nasal diseases. Further studies are needed to investigate the role of nasal microbiota variations as a trigger or a perpetuating factor in nasal diseases, especially in dogs with sinonasal aspergillosis.

Disclosures

No disclosures to report.

ESVIM-P-23

Canine sino-nasal aspergillosis in Italy (38 cases)

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Sino-nasal aspergillosis (SNA) is an important cause of chronic nasal disease in the dog, characterized by the formation of a superficial mucosal fungal plaque within the nasal cavity and/or frontal sinus of systemically healthy dogs. *Aspergillus fumigatus* is classically recognized as the most important etiological agent. Notably, this species is now considered a "group" (the Fumigati group) including several species distinguishable only via molecular exams. To date, few studies have analyzed fungal isolates involved in canine SNA cases at genetic level, showing that *A. fumigatus sensu stricto* was the main causal species. On the opposite, other species of the Fumigati group (the so called criptic species of the viridinutans complex) have been found associated to SNA in cats (in cats SNA is more invasive and has a poor prognosis, due to the involvement of the retro-orbital space). The aim of this study was to described the clinical and mycological features of a series of 38 cases of canine SNA in Italy. Cases were recruited by veterinarians belonging to ENDOVET (a group specialized in endoscopy). The diagnosis was achieved by a combination of exams (endoscopic visualization of fungal plaques and/or visualization of fungal elements at cytology/histology of nasal biopsies). Fungi were isolated from biopsies and identified by morphological keys and molecular analyses (sequencing of the ß-tubulin gene). Twenty isolates chosen randomly were tested *in vitro* against some antifungal agents commonly used in the dog with SNA (itraconazole, clotrimazole, enilconazole) and other agents more used in human patients (voriconazole, posaconazole).*A.fumigatus* sensu stricto was identified in most of the cases (34/38, 89%), with the remaining cases due to *A. flavus*, *Fusarium spp. and Paecilomyces spp*. (for one case a definitive identification was not achieved). *In vitro* MIC (Minimum Inhibitory Concentration) values were within expected limits, therefore resistance was not detected. A breed predisposition was not noted. On the contrary, in agreement with some past studies, a sex predisposition could be noted, since males accounted for 66% of the sample. Clinical signs more frequently observed are those already reported in the literature (eg. nasal discharge was present in 100% of the cases, it was monolateral in 82%). Frontal sinus involvement was confirmed in 47% of cases. Although

data on follow up visits were incomplete, the most effective treatment seemed to be represented by mechanical debridement of fungal plaques plus local treatment using clotrimazole cream or enilconazole solution (procedures conducted during endoscopy).

Disclosures

No disclosures to report.

ESVNU-P-1

Uroliths in dogs from Europe and China - a comparative study

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Our study aimed to evaluate and compare the nature of canine uroliths. The study involved 4204 dogs from predominantly Central Europe (CE) and 830 dogs from China (CN) during the year 2016-2018. Analyses were performed using infrared spectroscopy.

The global statistics of uroliths in CE:CN dogs were male (40%:45%), female (24%:31%), neutered (18%:15%) and spayed dogs (17%:9%). The population of CE:CN dogs consisted of 511:71 mongrels and 163:45 breeds.

The uroliths comprised struvite (CE:CN = 43.3%:48.3%), calcium oxalate (33.2%:41%), cystine (13.3%:4.8%), ammonium urate (5.1%:1.9%) and others (<5%). The CE:CN dogs had median ages (years): struvite (7:6), calcium oxalate (9:8), and cystine (5:3).

Struvite uroliths in CE:CN dogs accounted for: females (81%:81%), spayed (79.5%:74.4%), males (17%:33.9%) and neutered dogs (17%:26.5%).

Calcium oxalate uroliths in CE:CN dogs were: males (43.5%:46.6%), neutered (53.6%:42.8%), females (15%:15%) and spayed dogs (15%:15%).

Cystine uroliths in CE:CN dogs accounted for: males (28.6%:9.1%), neutered (9.6%:2.4%), females (1.2%/1.2%) and spayed dogs (0.7%/0%).

In CE cohorts, cystine uroliths were present in 11/13(=84.6%) of Irish terriers, a breed reported to have a genetic predisposition to cystine calculi. Cystine uroliths were also found in American Staffordshire terriers (25/30 = 83.8%), bulldogs (39/77 = 50.6%), French bullterriers (30/71 = 42.3%), huskies (5/12 = 41.7%) and Chihuahuas (61/177 = 34.5%). In CN cohorts having no Irish terriers population, cystine uroliths were primarily found in bulldogs (9/11 = 81.8%) and French bullterriers (7/9 = 70%).

Our study revealed that, in both CE:CN cohorts, uroliths were common in the order of struvites, calcium oxalates, cystines and ammonium urates. Among the CE breeds, Yorkshire terriers and Parson Russel terriers were more subject to calcium oxalate while pugs were to struvite formation. Among the CN breeds, miniature schnauzers, bichons and poodles were more subject to struvite uroliths.

Struvite uroliths in CE:CN dogs occurred equally in females but at rates substantially higher than those in males. Within males, CN dogs were 2 times more susceptible than their CE counterparts. Considering the role of urease-producing bacteria in the genesis of struvite uroliths, our data suggest the influence of gender as well as habitats.

Calcium oxalate uroliths were ~3 times more prevalent in male dogs than females in both CE:CN cohorts.

Cystine uroliths were substantially more prevalent in males than female dogs in both CE:CN cohorts. Notably, male CE dogs were 3 times more susceptible than the CN counterparts. Overall, our data suggest that canine urolith formation is widely dependent on breed, gender and environmental factors like diet and regional care practices.

Disclosures

Disclosures to report.

The authors Breu D1 and Wenk C2 are employed at Laboklin GmbH & Co KG, Bad Kissingen, Germany1 and Laboklin GmbH & Co KG, Basel, Switzerland2. Müller, E is owner/manager of the Laboklin GmbH & Co KG, Germany.

ESVNU-P-2

Significant Feline Proteinuria: a retrospective study of its aetiology in 61 cats

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Urinary Protein/Creatinine Ratio (UPC ratio) is currently the most frequent method for proteinuria quantification. Proteinuria can be classified in physiological versus pathological (pre-renal, renal or post-renal). Although chronic kidney disease (CKD) is assumed to be the most common cause of proteinuria in cats, few studies have been conducted to assess its aetiology.

The aim of this study was to determine the main causes of proteinuria in cats from the region of Lisbon, Portugal.

All the cats presented between 2016 and 2018 in a veterinary hospital and identified with significant proteinuria (UPC ratio > 0.4) were included. The cause of proteinuria, International Renal Interest Society (IRIS) staging and systolic blood pressure (SBP) were obtained from medical records.

Sixty-one cats were selected, including 28 females (21 spayed, 7 intact) and 33 males (26 neutered, 7 intact), with an average of 12 years old. No cats had physiological or pre-renal proteinuria, 46/61 (75%) presented renal proteinuria, 4/61 (7%) had post-renal proteinuria and 11/61 (18%) had a suspected mixed origin (renal + post-renal). In detail, the causes of renal proteinuria were CKD (39/61; 64%), CKD complicated with co-morbidities (2/61; 3%), acute kidney injury (AKI) (2/61; 3%) and diseases capable of decreasing renal permselectivity (3/61; 5%). Post-renal proteinuria was due to urinary tract infection (UTI) (3/61; 5%) and cystitis (1/61; 2%). Mixed-proteinuria was attributed to CKD + cystitis (7/61; 11%) and CKD + UTI (4/61; 7%).

Complete IRIS staging was obtained in 30 out of the 52 cats with CKD (58%). Two cats were on stage I (both hypertensive), 8 cats were on stage II (6 hypertensive, 2 non-hypertensive), 11 cats were on stage III (9 hypertensive, 2 non-hypertensive) and 9 cases were on stage IV (8 hypertensive, 1 non-hypertensive).

Regarding SBP, data was available in 36/61 cases. From these, 24/36 (67%) were severely hypertensive, 6/36 (17%) were hypertensive, 5/36 (14%) were pre-hypertensive and one cat was normotensive.

This study strengthens that pre-renal proteinuria is rare in cats and CKD is the most frequent cause of significant proteinuria in this species. Severe hypertension is a common finding, highlighting the relevance of SBP measurement in these cats. The fact that IRIS staging was only possible in 58% of the cats still reflects a poor awareness of veterinarians for this classification.More than contributing to better knowledge of causes of proteinuria, this study suggests that physiological and pre-renal causes do not induce significant proteinuria in cats.

Disclosures

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ESVNU-P-3

The role of vector-borne diseases in the aetiology of overt canine proteinuria: a retrospective study in 106 dogs

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Canine vector-borne diseases (CVBD) are highly prevalent in Southern Europe. They are a well-known cause of glomerular disease in dogs, being commonly associated with a significant proteinuria. However, few studies have evaluated the role of CVBD in the aetiology of proteinuria in these endemic countries. According to its origin, proteinuria can be classified in pre-renal, renal or post-renal and it is currently quantified by the Urinary Protein/Creatinine Ratio (UPC).

The aim of this study was to assess the main causes of proteinuria in dogs from the region of Lisbon in order to estimate the role of CVBD on it.

All dogs presented to a Lisbon veterinary hospital between January 2017 and December 2018 identified with significant proteinuria (UPC ratio > 0.5) and with an established primary diagnosis were selected. Based on the medical records, cases were classified according to proteinuria origin.

106 dogs were selected. From these, 54% were females and 46% were males, with an average of 9.5 years old (range between 0.83 and 16 years old). None had pre-renal proteinuria, 76% had renal proteinuria, 17% had post-renal proteinuria and 7% had a suspected mixed origin (renal+post-renal). Detailing renal proteinuria, 46% had a presumptive diagnosis of glomerulonephritis secondary to CVBD, 27% showed chronic kidney disease, 26% had systemic diseases possibly inducing an impaired glomerular permselectivity (bronchopneumonia, enteropathies, liver disease, hyperadrenocorticism, diabetes mellitus or neoplasia) and 1% had acute kidney injury (AKI). Post-renal proteinuria included dogs with pyometra and urinary tract infection (UTI). Mixed proteinuria was considered in dogs with systemic diseases (CVBD, CKD, AKI and hyperadrenocorticism) and a concurrent UTI and/or cystitis.

Considering dogs diagnosed with CVBD, 82% were positive on serology for a single-agent while 18% were positive for more than one agent. Detailing serology results, 90% of dogs were positive for *Leishmania infantum*, 13% *Rickettsia* spp, 8% heartworm disease, 5% *Borrelia burgdorferi*, 5% *Ehrlichia* spp, 3% *Babesia canis* and 3% *Anaplasma* spp.

Whilst pre-renal significant proteinuria was uncommon, CVBD were the main cause of renal proteinuria in these dogs. Leishmaniosis was the most frequent CVBD identified on serology. Despite the increasing preventive measures, these are expected results since CVBD are highly prevalent in Portugal.

This study suggests that CVBD are the most common differential diagnosis of renal proteinuria in dogs from Lisbon. These results might be extrapolated to other endemic southern European countries, but further studies are needed to confirm it.

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ESVNU-P-4

Evaluation of the diagnostic value of urinary albumin to protein ratio in proteinuric dogs

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Renal protein loss is mainly caused by either tubular or glomerular dysfunction, and the differentiation between these two types holds a high diagnostic and therapeutic significance. Tubular proteinuria is defined by the loss of low molecular weight proteins, whereas in glomerulopathies albuminuria is more pronounced.

We hypothesized that by assessing the urine albumin-to-total protein ratio (uAPR) we will be able to identify the source of proteinuria: whether it is solely of tubular origin or glomerular damage is present as well.

27 proteinuric canine left-over urine samples were used in this study. 11 samples belonged to laboratory beagles and 17 to clinical patients. In addition to urinary protein and creatinine concentration determination, albumin was measured by immunoturbidimetric method. The urinary albumin-to-creatinine ratio (UAC), urinary protein-to-creatinine ratio (UPC), and the ratio of UAC and UPC (uAPR) were calculated. The findings were compared to sodium dodecyl sulfate polyacrylamide gel electrophoresis (SDS-PAGE). Solely tubular proteinuria was considered 'tubular', while mixed (tubular and glomerular) and glomerular proteinuria were considered as 'non-tubular'.

The median (lower and upper quartile) of the UPC was 1.27 (0.48; 3.53), the UAC was 0.51 (0.10; 2.69), and the uAPR was 0.56 (0.15; 0.77) in the urine specimens. With receiver operating characteristic (ROC) curve analysis the ideal cut-off value was determined to be 0.37, below which the tubular proteinuria could be identified with high confidence (sensitivity = 93.75%, specificity = 75.0%, positive predictive value = 82.35%, negative predictive value = 81.81%).

Using this cut-off value, tubular proteinuria was diagnosed in 10 dogs (35.71%) and non-tubular proteinuria in 17 animals (60.71%). The uAPR results showed a high correlation with the electrophoresis outcomes. There was only one dog with a false negative result ('tubular' on uAPR and 'non-tubular' on electrophoresis), and 3 dogs had false positive results ('non-tubular' on uAPR and normal protein excretion pattern on electrophoresis).

Based on these findings we suggest that uAPR could be a simple and affordable method to identify the source of proteinuria, thus the

determination of uAPR may help in the diagnostic and therapeutic decision-making in proteinuric dogs.

The gold standard to differentiate between tubular and glomerular damage is histopathology, thus our future plan is to compare uAPR with histopathological diagnosis.

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ESVNU-P-5

N-acetil-β-D-glucozaminidase index as an early renal tubular damage marker in male cats with obstructive lower urinary tract disease

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Feline lower urinary tract disease (FLUTD) is a recurrent chronic disease commonly found in current practice. Male cats (2-8 years-old), neutered, sedentary and overweight present the highest risk of developing FLUTD.

The aim of the study is to evaluate the activity of the N-acetil-β-D-glucozaminidase (NAG) index as an early tubular damage marker in neutered male cats with obstructive FLUTD.

57 male cats of different breeds and ages, diagnosed with obstructive lower urinary tract disease were included in this study. Diagnosis was based on physical examination, haematological, biochemical, radiological and ultrasound exam.

Urine specimens collected via cystocentesis were subjected to complete urinary analysis (urinary biochemistry, urinary sediment, and culture and sensitivity tests). Complementary, urinary NAG index activity was evaluated in these samples.

Higher values of urinary NAG index were found in male cats with obstructive FLUTD and bacterial implication (median value of 55,52 $\pm$ 12,3 U/g) when compared with aseptic obstructive FLUTD were median NAG index values were found to be 33,12 $\pm$ 14,8 U/g.

Prolonged anuria consecutive to uretral obstruction as well as repeated obstructive episodes induces kidney tubular lesions. The use of this marker in current medical practice allows early diagnosis of patients with tubular damage consecutive to urethral obstruction and urinary reflux. Obstructed male cats with UTI have a greater risk of developing tubular lesions especially those with septic cystitis.

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No disclosures to report.

ESVNU-P-6

Non-symptomatic bacteriuria is common in young female boxer dogs

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Reflux nephropathy is considered a possible cause of end-stage kidney disease in young Boxer dogs. Furthermore, vesicoureteral reflux was found to be associated with urinary tract infection. The aim of this study was to evaluate the prevalence of bacteriuria in young Boxer dogs and to describe changes in the urine composition of these dogs.

In 165 clinically healthy Boxer puppies, urine was collected by cystocentesis. Urinalysis including urine culture was performed.

Dogs were examined at the age of 48 to 106 days (median 61 days). There were 22 male and 143 female dogs. Culture was positive in 36 female dogs (25% of the female dogs). None of the male dogs had a positive culture. Of the cultured bacteria, 82% were E. coli. Pyuria was seen in 52% of the culture positive dogs and in 4% of the culture negative dogs. Bacteria in the sediment were seen in 75% of the culture positive dogs and in 5% of the culture negative dogs. The combination of pyuria and bacteria in the sediment was seen in 42% of the culture positive dogs and in 2% of the culture negative dogs. Urine specific gravity ranged from 1.005 to 1.051 (median 1.034) and was significantly lower in culture positive dogs compared to culture negative dogs (median 1.024 vs. 1.037).

Bacterial colonization of the urine is common in young female Boxer dogs. The significance of this finding specifically concerning a possible association to the development of kidney disease has to be determined.

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Disclosures to report.

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ESVNU-P-7

Evaluation of symmetric dimethylarginine (SDMA) in canine acute pancreatitis

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Symmetric dimethylarginine (SDMA) is a sensitive renal biomarker whose concentrations increase earlier than creatinine as glomerular filtration rate decreases. So far in humans, SDMA is considered an important early biomarker of kidney dysfunction.

The aim of this study was to evaluate SDMA in dogs with acute pancreatitis (AP) and their association with kidney injury and severity of disease.

Dogs with AP, presented at the Veterinary Teaching Hospital of University of Pisa between 2017 and 2019, were prospectively enrolled. AP diagnosis was based on compatible clinical and laboratory parameters, abnormal SNAP cPL test (Idexx Laboratories) and positive abdominal ultrasound within 48 h from admission. Dogs with a history of renal diseases (clinical records/history, bloodwork and diagnostic imaging), urinary tract infection and/or on hemodialysis treatment were excluded, along with dogs that had received known nephrotoxic drugs (eg. non-steroidal anti-inflammatory drugs, aminoglycosides). For each dog, data about urea, creatinine and urinary output (UO) were recorded. Acute kidney injury (AKI) grading was made based on current IRIS consensus. Canine Acute Pancreatitis Severity (CAPS) was calculated for each dog at presentation and previously

described cut-off of 11 was used to divide dog into two groups (CAPS < and > 11). SDMA was measured using high performance liquid chromatography (HPLC). The SDMA was compared between UO groups (O, NO), presence of AKI and with CAPS score groups using Mann-Whitney *U*-test or Welch's *t*-test based on normality distribution. SDMA was correlated with urea and creatinine levels using Spearman's correlation test.

Fifty-one dogs with diagnosis of AP were enrolled with owners' informed consent. Sixteen dogs showed AKI and 11 of them were oligo-anuric. Overall median SDMA was 13.8 mg/dL (range 0.6-65 mg/dL). Twenty-three dogs (45%), of which 13 in non-AKI group, had SDMA above reference range (15 mg/dL). Median SDMA was significantly higher in AKI dogs (18.5 vs. 12.4 mg/dL; $P = 0.01$). Both urea and creatinine concentration showed a positive correlation with SDMA level in AKI dogs ($P = 0.01$ $r = 0.6$ and $P = 0.006$ $r = 0.7$, respectively) but the same correlations were not significant in non-AKI group. No association between SDMA and UO was found. Finally, dogs with CAPS>11 had higher SDMA compared to dogs with CAPS<11 (26.9 ± 5 mg/dL vs. 13.8 ± 1.5 mg/dL, $P = 0.03$). Based on our results, SDMA seems to be associated with disease severity (CAPS).

SDMA correlates well with kidney disfunction parameters (urea and creatinine) and results higher in AKI dogs. Interestingly, about 1/3 of non-AKI dogs presents abnormal SDMA that can be related to a subclinical kidney impairment.

Disclosures

No disclosures to report.

ESVNU-P-8

Evaluation of a point-of-care lateral flow immunoassay for detection of significant bacteriuria in dogs and cats

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There is a need for affordable point-of-care (POC) tests to accurately detect significant bacteriuria in dogs and cats.

Such tests may help practitioners in limiting unnecessary empirical antimicrobial treatment while improving patient outcome.

The aim of the study was to compare a lateral flow immunoassay POC (RapidBac™Vet) for detection of bacteriuria, to standard aerobic quantitative bacterial culture (QBC) at a reference laboratory.

The study was designed as a prospective cohort study. Urine samples were collected from dogs and cats presenting to the University Hospital for Companion Animals. Samples were subjected in parallel to RapidBac-Vet and QBC. POC results were interpreted by six investigators blinded to the gold standard results for coefficient of variation calculations.

Surplus urine samples from 79 dogs and 21 cats were included (58% by cystocentesis). Forty-four samples yielded ≥1000 colony forming units (CFU)/ml on QBC, of which 20 yielded heavy growth (≥100 000 CFU/mL). POC sensitivity and specificity and positive and negative predictive values were 71%, 77%, 71% and 77%, respectively. Eight of the 13 false negative samples grew *Staphylococcus* spp. By applying a cut-off of ≥100 000 CFU/mL, sensitivity, specificity and positive and negative predictive values changed to 80%, 65%, 36% and 93%, respectively. The intra-assay and inter-rater coefficients of variation were 1.4-3.23% and 83%, respectively.

These results suggest that RapidBac™Vet is of limited value as a sole discriminatory test for urinary tract infection. A negative test result does not rule out urinary tract infection but may justify withholding of antimicrobial therapy pending the urine culture result.

Disclosures

No disclosures to report.

ESVNU-P-9

Antioxidant enzyme activity in dogs with acute uraemia managed with haemodialysis

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Impairment in antioxidant enzyme activity is involved in several complications in human patients managed with intermittent haemodialysis (HD). Different factors, concerning uraemia and HD treatment, can promote oxidative stress in these patients. HD may generate oxidative stress due to several factors, such as reduced dialyzer biocompatibility, extensive contact between blood and synthetic surfaces of extracorporeal circuit, and poor dialysate sterility. The aim of the present study was to evaluate antioxidant enzyme activity between pre- and post- haemodialysis treatment in dogs with acute uraemia, and its correlation with systemic inflammatory response syndrome (SIRS), and disseminated intravascular coagulation (DIC). Ten uremic dogs managed with HD (HDG) and ten clinically healthy dogs (CG) were included. Enzymatic activities of catalase (CAT), superoxide dismutase (SOD), and glutathione peroxidase (GPx) were assessed in blood hemolysates of dogs of both groups. In HDG dogs, blood samples were taken immediately before HD, and at the end of HD (15 seconds into bypass at standard blood flow of 50 mL/min). HDG dogs were classified according to IRIS guidelines for AKI. The presence of SIRS and DIC were diagnosed on the basis of emathological, biochemical and coagulation profile according to the recently published criteria. Enzymatic activities of CAT, SOD and GPx were compared among CG, pre HD treatment (n = 17) and post HD treatment (n = 17), using the Tukey's Multiple Comparison Test. Enzymatic activities of CAT, SOD and GPx showed no significant differences between pre-dialysis and post-dialysis values, although GPx was significantly increased ($P < 0.0001$) in HDG compared to CG. Dogs with DIC (n = 4) showed significantly lower CAT ($P < 0.0001$) and GPx ($P < 0.0001$) levels, than dogs without DIC (n = 6). CAT and GPx activities in patients with SIRS (n = 3) appeared to be significantly lower than patients without SIRS (n = 7). In our cohort of dogs, intermittent HD did not seem to significantly affect antioxidant enzyme activity. However the reduced enzymatic activities in uremic dogs with DIC and SIRS is worthy of further investigations.

Disclosures

No disclosures to report.

ESVNU-P-10

Risk factors for urinary tract infection in dogs with natural occurring leptospirosis: a retrospective cohort study of 76 dogs

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Leptospirosis is a zoonotic disease. Due to the risk of dog-to human transmission of leptospirosis and the need of monitoring urine output in dogs with acute kidney injury it is highly recommended to place an indwelling urinary catheter during hospitalization. Urinary catheterization can predispose to urinary tract infection (UTI). The aim of this retrospective cohort study was to evaluate type and frequency of UTI in dogs with leptospirosis and to identify risk factors associated with UTI in leptospirotic dogs.The electronic data-base of the San Marco Veterinary Clinic P.O.A System-Plus 9.0 was searched between January-2008 and December-2018 for dogs with diagnosis of leptospirosis (n = 78). Diagnosis of leptospirosis was consistent clinicopathologic signs, a positive microscopic agglutination test (titer ≥1:1600 in vaccinated dogs, titer ≥1:800 in non-vaccinated dogs or ≥ 4-fold increase in convalescent titer) and/or a positive PCR (urine and/or blood). Dogs with leptospirosis to be included in the study need to met the following criteria: negative urine sediment and urine culture at arrival, ≥ 3 days hospitalization, and a urine sample for cytologic examination and bacterial culture during hospitalization. Dogs meeting inclusion criteria were divided in 2 groups: catheterized dogs (group 1) and non-catheterized dogs (group 2). Differences in UTI frequency between groups was evaluated (Fisher's exact test). Association between duration of catheterization and risk of developing UTI was analysed (Wilcoxon-Mann-Whitney test). Age, sex, diarrhoea, antecedent antibacterial and/or immunosuppressive treatment were evaluated for association with UTI (Chi-square test). Significance level for all statistical test was set at $\alpha < 0.05$. A total of 76 dogs met inclusion criteria: 25 in group 1 and 51 in group 2. Overall 5/76 (7%, all in group 1) dogs developed a UTI. Identified bacteria were: *Escherichia coli* (3 dogs), *Pseudomonas aeruginosa* (1 dog) and *Klebsiella pneumoniae* (1 dog). All UTI were defined as multidrug-resistant. Frequency of UTI was significantly higher in group 1 (5/25, 20%) compared to group 2 (0/51, 0%; OR = ∞; CI = 2.082 — ∞). Infected dogs had a significantly longer catheterization time (median = 6 days, IQR = 2.3) compared to non-infected dogs (median = 2 days, IQR = 2.6; $P = 0.026$). Age, sex, diarrhoea, antecedent antibacterial and/or immunosuppressive treatment were not significantly associated with UTI. Urinary catheterization and duration of urinary catheterization represented important risk factors for development of UTI in dogs with Leptospirosis.

Disclosures

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ESVNU-P-11

Accuracy of refractometric urine specific gravity determination in cats

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Urine-specific gravity (USG) is used as a proxy for urine concentration or osmolality and usually determined by using a refractometer. There is a long held belief that feline urine has higher specific refractivity than human or canine urine, thus human refractometer scales may result in falsely high readings for feline samples. There are some commercially available veterinary refractometers that are calibrated with different scales for cats and a conversion formula has also been recommended if a refractometer with human scale is used in order to avoid overestimation of the USG in cats. Two recent studies questioned whether a separate scale is necessary for the correct interpretation of the refractometer readings in feline urine.

The aim of this study was to compare canine and feline USG readings obtained by a commercial human refractometer with weight/volume measurement USG data. A second aim of the study was to test whether the centrifugation of the urine samples may have an effect on the accuracy of refractometric USG determination.

The USG of thirteen canine and fifteen feline urine left over specimens were compared using refractometry and precise weight/volume measurement. All urine samples were clear, pale to dark yellow in color. The bias between the two methods was determined by Bland-Altman analysis. The measurements were performed both on uncentrifuged and centrifuged samples. The effect of centrifugation was evaluated by paired T test.

The median (1st, 3rd quartiles) USG of the canine samples was 1015 (1008, 1024). The mean (±SD) USG of the feline samples was 1033 (±11). There were no significant differences between the refractometric USG values of canine or feline urine samples before and after centrifugation. The mean bias between the refractometric and weight/volume determined USG values of the feline samples was 1,8 ± 2,4. Using the generally accepted feline correction formula changed the mean bias to −3,1 ± 3,6. For comparison, the mean bias of the canine samples was −2,9 ± 5,7.

Based on our results we can conclude that it is unnecessary to use different scales or correction formulas during the refractometric USG determination of canine or feline urine samples. The use of the feline correction formula may result in a clinically important underestimation of the USG. The USG measurements can be performed both on native and centrifuged urine samples if the urine is clear. The inaccuracies of the refractometers irrespective of the species should be taken into consideration during the clinical decision making.

Disclosures

No disclosures to report.

ESVNU-P-12

Early detection of tubular damage in dogs infected with Leishmania infantum: use of N-acetyl-β-D-glucosaminidase (NAG) and glutamyl transferase (GGT)

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Renal damage associated with canine leishmaniasis induces nephritis and glomerulonephritis which cannot always be detected using classical laboratorial biomarkers (creatinine, urea, protein/creatinine ratio and urine specific gravity). N-acetyl-β-D-glucosaminidase (NAG) and glutamyl https://www.sciencedirect.com/topics/agricultural-and-biological-sciences/transferases (GGT) are hydrolytic enzymes found in the epithelial cells of the proximal tubule of the kidney being their presence in the urine associated to tubular damage. Hence, we aimed to evaluate the diagnostic power of urinary NAG and GGT as early markers of renal disease in dogs affected with leishmaniasis using a modified IRIS chronic kidney disease staging.

A prospective study was conducted in 5 groups of dogs:17 healthy dogs (C),13 dogs (G1) with an urinary protein/creatinine ratio (UP/C) ≤0.2 and plasma creatinine (CR) <1.4 mg/dL, 5 dogs (G2) with UP/C between 0.21-0.4 and CR < 1.4 mg/dL, 6 dogs (G3) with UP/C ≥ 0.41 and CR <1.4 mg/dL and 15 dogs (G4) with UP/C ≥ 0.41 and CR≥1.4 mg/dL.

Dogs presented to the University of Extremadura small animal hospital with a variety of clinical signs and diagnosed with visceral Leishmaniasis were included in the study. All dogs had haematology, biochemistry, blood protein electrophoresis, Leishmania ELISA (quantititative), abdominal ultrasonography, and full urine analysis and culture (including UP/C, NAG and GGT) carried out.

NAG was determined using a commercial kit (Diazyme®, Germany), GGT by a specific kit from RAL® (Spain). The results are all normalized to their respective urinary creatinine and expressed as ratios: uNAG/CR and uGGT/CR. All groups were not normally distributed and a Mann-Whitney-*U* test was used to compare among groups; $P < 0.05$ was considered statistically significant.

The urinary uNAG/CR (IU/g; mean ± SD) was 1.6 ± 0.7 in group C;5.77 ± 5.04 for G1;10.27 ± 6.96 in G2;12.61 ± 13.1 in G3 and 57.51 ± 54.36 in G4. For uGGT/CR (IU/g; mean ± SD) the values obtained were 0.8 ± 0.69 for group C;1.43 ± 1.58 in G1;5.07 ± 10 in G2;3.34 ± 5.72 in G3 and 17.75 ± 31.22.1 in G4.

uNAG/CR differed statistically in all groups compared to control and G1, G2 and G3 differed from G4; significant differences for uGGT/CR were only found between C and G4.

This study shows that uNAGC/CR appears to be a good biomarker for early detection of renal tubular damage at early stages of canine leishmaniasis, prior to the presence of azotaemia and proteinuria. On the contrary, uGGT/CR appears to be less useful for this purpose, as it is only consistently elevated at more advanced stages of leishmania-associated chronic kidney disease.

Disclosures

No disclosures to report.

ESVNU-P-13

Usefulness of urine neutrophil gelatinase-associated lipocalin (NGAL) and Cystatin C (CysC) in the diagnosis of renal disease in dogs affected with leishmaniasis

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Canine leishmaniasis is a highly prevalent zoonotic disease in Spain. All dogs affected present structural and/or functional abnormalities in their kidneys ranging the clinical presentation from asymptomatic to severely diseased individuals being the clinical signs, creatine and proteinuria used to assess the evolution of their condition. Evaluation of other markers of early renal damage could be used for redefining disease classification or for prognostic and/or therapeutic guidance purposes. Moreover, dogs with Leishmaniasis are optimal natural models for the study of tubular damage biomarkers such as NGAL and CysC in canine chronic kidney disease. We aimed to evaluate the diagnostic power of urinary NGAL and CysC as early markers of renal disease in dogs diagnosed with *L. infantum* using a modified IRIS chronic kidney disease staging.

Dogs presented to the University of Extremadura veterinary hospital with a variety of clinical signs and diagnosed with visceral Leishmaniasis were included in the study and 5 groups were stablished: 10 healthy dogs (C),13 dogs with an urinary protein/creatinine ratio or UP/C ≤ 0.2 and plasma creatinine or CR < 1.4 mg/dL (G1),7 dogs presenting UP/C 0.21-0.4 and CR < 1.4 mg/dL (G2),16 dogs with UP/C ≥ 0.41 and CR < 1.4 mg/dL (G3) and 16 dogs with UP/C ≥ 0.41 and CR≥1.4 mg/dL (G4).Some dogs also had other testing including thoracic radiographs, echocardiography or Leishmania PCR in a variety of tissues.

NGAL was determined using the canine NGAL Elisa kit (Abcam, USA) and the results are expressed as NGAL/creatinine ratio (uNGAL/CR). Cystatin C was measured using a turbidimetric latex assay (Spinreact, Spain) and is expressed as CysC/creatinine ratio (uCysC/CR). Mann-Whitney-*U* test was used to run all comparisons due to the non-gaussian distribution of the data with $P < 0.05$ considered as statistically significant.

The urinary uNGAL/CR (μg/g; mean ± SD) was 1082 ± 782 in group C;5210 ± 3676 for G1;38 446 ± 12 269 in G2;453 219 ± 322 898 for G3 and 4 306 983 ± 3 852 666 in G4. For uCysC/CR (μg/g; mean ± SD) the values obtained were 80 ± 54 in C; 485 ± 249 in G1; 475 ± 81 in G2; 9294 ± 7992 in G3 and 36 450 ± 22 571 in G4. For both biomarkers statistically significant differences were found between the control group and all the rest. G1 and G2 were significantly different compared to G3 and G4 for NGAL and CysC. Additionally, CysC values differed significantly between G3 and G4.

Our results demonstrate that urinary uCysC/CR and uNGAL/CR are highly sensitive biomarkers that detect tubular damage in non-azoemic, non-proteinuric dogs affected with canine leishmaniasis.

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ESVNU-P-14

Complicated UTI in dogs: uropathogens, antimicrobial resistance and comorbidity

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Complicated urinary tract infections (cUTI) occur in the setting of a urinary tract with underlying metabolic, functional, or structural abnormalities that typically require longer antibiotic courses and carry a higher risk of treatment failure.

UTI are major reasons for antibiotic prescription in dogs and the responsible bacterial populations have developed increasing resistances.

The aim of this retrospective study was to investigate the prevalence of pathogens, their susceptibility patterns, the comorbidities and the urinary sites involved (detected by ultrasonography) in a population of dogs affected with cUTI.

Four hundred thirty one urine samples collected by cystocentesis from 260 dogs underwent to urine culture for diagnostic purposes. Antimicrobial sensitivity tests were obtained by Kirby-Bauer method. Comorbidities were deduced by the analysis of patient clinical and ultrasound reports.

A control group of 360 dogs (4fold the dogs affected) was randomized among the whole canine population examined during the same period of time (2013-2017). Wilcoxon, Kruskal-Wallis and Chi-square tests were used for statistical analysis.

One hundred forty one urine samples (32.7%) from 90 dogs (34.6%) had a positive culture.

Crossbreeds (29%) and spayed females (42%) were prevalent and the mean age was 9.2 years.

A significant higher prevalence, among the "UTI-dogs" was observed for Labrador Retriever, English Bulldog, Golden Retriever, Beagle and Cocker Spaniel, spayed females and dogs between 8 and 13 years old.

Escherichia coli was the predominant pathogen (43%), followed by *Staphylococcus pseudintermedius* (8%), *Staphylococcus aureus* (8%), *Streptococcus faecalis* (7%), *Klebsiella pneumoniae* (6%), *Pseudomonas aeruginosa* (5%) and other 13 species. A predominance of single isolates (89.4%) compared to polymicrobial infections (10.6%) was observed.

Marbofloxacin was overall the most effective molecule (63.1% sensitivity), followed by Cefovecin (58.6%), Ceftriaxone (55.1%), Enrofloxacin (54%) and Pradofloxacin (53.3%). *Escherichia coli* showed the highest sensibility versus Cefovecin (70%), Marbofloxacin (67.2%), Trimetoprim/Sulphamide (64.3%), Ceftriaxone (63.5%), Pradofloxacin (61.5%) and Enrofloxacin (60.9%).

The most represented identified comorbidities were urolithiasis (25%), CKD (24%), hyperadrenocorticism (11%) and extra-urinary neoplasms (10%). Eight dogs were included because of the recurrence of the infections.

Of the 68 dogs underwent to abdominal ultrasound, 58 (85.3%) showed ultrasonographic abnormalities involving the urinary system: 36 (52.9%) in the upper tract, 41 (60.3%) in the lower tract and 19 of these in both (27.9%).

The high rate of antimicrobial resistance detected could lead to treatment failures and poor prognosis; additional guidelines are needed because of the public health concern determined by the zoonotic potential of the isolated bacteria.

Disclosures

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ESVNU-P-15

Cats at risk or with spontaneous CKD. What affects survival and prognosis?

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Chronic kidney disease (CKD) is among the major causes of morbidity and mortality in cats, with a significant prevalence up to 31% over 15 years old.

The aim of this retrospective observational study was to evaluate the prevalence of death, survival time and risk factors in a population of cats at risk or affected with CKD.

One hundred thirty three cats, from a starting population of 472 (years 2013-2018), were included in this study. One or more of the following criteria had to be observed during their first clinical examination: age over 9 years, serum creatinine (SCr) >1.6 mg/dL, borderline (0,2-0,4) or pathologic (>0,4) urinary protein/creatinine ratio (UPC), urine specific gravity (USG) <1.035. Furthermore, their "follow-up data" have been obtained through an online questionnaire filled by the owners (beginning of 2019). The nephropathic cats were staged according to IRIS guidelines, and not nephropathic cats were included in stage 0.

Wilcoxon test and Kaplan Meyer survival curve analysis were performed.

Median age of the population was 11 $\pm$ 4,31 years; male were overrepresented (55 vs 45%); Domestic Shorthair was the predominant breed (76%).

Forty nine (36,8%) cats were included in stage 0; 21 (15,8%) in stage 1; 48 (36,1%) in stage 2; 7 (5,3%) in stage 3; 8 (6%) in stage 4, with a mean sCr value of 1,98 mg/dL $\pm$ 1,54.

Sixty (45,1%) cats were naturally dead or euthanized at the time of the survey; 25 (18,8%) of these due to CKD. Some parameters were significantly different between the two groups: "dead by CKD" and "dead by other diseases". Serum creatinine was higher, while USG, red blood cells (RBC), white blood cells (WBC) and hematocrit (Ht) were significantly lower in "dead by CKD" cats. Survival time in nephropathic cats was related with age, IRIS staging, serum phosphorus, RBC, WBC, Ht. Cats staged as IRIS-2 survived longer than cats staged as IRIS-1, because other comorbidities are the reason for the consultation in stage-1 cats. A lower survival time was observed in cats with a body condition score different than normal (higher or lower). Lower survival was observed in hypertensive conditions when the whole population of cats was considered, and not only the CKD one.

Other than sCr, results from CBC and USG are to keep in consideration in a prognostic evaluation of cats at risk or affected with CKD. Age has to be considered a risk and a prognostic factor.

Disclosures

No disclosures to report.

ESVONC-P-1

Multicentric and prospective study on 271 cases of endonasal neoformations in the dog

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Tumors of the nasal cavity represent 1-2% of all canine neoplasms. They are more frequent in elderly animals, of epithelial origin and locally invasive with a low metastatic incidence.

All the subjects included in this multicenter and prospective study were examined between June 1, 2016 and December 31, 2018; the rhinoscopy revealed new tissue formation in the nasal cavity and in the nasopharynx. The case histories, including clinical exams, laboratory exams, X-rays, endoscopies, tomographies, histology, therapeutic choices, and follow-ups, of 271 subjects were collected and analysed.

The dogs were principally dolichocephalic breeds, 50% males and 48% females, with an average age of 10 and average BCS 2.9 (0-5). Most of the subjects (78%) underwent symptomatic treatment before the diagnosis, and in 43% symptoms had been present for over 3 months. The most common clinical symptom was nasal discharge (87%), unilateral in 55%. Serosanguinous discharge and/or epistaxis were present in 62% of the cases; stertor in 63%; facial swelling, deformation of the nasal planum, and/or exophthalmos in 20%.

Endoscopic examination found neoformation, which was unilateral in 34.7% of the cases and nasal and nasopharyngeal in 55%. The histological types of the neoplasms were classified as malign epithelial (70%), malign mesenchymal (12%), benign (11%), round cell tumors (4%), and other (3%).

Of the 271 subjects evaluated, 23 underwent endoscopic debulking (diode laser associated with grasping forceps), 31 metronomic therapy, 28 debulking and metronomic therapy, 4 radiation treatment, 5 radiation and metronomic radiotherapy, 8 chemotherapy, 2 surgery, 1 debulking and radiotherapy and 169 no specific therapy. Of the 166 deceased patients (61.3%), the average survival time was 160 days for malign epithelial tumors and 206 days for malign mesenchymal tumors. In patients that underwent debulking and metronomic therapy the average survival time was 442 days, in those treated with debulking alone 227 days, metronomic therapy alone 216 days, and in those that did not have therapy 115 days. To date, of the 29 subjects with benign neoplasia, 21 (72.5%) are alive.

In conclusion our study shows that nasal tumors are often diagnosed late, when the nasopharynx is already affected. The most common histological type is adenocarcinoma; benign neoplasia, which is rarely described in the literature, is also frequent. The good response to nasal debulking combined with metronomic therapy makes this multimode approach worthy of further evaluation, both for its reduced cost and for the high quality of life for the patients.

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ESVONC-P-2

Sensitivity of canine and human cancer cell lines towards thermoradiotherapy

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Hyperthermia (41°C to 43°C) combined with radiotherapy (HT-RT), or thermoradiotherapy, is used clinically in particular cases of human and canine cancer. Hyperthermia provokes changes at different levels: in the tumor microenvironment by increasing perfusion and oxygenation and inducing an immune response, in the tumor cells by induction of cell death and inhibition of DNA repair mechanisms. These changes increase the efficacy of radiation treatment towards a better tumor response. However, the molecular mechanisms of this cellular sensitization have not been fully elucidated. The aims of our study were firstly to screen human and canine cancer cell lines for their sensitivity towards hyperthermia-radiotherapy treatment, and investigate the role of heat-shock protein HSP70, and DNA repair proteins in the radiosensitization mechanism.

Survival curves after treatment were determined in a panel of human and canine cancer cell lines using a clonogenic survival assay. We analyzed the effect of HT-RT on cell proliferation and apoptosis. Further, we tested the influence of the HT-RT time gap, different temperatures and order of the treatment, using A549 cells that are sensitized by hyperthermia as positive controls. Levels of HSP70 and the DNA repair protein RAD51 were analyzed in HT-sensitive and -resistant cell lines by Western-Blot. Knockdown of HSP70 was performed in A549 cells using siRNA against HSP70. Additionally, we evaluated mRNA levels of BRCA2 in an *ex vivo* canine tumor model (soft tissue sarcomas, carcinomas, mast cell tumors), and whether they were affected by hyperthermia. Out of eight cell lines tested, only A549 and Abrams cells showed significant decrease in clonogenic cell survival when pre-treated with hyperthermia at 42°C. A549 showed high baseline levels of HSP70, which was further induced upon treatment. All other cell lines had low or non-detectable baseline expression levels, but showed strong induction upon treatment. Levels of RAD51 were not affected. Additionally, HSP70 knockdown did not affect clonogenicity after HT-RT. However, BRCA2 mRNA levels were lowered by heat, notably in *ex vivo* treated canine soft tissue sarcomas.

Our results show that a majority of cell lines are not radiosensitized *in vitro*, indicating that the tumor microenvironment is responsible for the major effect of hyperthermia. Interestingly, we discovered a heat-induced suppression of BRCA2 transcription, potentially inhibiting the homologous recombination repair pathway. This is promising in the light of future combination of hyperthermia with PARP inhibitors.

Disclosures

No disclosures to report.

ESVONC-P-3

Interest of the association of abdominal ultrasound and alanine transaminase (ALT) measurements in the determination of hepatic infiltration in case of nodal diffuse large B-cell lymphoma (DLBLCL)

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In canine nodal DLBCL one important step in diagnosis is the determination of the presence of a hepatic infiltration, which correspond to a stage IV based on the World Health Organization's staging for lymphoma. The assessment of liver involvement is classically made by ultrasonography and is confirmed by cytology, which remains the usual procedure. The efficacy to determine hepatic infiltration based on abnormal images has been evaluated at 77% with respective sensitivity, specificity, positive prognostic value (PPV) and negative prognostic value (NPV) of 73%, 81%, 77% and 76% (Crabtree 2010).

The aim of this work was to evaluate if the association of abdominal ultrasound and ALT measurements can help in the determination of hepatic infiltration.

A complete clinical staging was prospectively performed in 76 dogs (examined in 9 oncology referral centers) with a confirmed diagnosis of a nodal DLBCL. Liver ultrasound, cytology and plasma ALT measurements were performed for each dog. The ultrasonographic patterns characteristic for canine lymphoma in liver included coarse parenchyma, ill-defined hypoechoic areas, hypoechoic nodules, diffuse hypoechogenicity, and diffuse hyperechogenicity with or without hepatomegaly (Nyland 1984, Crabtree 2010). ALT analysis was performed in-house, and results were interpreted against each laboratory-specific reference ranges and classified as normal vs elevated. Ethics approval was granted by Royal Canin's Ethical Committee.

Hepatic infiltration, confirmed by cytology, was found in 70% of cases. To evaluate the interest of the association of liver ultrasound and ALT in the determination of infiltration, dogs were classified as positive if they had ALT outside laboratory range in combination with abnormal ultrasound images. Dogs with all other findings were considered as negative. Sensitivity, specificity, PPV and NPV were 17%, 100%, 100% and 34% respectively.

Our results suggest that the simultaneous finding of elevated ALT and abnormal ultrasonography allows to identify dogs with hepatic infiltration in case of nodal canine DLBCL. To confirm these findings further research is needed.

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Disclosures to report.

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ESVONC-P-5

Pet owner feedback on psychological support service in an Italian veterinary hospital: a survey data

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Pet owner burden has been recently explored in a few studies in veterinary medicine taking humans model. Veterinary studies identify this phenomena in pet owners with companion animals affected by chronic or terminal illness. Furthermore, clients burden may exacerbate occupational stressors of the veterinarians.

This study aimed to understand if pet owners are interested on a psychological support provided directly by the veterinary hospital.

A survey was given in the waiting room of single Italian veterinary hospital to dog and cat pet owners, coming to visit for several medical reasons (3rd January-23th March 2019). The survey included questions on socio-demographic data, on the emotional experiences of the owner during the disease and the care of their pets, and on their opinion of having a psychologist for their support.

A total of 350 surveis were administrated, and 268 were returned (76.58% of adherence to the study). Twelve were discarded because incomplete. The sample was composed of 62,9% female and 37.1% male. The most represented age group was 40/60 years (55.5%), had a dog (76.9%) instead of a cat (12.5%) or both (10.6%). Most of the pets presented to the department of internal medicine (43%), followed by oncology (14.8%) and emergency (10.9%); 95.7% of the sample declared to have anxiety, was concerned or demoralized for the health of their animals, and 69.7% of the subjects reported that would have appreciated to receive help from someone competent. In contrast, 30.3% declared “non-desiring” support. The most reported motivation for their decision was the desire to live their emotions alone (50.6%). Sixty-six % of the sample would use the service itself and/or suggest it to its family members in case of difficult decisions, poor prognosis, or emotional management particularly, those who claimed to normally feel anxiety and concern about the health of their animals (47.7%). They were among those who would most likely use the service (32%). Finally, those who declared themselves probable beneficiaries of the service of psychological support, considered in 57% of the cases the hilliness and mourning of their pet comparable, in terms of distress, to the illness and the loss of a loved one.

Our survey showed that a large percentage of owners express the desire to be followed by a professional figure in the field of psychological support, during the treatment of their pets. To our knowledge, this aspect has never been investigated before in an Italian veterinary hospital.

Disclosures

No disclosures to report.

ESVONC-P-6

Evaluating the myelosupressive effects of a single dose of vincristine in dogs with lymphoma

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Lymphoma is the most common haematopoetic neoplasm in the canine population. Chemotherapy protocols, such as COP, CHOP and LOPP are used to treat lymphoma due to its chemosensitive nature. These protocols routinely involve vincristine. Current literature reports the most common vincristine induced toxicity to be gastrointestinal effects but also suggests a degree of myelosupression especially during combination protocols. Little research has focused on the myelosupressive effects of vincristine alone and after a single dose. Neutropenia leading to treatment delays or dose reductions, have been associated with longer remission times.

The aim of this study is to investigate the myelosupressive effects of vincristine in dogs after a single administration.

The records of dogs with previously untreated, confirmed lymphoma receiving vincristine between July 2015 to March 2019 were analysed. Patients were included if they had a haematology performed prior to receiving vincristine and repeated within 5-14 days, prior to receiving a second dose of chemotherapy. Patients were excluded if they received any other chemotherapeutic medication during this time, oral prednisolone therapy was permitted.

Forty-four dogs treated with 0.5-0.7 mg/M^2 intravenous vincristine were included in the study, all of the dogs had lymphoma. Boxers (4), Labradors (4) and Spaniels (6), were over-represented. The study population had a mean age of 7.8 years, ranging from 2.7-13 years. Mean body weight was 22.9 kgs ranging from 3.4-63kgs.

Four dogs (9%) experienced neutropenia (neutrophil count <2.0 x 10˄9/L) following a single administration of vincristine, two of which were borderline neutropenic at the start. Two dogs developed VCOG grade 4 neutropenia, one dog grade 3 and one grade 2. The study population had a mean neutrophil count of 9.96 x 10˄9 (range 1.25-55.22 x 10˄9/L), prior to vincristine administration and a mean of 8.7 x 10˄9/L (range 0.4-35.2 x 10˄9/L), one week post vincristine. 27/44 (61%) cases had a decreased neutrophil count on the second sample. Neither weight nor age influenced likelihood of developing neutropenia.

Fifteen of the cases were thrombocytopenic (<150 x 10˄9 /L) prior to vincristine, compared to only two post vincristine, with a mean of 213.65 and 372.45 respectively. 37/43 (84%) of the cases showed an increase in platelet count following a single administration of vincristine.

This study shows that a small proportion (9%) of dogs receiving vincristine, initially as a sole agent, but as part of a chemotherapy protocol for lymphoma, developed a significant neutropenia within one week of vincristine administration.

Disclosures

No disclosures to report.

ISCAID-P-2

Circulating immune complexes levels correlate with the progression of canine leishmaniosis

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Dogs are the main domestic reservoir of *L. infantum*. In *Leishmania* infected dogs unable to control the infection, a large and uncontrolled humoral immune response is elicited, which is inefficient against parasites. The high concentration of antibodies and circulating antigens in canine leishmaniosis can result in the formation of Circulating Immune Complexes (CICs). Their deposition in tissues has been associated with tissue damage and especially glomerulonephritis and renal failure. However, little is known about the relationship between the presence of CICs and the progression of the disease. The objective was to evaluate the levels of CICs and their correlation with the severity of the disease in serum samples from healthy and infected animals.

A total of 44 dogs, classified according to the LeishVet criteria (Healthy (n = 13), Infected Asymptomatic (n = 12) and Infected Symptomatic -stage I (n = 9), II (n = 17), III (n = 8) and IV (n = 1)), were included in the study. CICs were isolated from serum samples using a slightly modified PEG-precipitation method, and their levels measured by ELISA. The protein content was estimated by bicinchoninic acid (BCA) protein assay. A Nanoparticle Tracking Analysis (NTA) of CICs was done in order to investigate the relationship between CIC molecular size distribution and the progression of the disease.

Results showed a statistical significant correlation between CICs levels and the stage of the pathology in infected dogs. As expected, healthy and infected asymptomatic animals did not show CICs related with the infection and there was a direct relationship of levels of CICs, total protein concentration and progression of the disease. It was also confirmed the correlation between IFAT titers and CICs levels. In addition, data showed that dogs with more severe clinical signs presented large size protein aggregates whereas higher concentration of smaller size aggregates were observed in non infected and asymptomatic dogs. This fact demonstrates a clear positive correlation between clinical stage and the size of precipitated-CICs.

This is the first study correlating the CICs levels with the progression of the disease. in canine leishmaniosis. The measurement of CICs probably represents a valuable tool to not only diagnose but also to predict disease progression and activation in asymptomatic but seropositive dogs or even follow up the efficacy of treatments. Even more, the measurement of CICs as a biomarker of the progression of the disease could provide interesting information about vaccines or immunotherapy treatments to confirm the control of the disease.

Disclosures

Disclosures to report.
Employee of Laboratorios LETI.

ISCAID-P-3

Clinicopathological findings in canine leishmaniosis and its association with signalment

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Canine leishmaniosis (CanL) is a vector-borne disease caused by *Leishmania infantum*.The type of predominant individual immune response

is crucial in the presentation of the disease and determinates the clinical signs and clinicopathological abnormalities in each dog.

Age, sex and breed seem to be determinant in the type of clinical manifestations that dogs develop as well as the outcome of infection. The objective of the study was to define clinicopathological findings of CanL and its association with signalment.

A total of 123 dogs with a diagnosis of leishmaniosis were retrospectively included. The information obtained through the clinical history, physical examination and laboratorial tests of each dog was used to fill out a database that included signalment, clinical signs, laboratorial abnormalities and clinical stage of disease.

Most dogs studied were classified as moderate clinical stage of canine leishmaniosis (70.2%). Young dogs have less tendency to develop systemic signs (P = 0.0059), renal (P = 0.0015) and hematologic (P = 0.0267) abnormalities, while dermatologic signs appear to be more common in young dogs compared with old ones (P = 0.0451). Young dogs showed proteinuria less often than older dogs (P = 0.0029). Dogs younger than 3 years did not present renal azotemia, while older dogs showed occasionally renal azotemia (P = 0.0284). Younger dogs were mainly classified as Stage I or II-mild-moderate disease, and very rarely as Stage III or IV- severe or very severe disease, compared with dogs older than 3 years old (P = 0.0153). Pure breed dogs seem to have significantly more tendency to develop ulcerative dermatitis compared to mixed breed dogs (P = 0.0460).

This study describes, for the first time, that age appears to be associated with differences in clinicopathological findings of CanL. Young dogs appear to present less severe manifestation of disease and are more prone to develop dermatologic signs than adult-old dogs. Moderate clinical stage is commonly found in CanL.

Disclosures

No disclosures to report.

ISCAID-P-4

Correlation between the molecular epidemiology of canine Babesia species and the distribution of vector ticks on dogs in Taiwan

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Babesia gibsoni and *Babesia vogeli* have both been identified in canine babesiosis in Taiwan, where information on the epidemiology of the disease is limited. Although direct transmission of *B. gibsoni* between fighting dogs has been reported from the USA, Korea and Romania, this mode of transmission does not occur in Taiwan. The objective of our study was to correlate the distribution of *Babesia* with the distribution of ticks infesting dogs in Taiwan.

A total of 389 surplus blood samples and 3037 ticks were collected from 389 roaming and free ranging owned dogs, during neutering procedures, at various residential sites in Taiwan between January 2014 and December 2017. The prevalence of *B. gibsoni* and *B. vogeli* was determined by PCR, whereas all ticks were identified under a stereomicroscope using various morphological keys.

An average of 7.8 ticks was collected from 261 dogs in the north of Taiwan, 83 dogs in the middle and 45 dogs from the south of Taiwan. Five different species of ticks were found: *Rhipicephalus sanguineus* (throughout Taiwan), *Rhipicephalus haemaphysaloides* (only in the north), *Haemaphysalis hystricis* (only in the north and middle of Taiwan), *Amblyomma testidunarium* and *Ixodes ovatus* (both only in the north). The prevalence of *B. gibsoni* and *B. vogeli* infection was 13.4% (56/389) and 10.3% (40/389), respectively. Most positive *B. gibsoni* dogs were found in the northern part of the country 51/56 (91%), whereas a few were found in the middle part 5/56 (9%). *Babesia vogeli* infections were distributed as follows: 29/40 (72.5%) in the north, 3/40 (7.5%) in the middle and 8/40 (20%) in the south of the country. None of the dogs in the south were infected with *B gibsoni*, which correlated with the absence of *H. hystricis*, a tick recently identified as the local vector for *B gibsoni*. *Babesia vogeli* was more equally distributed coinciding with the occurrence of *R.sanguineus*, which tick is present throughout Taiwan. These findings are discussed in relation to the local clinical relevance and treatment of canine babesiosis in Taiwan.

Disclosures

No disclosures to report.

ISCAID-P-5

Risk factors of Babesia gibsoni infection from client-owned dogs

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Babesia gibsoni (*B. gibsoni*) is increasingly recognized as an anemic cause of canine tick-borne disease worldwide. Taiwan is an epidemic area in Asia. The purpose of this study was to investigate the risk factors associated with *B. gibsoni* infection. A total of 112 dogs with compatible clinical signs suggestive of *B. gibsoni* infection e.g. pale mucous membranes, apathy, anorexia, fever, abnormal urine color and ruling out of large piroplasmas infection (ie. *B. canis* or *B. vogeli*) were collected from National Taiwan University Veterinary Hospital between January 2014 to December 2015.

Polymerase chain reaction (PCR) test for *B.gibsoni* was performed firstly to divide the dogs into positive (59 dogs) and negative (53 dogs) groups. Factors including environment, season, breed, gender, intact or neuter status, living lifestyle, external parasites prevention, urine color, mucous membrane color and history of babesiosis were analyzed by using of chi-square test initially. Variables with P-value ≤0.1 were further analyzed with a stepwise multivariate logistic regression analysis. Differences were considered to be statistically significant when their associated P-values were ≤ 0.05. The odds ratio of irregularly external parasites prevention (P = 0.001, OR = 4.623, CI:1.935-11.044) and presenting of dark brown urine (P = 0.005, OR = 3.336, CI:1.432-7.774) were 4.623 and 3.336, respectively.

Therefore, the both factors were most likely to be associated with infection.

The results revealed that regular prevention of external parasites and observation of urine color are very important in babesiosis epidemic areas.

Disclosures

No disclosures to report.

ISCAID-P-6

Acantocheilonema reconditum in hunting dogs from Southern Italy: distribution, risk factors and haemato-biochemical findings

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Acantocheilonema reconditum is a parasite transmitted by fleas, lice and ticks and is included among the filaroid species infecting dogs. Contrary to the more well-studied *Dirofilaria immitis* and *Dirofilaria repens*, *A. reconditum* is believed to be less pathogenic, as adult worms are localized in the subcutaneous tissues and in the perirenal fat. Although previous studies reported the absence of clinical symptoms in infected dogs, there are few data regarding the haematological and biochemical changes that could be potentially caused by this parasite.

Because hunting dogs are frequently exposed to vector-borne pathogens, the aim of the present study was to assess the prevalence, risk factors and potential hematobiochemical abnormalities associated with *A. reconditum* infection in this specific canine population. Blood samples were collected from 3020 hunting dogs living in Campania region, and were tested by a modified Knott technique to count and identify microfilariae. Out of 3020 dogs tested, 84 were positive to *A. reconditum*, with an overall prevalence of 2.78% (95% CI: 2.19% - 3.37%). The number of microfilariae/ml ranged from 1 to 442. After excluding dogs co-infected by different filarial worm species and/or other vector-borne pathogens common in Southern Italy, n. 74 dogs showed *A. reconditum* single infection. The main clinical features observed were dehydration (n. 1), fever (n. 1), congested mucous membranes (n. 2) and exercise intolerance (n. 2). Complete blood cell count results revealed leukocytosis (n. 16), anaemia (n. 2), thrombocytopenia (n. 8), eosinophilia (n. 1). Biochemical data showed increased serum values of total globulins (n. 14), albumins (n. 9), gamma glutamyl transferase (n. 2) and alkaline phosphate (n. 1), hypoalbuminemia was observed in one dog.

Dogs co-infected with other vector-borne pathogens were not included in the statistical analysis. The living area ($P < 0.0001$), type of hunted species ($P = 0.0004$) and ectoparasite infestation ($P = 0.018$) were variables significantly associated to *A. reconditum* infection. Living in Caserta province (OR = 6.0, 95% CI: 2.6-14.2) and in Napoli province (OR = 7.0, 95% CI: 2.4-20.2), hunting of wild mammals (OR = 2.9, 95% CI: 1.6-5.1) and ectoparasite infestation (OR 1.9, 95% CI: 1.1-3.4) represented risk factors. Concerning the haematochemical parameters of infected dogs, a significant negative correlation between microfilaraemic load and serum albumin level was found (Pearson Correlation Coefficient: –0.35; $P = 0.025$).

The obtained data confirm the circulation of *A. reconditum* within the hunting dog population of Southern Italy and provides more information about the pathogenic potential of this filarial worm.

Disclosures

Disclosures to report.

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ISCAID-P-7

Effect of human antiretroviral compound Tenofovir in the treatment of cats naturally infected with feline immunodeficiency virus (FiV)

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Feline immunodeficiency virus (FiV) is one the most common infectious agents of cats. FIV is a lentivirus that shares many properties with human immunodeficiency virus (HIV), can cause an acquired immune deficiency syndrome (AIDS) due to gradual loss in T helper cell numbers and function, characterized by increased susceptibility to secondary pathogens. Both viruses preferentially infect CD4+ T lymphocytes, leading to an inversion of the CD4+/CD8+ lymphocyte ratio. Tenofovir, a Nucleotide Analogue Reverse Transcriptase Inhibitor, is effective against FIV in vitro, and there is some evidence that tenovovir might have greater anti-FIV efficacy with less cytotoxicity than other antiretroviral compounds.

In the present study the therapeutic efficacy of the human antiretroviral compound used in acquired immune deficiency syndrome, was investigated in the treatment of cats naturally infected with feline immunodeficiency virus (FiV). Cats presenting symptoms of recurrent infections (sinusitis, stomatitis) were tested for their FIV and FeLV status by IDEXX SNAP FIV/FeLV Combo test. Cats were included in this study if they tested positive for FiV and presented chronic oral or nasal inflammation. Nine cats met the mentioned inclusion criteria. Tenofovir (Virofob, Alvogen) was administered orally, once daily at a dose of 50 mg/cat for 30 days, alongside specific treatment. For the experimental group, day 0 and day 30 measures included complete blood count, CD4+/CD8+ ratio, BUN and creatinine. One of the nine cats did not tolerate Tenofovir administration and was excluded from the study after five days, due to severe hypersalivation.

Tenofovir had a benefical effect on the severity of oral and nasal inflammation and induced an improvement in the general condition of the FiV infected cats. The cats showed an increased CD4+/CD8+ lymphocyte ratio after treatment without signs of nephrotoxicity or myelotoxicity.

This study suggests that Tenofovir, a human antiretroviral compound, is effective in the treatment of cats naturally infected with feline immunodeficiency virus (FiV) and has no side effects on bone marrow activity and kidney function in the short-term administration.

Disclosures

Disclosures to report.

ISCAID-P-8

The many faces of Lyme borreliosis in dogs: a review of 29 suspected clinical cases

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Canine Lyme borreliosis (LB) is a disease common and well-documented in the northeastern and midwestern parts of the USA, while in Europe the clinical presentation and the existence of the disease are controversially discussed. Fever and arthritis are clinical manifestations most often associated with LB; other presentations such as renal, cardiac, neurological, and muscular disorders are suspected to be sequelae of *Borrelia burgdorferi* (*Bb*) infection, but were not reproduced experimentally. The aim of this study was to document various presentations of canine LB in North America and Europe. Twenty-nine dogs were included in this retrospective study based on clinical signs consistent with LB and at least positive antibody detection. Nineteen dogs lived in endemic areas in the USA, while 10 dogs resided in Europe. Medical records were available for 28 cases, and a phone contact with the owner and the attending veterinarian was established for the 29th case.

LB was discovered accidentally in three cases, presented to veterinarians for wellness exams and vaccinations. Clinical signs were apparent to the examining veterinarians but not to the owners. Orthopedic disorders were reported for 20/29 cases (69%). Four dogs (14%) showed only general signs of disease. Kidney failure occurred in four cases (14%); one dog (3%) died of a dilated cardiomyopathy and one suspicious dog presented barking troubles that responded well to doxycycline. Specific antibody levels against *Bb* did not correlate with clinical signs and severity of the disease but were useful for the follow-up checks. Furthermore, concurrent specific antibodies against *Bb* and *Anaplasma phagocytophilum* were detected in 5/29 cases (17%). Borrelial DNA was found in synovial fluid of four dogs with orthopedic disorders and from one heart sample. Recovery after antibiotic treatment was observed in all dogs with orthopedic or general disorders. Dogs with renal and cardiac manifestations showed a poor prognosis as the dog with cardiac presentation and 3/4 cases with kidney failure died. Application of parasiticides was not always recorded; 6/29 dogs (21%) did not receive regular treatment, 7/29 were treated regularly with isoxazoline systemic products (24%), 2/29 with fipronil-based spot-ons (7%) and 2/29 (7%) with collars (one flumethrin-based and one unrecorded brand).

This study suggests that canine LB may present itself in various clinical forms and should be considered not only in cases of musculoskeletal problems or impaired general condition, but also in cases of renal and cardiac disorders. Further research is required to investigate this disease in dogs.

Disclosures

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Marina Gatellet and Marie Varloud are employees of Ceva Santé Animale. A consent form was signed by the veterinary clinics or the owners prior to enrolment.

ISCAID-P-9

Distribution and risk factors of canine hemotropic mycoplasmas in hunting dogs from Southern Italy

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Mycoplasma haemocanis (Mhc) and *Candidatus Mycoplasma haematoparvum (CMhp)* are two species of canine hemoplasma that may cause hemolytic anemia and chronic disease in canine species. While understanding is limited, blood transfusions, bloodsucking arthropods, biting and fighting are suspected routes of hemoplasma transmission in dogs. The aim of the present survey was to determine the prevalence of hemotropic mycoplasma infections in hunting dogs from Southern Italy and assess related risk factors. Blood samples were collected from 1433 hunting dogs in the Napoli, Avellino and Salerno provinces of Campania region of Southern Italy, and tested by real time polymerase chain reaction (RT-PCR) assays for amplification of *Mhc* and *CMhp* DNA. The dogs had no clinical signs at the time of sampling. The overall PCR positive rates were13.1% for *Mhc* and 11.4% for *CMhp*. Coinfection with both hemoplasma species was found in 4% of animals. Statistical analysis revealed living in Salerno province (*Mhc*: OR = 2.94, 95% CI: 2.10-4.11; *CMhp*: OR = 2.27, 95% CI: 1.61-3.20), hound breeds (*Mhc*: OR = 4.0, 95% CI: 2.86-5.59; *CMhp*: OR = 1.61, 95% CI: 1.16-2.24), pack size more than 10 animals (*Mhc*: OR = 1.67, 95% CI: 1.14-2.47; *CMhp*: OR = 1.61, 95% CI: 1.06-2.44) and wild mammals hunting (*Mhc*: OR = 3.53, 95% CI: 2.51-4.98; *CMhp*: OR = 15.7, 95% CI: 8.83-28.0) as associated risk factors for both canine hemoplasma infection. Adult age was a variable significantly associated only to infection by *CMhp* (OR = 1.93, 95% CI: 1.25-2.97). To the authors' knowledge, this is the first large-scale molecular survey on *Mhc* and *CMhp* infections in dogs living in Southern Italy. The obtained data confirm the circulation of these two species of canine hemoplasma within the hunting dog population of Southern Italy, although their pathogenic potential and the possible epidemiological relationships between hunting dogs and sympatric wild animal populations are still unclear.

Disclosures

Disclosures to report.
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ISCAID-P-10

Prevalence of vector-borne diseases in free-roaming cats

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Though the role of cats as a reservoir for *B. henselae* has long been established, the prevalence of infection or exposure of cats to other *Bartonella* species or to other vector-borne diseases remains unreported. Using serology and polymerase chain reaction (PCR), the prevalence of exposure and bacteremia of vector-borne infections (*Anaplasma*spp, *Babesia*spp, *Cytauxzoon*spp, *Ehrlichia*spp, *Rickettsia*spp, *Piroplasma*,*Mycoplasma*spp, and *Bartonella*spp.) was assessed in a population of free-roaming cats in a rural area of the Midwestern United States. Serum and EDTA blood samples were collected from 65 free-roaming cats captured as part of a community spay and neuter program. Testing with PCR revealed that 9 cats (14%) were positive for *Bartonella* species (6 for *B. henselae*and 3 for *B. clarridgeiae*) and 7 cats (11%) were positive for *Mycoplasma* species (6 for *M. haemominutim* and 1 for *M. haemofelis*). Serological testing revealed that 50 cats (83%) were positive for antibodies against *Bartonella* species. More specifically, 50 cats were positive for *B. koehlerae*, 47 were positive for *B. henselae* and 42 were positive for *B. vinsonii* subspecies *berkhoffi*. A significant positive correlation was identified between positive *Bartonella* spp. PCR results and the presence of antibodies against *B. vinsonii berkhoffi* and *B. henselae*. All cats that were positive for *Bartonella* by PCR had positive serologies for all three species of *Bartonella* tested. Odds ratio analysis of age, sex and weight revealed that male cats were at increased risk for exposure to *B. vinsonii berkhoffi*.

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I (Jean-Sebastien Palerme) am a consultant for Infiniti Medical, LLC.

ISCAID-P-11

Retrospective analysis of cases tested for leptospirosis at a university teaching hospital

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Pathogenic *Leptospira* species pose a risk to canine and human health worldwide. There is very little published data on positive cases of leptospirosis seen in veterinary practice in the United Kingdom. This study aims to analyze all cases tested for leptospirosis in a UK university teaching hospital between 2011 and 2018.

Data was retrospectively collected from a university database; all dogs that were screened for leptospirosis via urine PCR, blood PCR, or serum MAT were included (n = 153). Cases vaccinated within 12 months with an MAT titre<1:800, in addition to testing negative for both urine and blood PCR, were excluded from the positive category (n = 17). Of the remaining 136 cases, 39 tested positive for leptospirosis.

The number of positive cases among those tested was 11/30 (36.6%) in 2016, 18/64 (28.1%) in 2017 and 10/56 (17.8%) in 2018; the 3 cases tested between 2011 and 2016 were all negative. Of the positive cases, 61.5% (24/39) had been vaccinated with a leptospiral vaccine within 12 months and 53.8% (21/39) had received antibiotic therapy prior to referral.

The most common presenting clinicopathological abnormalities of positive cases were increased hepatic (13/39) or renal (12/39) values, with four of these cases presenting with both. None of the positive cases presented with pulmonary haemorrhage or dyspnea.

Primary leptospirosis was the final diagnosis for 27 of the 39 positive cases, while six cases had an unrelated final diagnosis (neoplasia (n = 3); biliary mucocele (n = 1); necrotizing fasciitis (n = 1); pericardial effusion (n = 1)). Three of the positive cases had a final diagnosis of chronic renal insufficiency, two of these secondary to congenital dysplasia, and three cases had a final diagnosis of chronic hepatopathy.

Positive cases were predominantly medium to large breed dogs, with Labradors, followed by beagles, cross breeds, and border collies being most commonly observed in the positive category.

In conclusion, prior vaccination or antibiotic use should not preclude testing for leptospirosis. In contrast to cases reported in mainland Europe, clinical pulmonary disease does not appear to be a feature in these cases. Leptospirosis appears to have been detected incidentally in some cases, potentially signifying a population of dogs with subclinical infection.

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ISCAID-P-12

Canine urine culture and antimicrobial susceptibility patterns over an eight-year period: increasing antimicrobial and multidrug resistance

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Urinary tract infections (UTIs) are common in dogs but appropriate use of antimicrobial drugs is necessary to prevent emergence of multidrug resistant (MDR) bacteria. Awareness of the prevalence of urinary tract infections, causative agents and resistance patterns is essential to guide appropriate therapy.

The aims of the present study were to describe the prevalence of bacterial UTIs in dogs, identify the most commonly isolated microorganisms, and analyze the progression of susceptibility patterns over the study period.

The results of canine urine culture and antimicrobial susceptibility tests performed between January 2010 and December 2017 at the Veterinary Teaching Hospital of the Alfonso X El Sabio University were retrieved from the laboratory database. All samples were collected by cystocentesis and cultured within 24 hours of collection. Antimicrobial susceptibility was determined using the Kirby-Bauer disc diffusion method. Multidrug resistance was defined as resistance to at least one antimicrobial agent in more than three different antimicrobial categories.

A total of 3420 urine samples were identified, with positive culture results in 771 (22.5%). There was no increase in the frequency of positive bacterial cultures over the study period (interval relative risk (IRR) 0.98, 95%CI 0.92-1.0, P = 0.565). The relative effect of developing UTI was significantly higher in females than males (RE 1.42, 95%CI 1.26-1.61, P < 0.001). The most commonly isolated microorganisms were *Escherichia coli* (52.9%), *Staphylococcus* spp. (12.0%), *Enterococcus* spp. (5.8%), *Pseudomonas* spp. (5.7%) and *Streptococcus* spp. (5.6%). The overall prevalence of resistance within the Enterobacteriaceae family was 45.6% for cefazolin, 33.8% for pradofloxacin, 32.1% for trimethoprim-sulfamethoxazole, 30.9% for cefuroxime, 29.3% for enrofloxacin, 26.5% for marbofloxacin, 25.8% for amoxycillin clavulanate, 18.4% for cefovecin and 11.5% for fosfomycin. The prevalence of MDR infections ranged from 2.1% in 2010 to 8.6% in 2017 which meant an increased trend of MDR bacteria of 22% (IRR 1.22 CI95% 1.06-1.42 P = 0.005).

The high frequency and increasing trend of antibiotic resistance observed in this study is concerning and has implication for veterinary and public health. These results emphasise the importance of performing urinary culture and antimicrobial susceptibility testing to allow appropriate selection of therapy. Although resistance to fosfomycin was comparably low, this antimicrobial agent is not licensed for veterinary use in Europe, and given its use in human MDR and methicillin-resistant *Staphilococcus aureus* infections, the use of this antibiotic must be reserved for human medicine.

Disclosures

No disclosures to report.

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The use of MRI and gadoxetic acid to differentiate hepatic parenchymal hyperplastic lesions in dogs

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Magnetic resonance imaging (MRI) using gadoxetic acid (Gd-EOB-DTPA) is widely used in human medicine to characterize hepatic nodular lesions. In veterinary medicine there are few reports of Gd-EOB-DTPA use in liver examinations in dogs.

The aim of the study was to describe a contrast enhancement pattern for different types of liver lesions after administration of Gd-EOB-DTPA in dogs.

The study was carried out on six dogs with a presumptive diagnosis of a focal liver lesion. A clinical examination, laboratory blood tests and abdominal ultrasound were carried out prior to MRI. The animals were examined using a 1.5-Ingenia Philips MRI system. The imaging protocol consisted of breath triggered pre-contrast T1, T2 and post-contrast T1 weighted sequences performed in transverse plane. Gd-EOB-DTPA was administered intravenously at 0.1 mL/kg, followed by 15 mL of a 0.9% saline solution. The post-contrast T1-W sequences were acquired 26 minutes after contrast administration. Samples for histopathological examination were collected from all the cases (surgical resection - 1 case, core-needle biopsy - 4 cases, necropsy - 1 case).

Parenchymal liver metastasis was found in one case. The lesion was strongly hypointense compared to the surrounding liver tissue in both pre-contrast sequences and no signal-enhancement following contrast administration was observed. Focal nodular hyperplasia was observed in one case. It was isointense compared to the surrounding liver tissue in pre-contrast sequences, and it showed signal-enhancement post contrast, similar to that observed in the surrounding healthy tissue. A non-enhancing central scar was observed within this lesion. A hepatocellular adenoma was diagnosed in two cases. The lesions were T1-W hypointense and T2-W hyperintense on pre-contrast images compared with the surrounding liver tissue. In both cases, contrast enhanced MRI of the lesions were observed. In one case, a hepatic carcinoid was found in the liver parenchyma. The tumour was weakly hypointense in T1-W pre-contrast sequences, while it was weakly hyperintense with a strongly hyperintense centre in T2-W images. No contrast enhancement was noted. A hepatocellular carcinoma was also diagnosed in one case. This lesion was heterogenous in pre-contrast sequences, with moderate T1-W hypointensity and moderate T2-W hyperintensity. It did not show contrast enhancement and remained strongly hypointense compared to the surrounding tissue.

The obtained results indicate that contrast-enhancement patters in dogs with various hepatic neoplastic lesions are similar to those in humans. The enhancement patterns used in human medicine to assess hepatic hyperplastic parenchymal lesions may be of use in veterinary medicine.

Disclosures

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