| 1 | Campbell, FE | Effect of hydration status on echocardiographic measures of the left heart in normal cats |
| 2 | Ohad, DG | Spontaneous feline cardiomyopathy as a model for diastolic heart failure (DHF): Is color M-mode trans-mitral flow propagation velocity sensitive enough? |
| 3 | Schober, KE | Doppler echocardiographic assessment of E:Ea and E:Vp as indicators of left ventricular filling pressure in normal cats and cats with hypertrophic cardiomyopathy |
| 4 | Schober, KE | Doppler echocardiographic assessment of the E:Ea ratio as an indicator of left ventricular filling pressure in normal dogs and dogs with heart disease |
| 5 | Falk, T | Arteriosclerotic changes in myocardium, lung and kidney in dogs with chronic congestive heart failure and myxomatous mitral valve disease |
| 6 | Borgarelli, M | Decreased systolic function and inappropriate hypertrophy in small breed dogs with chronic mitral valve disease |
| 7 | Tidholm, A | Survival in dogs with dilated cardiomyopathy and congestive heart failure treated with digoxin, furosemide and propranolol: a retrospective study of 62 cases |
| 8 | Van Israël, N | Athlete heart or DCM in a springer spaniel family? |
| 9 | Moesgaard, SG | Neurohormonal and circulatory effects of short-term treatment with enalapril and quinapril in dogs with asymptomatic mitral regurgitation |
| 10 | Santilli, RA | Anatomical distribution and electrophysiological properties of accessory pathways in seven dogs with orthodromic atrioventricular reciprocating tachycardia |
| 11 | Jenni, SD | Coronary flow reserve measured by positron emission tomography in healthy cats: Adenosine dose finding study |
| 12 | Schellenberg, S | Analytical validation of various immunoassays for the quantification of cardiovascular peptides in dogs |
| 13 | Hogan, DF | Neuroendocrine effects of digoxin on early mitral regurgitation in dogs |
| 14 | Rishniw, M | Molecular diagnosis of canine filariasis—taking the guesswork out of microfilarial identification |
| 15 | Olsen, LH | Use of electronic stethoscope for diagnosing mild mitral regurgitation in dogs |
| 16 | Hildebrandt, N | Experiences with dual-chamber pacemaker implantation in dogs |

**ESVNU—European Society of Veterinary Nephrology and Urology**

| 17 | Le Garréres, A | Effect of mild overhydration on plasma exogenous creatinine clearance test in healthy cats |
| 18 | Arons, J | Polyuria-polydipsia in dogs: usefulness of GFR assessment through creatinine clearance testing for differential diagnosis |
| 19 | Lund, HS | Bacteriuria in feline lower urinary tract disorders (FLUTD) |
| 20 | Wehner, A | Association between glomerular filtration rate, proteinuria and hypertension in dogs with naturally occurring renal and non-renal diseases |

**ESVE—European Society of Veterinary Endocrinology**

| 22 | Boretti, FS | Thyroid enlargement and its relationship to serum t4 status in clinically suspected hyperthyroid cats |
| 23 | Watson, SG | Somatic mutations of the thyroid stimulating hormone receptor gene in feline hyperthyroidism |
| 24 | Kenefick, S | Autonomic neuropathy in dogs with naturally occurring diabetes mellitus |
| 25 | Furrer, D | Effect of amylin on plasma concentration of glucose, insulin and glucagon in an arginine stimulation and meal response test in cats |
| 26 | Fracassi, F | Acromegaly due to a somatroph adenoma in a dog |
| 27 | Sieber-Ruckstuhl, N | Effect of trilostane on cortisol and cortisone concentrations in dogs with pituitary-dependent hyperadrenocorticism |
| 28 | Diaz Espineira, M | TRH-induced GH secretion in dogs with primary hypothyroidism (PH) |
VBPS—Veterinary Blood Pressure society
29 Jepson, RE Feline hypertension: the associations between long-term blood pressure control and survival
30 Espada, Y Effect of sedation on resistive and pulsatility index in dogs
31 Schellenberg, S Effect of adaptation on indirect blood pressure measurement in conscious untrained beagle dogs

ESVCN—European Society of Comparative Veterinary Nutrition/ESVONC European Society of Veterinary Oncology
32 Lhoest, E Is there a difference between energy balance of cats hospitalised for surgical or for medical reasons?
33 Morgan, D The effect of dietary fish oil on puppy trainability

ESVONC—European Society of Veterinary Oncology
34 Simon, D Malignant lymphoma in the dog: Results of treatment with a 12-week maintenance-free chemotherapy protocol
35 Vajdovich, P Changes of free radical and antioxidant parameters in blood of dogs with lymphoma during the course of treatment
36 Setoguchi, A Clinical features of low-grade lymphoma in 8 dogs
37 Brearley, MJ Coarse fractionated radiation therapy for pituitary tumours in cats: a retrospective study of 8 cases
38 Balogh, L How to improve the target/non-target ratio—locoregional application of radiopharmaceuticals
39 Meyer, B HMGA2 expression in canine prostatic tissues—A potential diagnostic tool?
40 Dank, G Preliminary results of the use of carprofen in canine mammary tumors
41 von Euler, H Development of quantitative real-time RT-PCR assays for detection of metastatic disease in canine melanoma
42 Stell, A Identification of a short interspersed nuclear element (SINE) insertion in the gp100 gene of a poorly pigmented canine oral melanoma tumour
43 Kessler, M Long term remission of a primary malignant lymphoma of the urinary bladder in a dog—a case report
44 Tabar, L Primary pulmonary hemangiosarcoma (HSA) in a German shepherd dog with spontaneous pneumothorax

ESVIM—European Society of Veterinary Internal Medicine
47 House, A Pattern recognition receptor mRNA expression and response to stimulation in a canine macrophage cell line
48 Barr, SC Serologic responses of dogs after vaccination with a commercial leptospirosis vaccine
49 Jensen, J Prevalence of Anaplasma phagocytophilum in dogs in Germany
50 Willi, B Prevalence and clinical importance of the two known and a third novel feline haemoplasma species in cats in Switzerland
51 Ruiz de Gopegui, R Review of clinicopathological findings and coagulation disorders in 45 cases of canine babesiosis
52 Resk, N Human dander as a potential allergen source in atopic dogs—allergen characterization and IgE profiling
53 Bohm, M Central and peripheral Babesia canis rossi parasitaemias and their association with outcome of infection
54 Rieker, T First European report of B. gibsoni (Asian genotype) infection in two American pit bull terriers without staying abroad, detection by PCR and sequencing
55 Billen, F Histochemical and immunohistochemical characterization of canine nasopharyngeal lymphoid tissue
56 Forcada, Y Frequencies of feline blood types in cats at the Royal Veterinary College, London, UK
57 Mischke, R Effectivity and safety of different dosages of low molecular weight heparin in dogs suffering from gastric volvulus/dilatation complex
58 Peeters, D Quantification of mRNA encoding cytokines and chemokines in nasal biopsies from dogs with sino-nasal aspergillosis

ESCG—European Society of Comparative Gastroenterology
59 Roura, X Prevalence and development of microalbuminuria in dogs with leishmaniosis
60 Kellumi, HB Clinical presentation and long-term outcome in 21 cats with third degree atrioventricular block (1997–2004)
61 Schulz, B Prevalence of enteral viruses in 936 dogs with acute haemorrhagic diarrhea
62 Suchodolski, JS Reproducibility of endoscopic collection of duodenal juice and evaluation of sample stability for 16S rDNA analysis of the duodenal microflora in dogs
| 63 | Berghoff, N | Determination of intestinal permeability and mucosal absorptive capacity in Norwegian Lundehunds using a four-sugar blood test |
| 64 | Harmoinen, JA | Dynamics of jejunal microbiota during food deprivation |
| 65 | Rinkinen, ML | Probiotic lab caused persistent changes in the canine jejunal microbiota |
| 66 | Dossin, O | Real time PCR quantification and genetic identification of Helicobacter strains in a group of dogs with digestive disorders |
| 67 | Schreiner, N | No changes in histological scoring, total number of infiltrating cells and number of T cells after treatment in dogs with chronic enteropathies |
| 68 | Schneider, M | Coils-embolization of extrahepatic portosystemic shunt in dogs |
| 69 | Watson, PJ | Prevalence of chronic pancreatitis at post mortem examination in an unselected population of first opinion dogs |
| 70 | Hoffmann, G | Copper-associated chronic hepatitis in Labrador retrievers: 15 clinical patients and their family |

**ESFM—European Society of Feline Medicine**

| 71 | Helps, CR | Detection of feline autosomal-dominant polycystic kidney disease (AD-PKD) by real-time PCR genotyping |
| 72 | Peters, IR | Real-time quantitative PCR assays for the diagnosis of three haemoplasma species in feline blood samples |
| 73 | Dye, C | Quantitative real-time RT-PCR for feline coronavirus RNA in the blood and tissues of cats |
| 74 | Baxter, KJ | Tryptophan metabolism is altered in cats infected with feline immunodeficiency virus |
| 75 | Schoeman, JP | Seroprevalence of FIV and FeLV infection and determination of FIV subtypes in sick domestic cats in South Africa |
EFFECT OF HYDRATION STATUS ON ECHOCARDIOGRAPHIC MEASURES OF THE LEFT HEART IN NORMAL CATS. FE Campbell, MD Kittleson. Veterinary Medical Teaching Hospital, School of Veterinary Medicine, University of California, Davis, USA.

The aim of this randomized crossover study was to determine the effects of hydration status on the echocardiographic measures of the left heart in normal cats (n = 10; median age 3 y, weight 3.8 ± 0.8 kg). This population was identified via physical examination, serum biochemistry, PCV, urinalysis, thoracic radiography and 2-D, color flow Doppler, and Doppler tissue imaging echocardiography. Three protocols were employed, including dehydration and intravenous fluid administration at two fluid rates, with a 6-7 day washout period between protocols. To create dehydration, water was withheld and cats were administered (2–4 mg/kg IV q1–2hr to a total dose of 14 mg/kg) over 7hr. Normal saline was administered at a standard maintenance rate (2.5–3 mL/kg/hr IV for 24 hr) and at a rate typically used during surgery (10 mL/kg/hr IV for 7 hr). Prior to and at the completion of each protocol, cats were sedated, the urinary bladder emptied, and body weight, PCV, and TP determined, followed by an echocardiographic examination that was performed prior to randomization and at the completion of each protocol. From the right parasternal short-axis views, the left ventricular (LV) interventricular septum and LV free wall thickness in diastole (IVSd and LVFWd), LV internal diameter in diastole (LVIDd) and systole, largest left atrial (LA) diameter and aortic root diameter in diastole (Ao) were determined. Left ventricular chamber size (LVCAd) and left atrial area (LAa) were determined by tracing the endocardial borders of the LV and LA in diastole. All measures were taken from 4 to 5 consecutive cardiac cycles and averaged. Data were analyzed using one-way repeated measures ANOVA with Bonferroni post hoc test.

The dehydration protocol resulted in a mean (±SD) 5.6% (+0.3%) reduction in body weight (P < 0.001) and a 24% (+7%) increase in PCV and TP (P < 0.0001). Significant and similar increases occurred in IVSd (4.5 ± 0.4 to 5.8 ± 0.6 mm; P < 0.001) and LVFWd while LVIDd (12.6 ± 1.8 to 9.3 ± 1.7 mm; P < 0.001), LVFWd (11.5 ± 0.7 to 8.9 ± 0.1 cm; P < 0.005), LVFWd (1.4 ± 0.2 to 1.2 ± 0.1; P < 0.005) and LA: Ao ratio (1.4 ± 0.2 to 1.0 ± 0.2 cm²; P < 0.05) decreased. Body weight was not altered by either fluid protocol and at the lower fluid rate only LA:Ad increased (1.8 ± 0.5 cm²; P < 0.05). The higher fluid administration rate increased LVFWd compared to baseline (14.4 to 2.0 mm; P < 0.01) and increased LVAd (to 1.6 ± 0.3 cm³; P < 0.01), LA: Ao ratio (to 1.7 ± 0.1; P < 0.001), and LA:Ad (to 2.3 ± 0.6 cm³; P < 0.001) relative to the lower fluid administration rate and to baseline. In conclusion, hydration status alters the echocardiographic measures of the left heart in normal cats. LV myocardial mass, left atrial diameter, and LA:Ao ratio may be useful as a sensitive and readily attainable parameter of diastolic dysfunction in spontaneously occurring feline cardiomyopathy. Larger scale, controlled, and blinded validation studies are warranted to enable taking full advantage of this spontaneously and physiologically occurring animal model of human DHD.

SPONTANEOUS FELINE CARDIOMYOPATHY AS A MODEL FOR DIASTOLIC HEART FAILURE (DHF): IS COLOR M-MODE TRANS-MITRAL FLOW PROPAGATION VELOCITY SENSITIVE ENOUGH? DGI Ohad. The Koret School of Veterinary Medicine, the Hebrew University of Jerusalem, Rehovot, Israel.

Traditional Doppler measures of trans-mitral or pulmonary venous flow patterns prove technically challenging and/or confounded by loading conditions. Transmitral propagation velocity (Vp) is considered both a sensitive and recently introduced echocardiographic index (E:Ea ratio) combines spectral Doppler early transmitral flow velocity (E) with mitral annulus peak early diastolic tissue Doppler velocity (Ea) or Doppler peak early diastolic flow propagation velocity (VP) have been shown useful in the Doppler-echocardiographic prediction of LVFP in people. In this study we hypothesized that the Doppler derived E:Ea and Vp ratios predict the radiographic diagnosis of CHF.

A total of 131 cats, 52 normal cats, 56 cats with HC and without CHF, and 23 cats with HCM and CHF were studied. Diagnostic procedures included Doppler echocardiography (DE), thoracic radiography, and blood pressure measure-ment. Normal cats were used to establish reference values for E:Ea and Vp. Cats with HCM were compared to normal cats matched for age, body weight, heart rate, blood pressure, and left ventricular shortening fraction (n = 26). In normal cats, E:Ea (mean, 95% confidence interval) measured at the septal or lateral mitral valve annulus was 6.8 (6.0 to 7.4) and 7.3 (6.8 to 8.0), respectively (P = 0.268). The E:Ea ratio was 1.16 (1.01 to 1.30). Age, heart rate, blood pressure, and left ventricular shortening fraction did not affect E:Ea. Body weight had an effect on E:Ea (r = -0.32, P = 0.058), age affected E:Vp (r = 0.44, P = 0.003). The E:Ea ratio was increased (P < 0.05) in cats with non-congestive HCM (8.6, 7.9 to 9.4) and cat with HCM and CHF (17.1, 15.4 to 18.9), as was the E:Ea ratio (1.45, 1.21 to 1.70 vs 2.38, 1.81 to 2.94). Using a cut-off of 12.0 for E:Ea and 2.0 for E:Vp to separate cats with HCM and CHF from cats with HCM and without CHF, sensitivity was 76%, specificity was 90% and 81%, positive predictive value was 76% and 59%, and negative predictive value was 95% and 83%, respectively. Kappa statistics revealed a Kappa of 0.74 (“substantial agreement”) and 0.43 (“moderate agreement”) for E:Ea and E:Vp in comparison with radiography in the prediction of CHF! The E:Ea ratio appears to be a useful Doppler-echocardiographic index of CHF in cats with HCM.
DMD (7.57, 7.04 to 8.10), and non-congestive MVD (7.41, 6.64 to 8.18) or DCM (6.84, 6.21 to 7.48) had similar E/Ea (P > 0.05). However, in dogs with CHF (ESV-I 3.34, 11.36 to 16.83) or DCM (2.04 vs. 0.74 cm² ± 0.18), whereas the l/R ratio was significantly decreased in affected dogs (0.41 ± 0.12 vs. 0.53 ± 0.11).

Data from this study show that although FS in affected dogs is increased, as it should be expected in volume overload caused by CMVD, the increased ESV-I in these dogs suggests that a mild systolic dysfunction was present also with moderate heart failure. In fact, ESV-I is believed to be a reliable indicator of systolic function. Inadequate hypertrophy of left ventricle could be responsible for this finding, as it has been demonstrated in experimental models.

**SURVIVAL IN DOGS WITH DILATED CARDIOMYOPATHY AND CONGESTIVE HEART FAILURE: A PROSPECTIVE STUDY OF 62 CASES.** A Tidholm, Albano Animal Hospital of Stockholm, Danderyd, Sweden.

The objective of the study was to retrospectively evaluate the effect of β-blocker therapy in dogs with DCM. Inclusion criteria were as follows: 1) echo-cardiographic evidence of left ventricular eccentric hypertrophy, left atrial dilatation and fractional shortening <25%; 2) radiographic evidence of left-sided or biventricular cardiac enlargement and pulmonary edema or pleural effusion; 3) medical treatment consisting of digoxin, furosemide and propranolol. Survival analysis was based on the Kaplan-Meier method.

Sixty-two dogs of 25 different large and medium-sized breeds were included in the study. Forty-five dogs (73%) were male and 17 (27%) were female. Age at initial presentation ranged from 10 months to 12.5 years (mean, 7 ± 2.5 years). Body weight ranged from 12.2 to 69.9 kg (mean, 30.2 ± 12.7 kg). Echo-cardiographic measurements, indexed according to Kittleton and Kienle, were as follows: LVEDD ranged from 15.3 to 31.2 (mean, 21.4 ± 3.3), LVESD ranged from 9.7 to 20.5 (mean, 13.7 ± 2.5), LAD ranged from 5.6 to 28.5 (mean, 15.8 ± 3.7) and Ao ranged from 2.5 to 9.11 (mean, 6.5 ± 1.3). LAA/Ao ranged from 1.5 to 3.4 (mean, 2.04 ± 0.35) and FS ranged from 4 to 22% (mean, 12 ± 4). Pulmonary edema was present in 60 dogs, and pleural effusion in 2 dogs. Heart rate ranged from 140 to 270 beats/min (mean, 186 ± 38). Thirty-one dogs (50%) presented with atrial fibrillation, and ventricular premature complexes were found in 9 dogs. All dogs were initially treated with digoxin (mean dose 0.009 mg/kg per day) and furosemide (mean dose 3.6 mg/kg per day). Propranolol (mean dose 2.4 mg/kg per day) was added after signs of CHF had been resolved, and similarly one week after initial presentation. Additional treatment consisted of levotirudoxine (15 dogs), spironolactone (4 dogs) and enalapril (2 dogs).

Survival time ranged from 8 to 1335 days (median, 126 days, mean 336 days). Nine dogs were censored in the analysis, 8 of them because euthanasia was performed for reasons unrelated to cardiac disease, and 1 dog was lost on follow-up. Fifty-two dogs were euthanized, nine dogs died suddenly. Post mortem examination was performed in 33 dogs where the attenuated wavy fiber form of DCM was found in 32 dogs and the fatty-infiltration-degenerative form in 1 dog.

The safety and efficacy of the use of β-blocking agents in the treatment of congestive heart failure (CHF) in dogs have been debated. In comparison with previous studies, the present study showed a prolonged survival time when propranolol is added to conventional treatment with digoxin and furosemide, and that survival time in dogs with DCM and CHF treated with digoxin, furosemide and propranolol is comparable to conventional treatment including ACE-inhibitors.

**ATHLETE HEART OR DCM IN A SPINGRER SPANIEL FAMILY?** N Van Issel1, J Dukes-McEwan1, V Biourge2, JLY N Van Israël3, ILY® N Van Israël4. 1Department of Basic Animal and Veterinary Sciences, The Royal Veterinary and Agricultural University, Frederiksberg, Denmark; 2Department of Pathology, Swedish University of Agricultural Sciences, Uppsala, Sweden.

An athletic heart (AH) indicates the presence of morphological (increased LV diameter and wall thickness) and functional cardiac changes (reduced FS, increased SV, bradycardia) as a result of strenuous repetitive exercise. In humans and horses training also influences the development of atroventricular valvular (AVV) regurgitation, but this finding has not been elucidated in the study. All dogs underwent an echocardiographic examination, which included transthoracic 2-D and M-mode. Right parasternal M-mode recordings were obtained from short-axis views with the dogs positioned in right lateral recumbency. A mean of three consecutive measurements was considered for each variable. M-mode measurements included left ventricular end-diastolic diameter (EDD), end-systolic di-ameter (ESD), fractional shortening (FS), and left ventricular diastolic wall thickness (h). From EDD and ESD using Teicholz formula end-diastolic (EDV-l) and end-systolic ventricle (ESV-l) were calculated. Left ventricular radius (RL) was calculated by dividing EDD by two. Systolic function was investigated using FS and ESD, whereas the l/R ratio was used as a parameter of left ventricular hypertrophy. Data are presented as mean ± SD.

Achondroplasia affected dogs were significantly older (8.7 ± 3.5 vs 11 ± 3.2 years). There were no differences concerning sex and weight between the 2 groups. Affected dogs presented a significant increased FS (45.6% ± 8.04 vs 40.06% ± 8.9) and ESV-I (30.0 ml/m² ± 2.3 vs 21.18 ml/m² ± 13.9). The l/R ratio was obtained in 18 affected and in all normal dogs. The l/R was not significantly different between the groups (2.3 vs 1.67). LVMI 0.4 vs 0.74 cm² ± 0.18, whereas the l/R ratio was significantly decreased in affected dogs (0.41 ± 0.12 vs. 0.53 ± 0.11).

Data from this study show that although FS in affected dogs is increased, as it should be expected in volume overload caused by CMVD, the increased ESV-I in these dogs suggests that a mild systolic dysfunction was present also with moderate heart failure. In fact, ESV-I is believed to be a reliable indicator of systolic function. Inadequate hypertrophy of left ventricle could be responsible for this finding, as it has been demonstrated in experimental models.
been associated with the development of DCM in Springer Spaniels. The fit dogs showed initial echocardiographic characteristics of early DCM, where the non-fit dogs had normal echocardiographical parameters. All fit dogs had LVDd in the higher range of normal, LVDs out of reference range, normal ventricular wall thicknesses, but FS < 25% (18–23%). Obvious mitral and tricuspid valve regurgitation was present in all fit dogs, despite the AV valves having a normal appearance. No atrial enlargement was visible. Pulmonary insufficiency was visible in 2/3 dogs. Plasma taurine levels were within reference range (>50 nmol/ml). At long-term follow-up none of the dogs showed clinical signs. One of the non-fit dogs had now reached the level of fitness of the fit dogs. At this time all fit dogs had audible murmurs over the left apex (1–3/6), and one dog had an audible murmur over the tricuspid valve. Exercise did not make the murmurs disappear. Echocardiographically, LVDd had now further increased exceeding reference range, as did LVDs. FS had continued to decline in 2 dogs (min 13%), but remained stable (≤25%) in the others. Mitral and tricuspid regurgitation was more marked in all dogs without associated atrial enlargement.

In conclusion, traditional echocardiography might be very misleading in the differentiation between AH and DCM. The dog’s physical condition should be taken into consideration and long-term follow-up is advised before condemning these animals.

NEUROHORMONAL AND CIRCULATORY EFFECTS OF SHORT-TERM TREATMENT WITH ENALAPRIL AND QUINAPRIL IN DOGS WITH ASYMPTOMATIC MITRAL REGURGITATION. S. C. Hall, A. T. Mann, B. J. Siegrist, S. F. M. Glaus, D. R. Stevenson, A. L. Pedersen, T. T. Torellin, I. Heggsstrøm, H. D. Pedersen. 1Department of Basic Animal and Veterinary Sciences, The Royal Veterinary and Agricultural University, Fredensborg, Denmark; 2Department of Clinical Chemistry, VU University Medical Center, Amsterdam, The Netherlands; 3Faculty of Veterinary Medicine, Swedish University of Agricultural Sciences, Uppsala, Sweden.

Mitril regurgitation (MR) is a common cause of heart failure in dogs. Cavalier King Charles Spaniels (CKCS) are predisposed to the disease and often develop asymptomatic MR at a young age, which seems to be associated with a decreased production of nitric oxide (NO) (measured as the stable metabolites nitrate and nitrite in plasma (NOx)). Activation of angiotensin-converting enzyme (ACE) in heart failure might lead to reduced endothelial NO release. Accordingly, ACE inhibition has been shown to improve endothelial function in humans, most likely by increasing the availability of NO. In that context, quinapril has been shown to have a superior effect on endothelial function compared to enalapril. In CKCS with MR enalapril does not prolong the asymptomatic period where endothelial dysfunction seems to be present. Quinapril, however, might be able to prolong the asymptomatic period of MR—provided that the results seen in humans regarding improved endothelial function can be reproduced in dogs.

The aim of the study was to compare the effect enalapril and quinapril on neurohormonal and circulatory parameters in CKCS with asymptomatic MR. Ten CKCS with mild to severe untreated MR completed a protocol were they were treated with quinapril and enalapril (0.5 mg/kg PO SID for 7 days), in a double-blind cross-over study with a wash out period of 7 days between treatments. Blood samples were drawn and echocardiography was performed at 0, 7, 14, and 21. Both treatments reduced ACE activity (P < .001) and increased renin activity (P < .001) and atrial natriuretic peptide concentration (P < .005). The ACE inhibitors had no effect on the plasma concentration of NOx or asymmetric dimethylarginine (ADMA). On day 0, a lower NOx concentration (P = .02) was found in samples taken in the clinic as compared to samples taken in the home of the dogs. Quinapril caused a significant reduction in a greater number of variables reflecting the severity of MR (e.g. jet size and left ventricular end diastolic diameter) than was found with enalapril. However, in terms of specific parameters, there was no significant difference between the effects of the two treatments on MR.

These results suggest that ACE inhibitors do not affect NOx and ADMA concentrations in asymptomatic dogs with MR, however, stress in connection with clinical examination may influence NOx concentrations in these dogs.

CORONARY FLOW RESERVE MEASURED BY POSITRON EMISSION TOMOGRAPHY IN HEALTHY CATS: ADENOSINE DOSE FINDING STUDY. T. S. Jensen, T. Schleppe, N. Friis, D. P. Siegrist, M. Belpolasi, S. R. Kaestner, C. E. Reusch, P. A. Kaufmann, T. M. Glaas. 1Divisions of Cardiology, 2Clinic for Small Internal Medicine, 3Anesthesiology, 4Vetsuisse Faculty, and 5Nuclear Cardiology and Echocardiography, Cardiovascular Centre, University of Zurich, Switzerland.

In humans, positron emission tomography (PET) is the gold standard to measure myocardial blood flow (MBF). Coronary flow reserve (CFR) is defined as the ratio of maximal MBF e.g. stimulated by administration of vasodilators, divided by resting MBF. An adenosine constant rate infusion for 7 minutes is used to determine the maximal MBF, and normally coronary flow increases 2.5–4 fold. The purpose of this study is to establish a protocol to determine MBF and CFR by PET in healthy cats for future reference in cats with hypertrophic cardiomyopathy. Emphasis was placed on the adenosine dose to produce maximal CFR and its adverse effects.

In the first part MBF was measured at rest and during adenosine at the human standard dose and rate of 0.14 µg/kg/min using PET with ‘N-ammonia (‘N-NH3) and 15 O-water (‘H2 O). There was no increase of MBF and ‘H2O was found to be unsuitable. For the main study, in 4 healthy cats anesthesia was induced with propofol, maintained with isoflurane/oxygen by inhalation. In two cats (cat 1 and 2) also direct arterial blood pressure (BP, indicated as mean MAP) was monitored invasively through a catheter placed into the femoral artery. After MBF baseline measurement, each cat was challenged with adenosine infusions at different doses 50 min apart, randomly assigned. The doses were 0.28 (2 cats), 0.56 (3 cats), 0.84 (2 cats) and 1.12 µg/kg/min (1 cat) for 7 minutes. In the individual cats baseline MBF were 1.72, 1.59, 1.54, 1.38 ml/g/min, maximal MBF were 2.68, 2.45, 2.32, 2.51 ml/g/min, resulting in a CFR of 1.55, 1.54, 1.50, 1.81. MBF did not correlate with adenosine dose, instead MBF decreased in 2 cats at 0.84 and 1.12 µg/kg/min, respectively. In cat 2 with invasive BP measurement there was MAP decrease from a baseline of 89 mmHg to 56 mmHg and 61 mmHg during 0.28 µg/kg/min and 0.84 µg/kg/min adenosine, respectively, returning to baseline within one min at the end of infusion. In cat 2 there was no relevant MAP decrease at 0.56 µg/kg/min and 1.12 µg/kg/min. At 1.12 µg/kg/min adenosine, the cat developed palpable reflex and a rise in heart rate from 159 to 172/min, which ceased/returned to baseline at the end of infusion. ECG abnormalities occurred only in one cat at 0.56 µg/kg/min adenosine, consisting of a short P wave not following any QRS complex; in the same cat, 1.12 µg/kg/min did not result in any abnormalities.

In conclusion, CFR assessed by PET under adenosine infusion is markedly lower in anesthetized cats compared to awake healthy humans. Increasing adenosine doses do not increase CFR and even high doses are tolerated without relevant adverse effects. A dose of 0.56 µg/kg/min seems adequate to study CFR in cats.
ASSAY VALIDATION OF VARIOUS IMMUNOASSAYS FOR THE QUANTIFICATION OF CARDIOVASCULAR PEPTIDES (BNP, ProANP, and ANP) IN CANINE PLASMA SAMPLES. Neuroendocrine parameters of 3 plasma samples from 3 dogs were evaluated by the following methods: 1) ELISA for proAtrial Natriuretic Peptide (Vetsign CardioScreen, Guildhay Ltd, England: proANP), two radioimmunoassays for Brain Natriuretic Peptide (Peninsula Lab Inc, Belmont, CA; BNP), and Phoenix Pharmaceuticals Inc, Belmont, CA: BNPx), three assays for Endothelin-1 (ELISA, Biomedica, Vienna, Austria: ET-1x, EIA, Phoenix Pharmaceuticals Inc, Belmont, CA: ET-1x, EIA; IBL-Batburg, Germany: ET-1x), and two assays for Big Endothelin-1 (ELISA, Biomedica, Vienna, Austria: Big-ETx, EIA for rat Big ET-1, IBL-Batburg, Germany: Big-ETx). Validation included determination of intra-assay variability and dilutional parallelism. Intra-assay variability was determined by calculating the coefficients of variation (CV) for 10 replicates of 3 samples. Dilutional parallelism was assessed by serial dilution of 3 high-concentration or spiked samples. Results were evaluated for dilutional parallelism comparing observed with expected values. The correlation coefficient between observed and expected peptide concentrations was calculated.

For all ANP assays, CVs of plasma samples were 4.9%, 10.3% and 4.2%, for BNPx, 47.2%, 21.2% and 24.3%, for BNPx, 8.2%, 12.7% and 21.8%, for ET-1x, 2.1%, 2.4% and 5.3%, for ET-1x, 7.3% and 8.0% and 7.9%, for ET-1x, 17.5% and 18.8%, for Big-ETx, 5.0% and 2.4% and 8.6%, and for Big-ETx, 3.8% and 5.6% and 9.5%. Observed to expected (OE) ratios for four serial dilutions of three plasma samples ranged from 100.3–206.2% (median: 122.2%) with a correlation coefficient (R) of 0.992 for proANP, 59.2–146.5% (99.4%) and R = 0.995 for BNPx, 71.7–123.6% (98.8%) and R = 0.996 for BNPx, 161–600.5% (525.4%) and R = 0.844 for ET-1x, 94.4–460.2% (85.7%) and R = 0.667 for ET-1x, 56.3–205.6% (95.2%) and R = 0.996 for ET-1x, 241.9–1407% (579.1%) and R = 0.008 for Big-ETx, and 96.5–132.9% (111.8%) with a correlation coefficient of 0.992 for Big-ETx.

In conclusion, the study showed large differences between the performance of the assays. Considering high CVs and poor correlations and recovery in dilution studies several assays do not seem to be appropriate for use in dogs. Nevertheless, the assays for proANP, BNPx, ET-1x, and Big-ETx are expected to give reliable results in dogs, and, depending on additional validating studies, may be useful to study cardiovascular compensatory mechanisms in this species.
In conclusion, a low percentage of murmurs was found on the PCGs from dogs with mild mitral regurgitation. Still, electronic stethoscope might improve the accuracy of diagnostic procedures of less experienced observers. Further studies are needed to evaluate the use of the newer versions of the electronic stethoscope system for diagnosing mild mitral regurgitation in dogs.

16 EXPERIENCES WITH DUAL-CHAMBER PACEMAKER IMPLANTATION IN DOGS. N Hildebrandt1, WA Stiertmann2, M Wehner1, J Schneider1, H Neu2, M Schneider2. Small Animal Clinic (Internal Medicine) and ‘General Surgery, Justus-Liebig-University, Giessen, Germany.

Between December 1997 and November 2004 in 33 dogs (median body weight 27.0 kg, range 7.0–40.0 kg; median age 95.2 months, range 21.1–160.6 months) with a second (n = 3) or third (n = 30) degree AV block, unresponsive to a medical treatment and clinical signs like syncope or exercise intolerance, a dual-chamber pacemaker system was implanted. The most common breeds were German Wirehair Pointer (n = 7), Cocker Spaniels (n = 2), Labrador Retriever (n = 2) and mixed breed dogs (n = 9).

Under general anaesthesia and fluoroscopic control one lead was placed into the right ventricle and the second lead into the right atrium by a transvenous access. Lead fixation to the myocardium was accomplished by an active screw in tip and a passive lead tip with siltic tip. With a pacing system and at the threshold voltage, the system impedance and the amplitude of R- and P-wave were measured. The pulse generator was placed beneath the skin of the neck prescapular. Postoperative care included a neck bandaging and antibiotic treatment with amoxicillin/clavulanic acid (20 mg/kg body weight) over a 10-day period. In the time of hospitalization the programming was checked and if necessary reprogrammed by telemetry.

In all 33 patients (100%) a dual-chamber pacemaker implantation and a programming in a DDD pacing mode was possible. One patient died postoperatively because of a larynx edema. Malfunctions of the ventricular lead did not occur. Atrial lead disturbances were present in four patients and included lead dislodgement (n = 2), lead malfunction (n = 1) and improper atrial sensing (n = 1). In all four dogs these problems were eliminated by a second intervention. Consequently in 32/33 patients an atroventricular synchrony was restored.

In conclusion the transvenous implantation of a dual-chamber pacemaker in dogs with AV block and clinical signs is possible. A reconnection of atrium and ventricle could be achieved by programming the pacemaker in a DDDR- or DDDDR-mode. The heart frequency adaptation is then achieved accordingly to the physiological centre, the sinus node. This results in a normal exercise efficiency.

17 EFFECT OF MILD OVERHYDRATION ON PLASMA EXOGENOUS CREATININE CLEARANCE TEST IN HEALTHY CATS. A Le Garre1, S Noël1, F Billen1, HP Lefebvre1, C Clercx2. ‘Dep. Medicine, Faculty of Veterinary Medicine, University of Liège, Liège, Belgium; ‘Physiology and Pharmacology, National Veterinary School of Toulouse, France.

Determination of glomerular filtration rate (GFR) in companion animal practice allows early diagnosis of impaired renal function. Assessment of GFR by plasma exogenous creatinine clearance (PECCT) test has been described in dogs and cats. Hydration status may affect GFR value. The aim of this study was to evaluate the effect of IV mild overhydration on GFR estimated by pECCT. Besides, 2 analytical conditions were compared for determination of creatinine.

PECCT was measured for GFR assessment in 10 healthy cats (6M and 4F; 7 months to 17 years, 3.6 to 6.5 kg). Health was based on clinical examination and biochemistry and panel results were obtained in all cats. Creatinine (40 mg/kg) was injected by IV bolus in normohydrated 10-hours fasted conscious animals and jugular blood samples were obtained before, 30, 120 and 600 minutes after injection. Immediately at the end of the first test, cats were placed under perfusion with balanced fluid (Ringer lactate) using an infusion pump, at a rate of 4 ml/kg/hour during 48h; the test was repeated after 38h of infusion. Water was given ad libitum for the whole period and the test was stopped at 20°C at the end of each test and all plasma creatinine values (Pl-creat) were assayed simultaneously by an enzymatic method (Hitachi 917). Additionally, Pl-creatinine concentration versus time curve using a Winnonlin noncompartimental analysis program.

The healthy dogs and the clinical cases had a mean GFR of respectively 3.1 mL/min/1.73 m2 and 1.9 mL/min/1.73 m2. The dogs with normal GFRs additional testing assessed etiologies of pu/pd different from renal insufficiency in 4 out of 5 cases. The PECCT required a 10-hour hospitalisation and was easy to perform, requiring nothing more than accurate sample prevelation and handling. The results of this study suggest that the PECCT is a useful aid in the diagnostic work-up of pu/pd in the absence of an obvious etiology.

18 POLYURIA-POLYDIPSIA IN DOGS: USEFULNESS OF GFR ASSESSMENT THROUGH CREATININE CLEARANCE TESTING FOR DIFFERENTIAL DIAGNOSIS. M Diederich1, J Arons1, J A Le Garre2, J Van der Heyden1, H Lefebvre1, K Gommeren1, D Paape5, S Stümpel1. ‘Dep Med Clin Biol Sm Anim, Faculty of Veterinary Medicine, Ghent University, Belgium; ‘Phys Therp, National Veterinary School of Toulouse, France.

The plasma exogenous creatinine clearance test (PECCT) in blood was established by Watson et al (2002) and has been found to be a reliable estimate of the glomerular filtration rate (GFR).

The objectives of this study were to determine GFR-values in a group of healthy dogs from different sizes and to evaluate the usefulness of the PECCT in clinical cases with polyuria/polydipsia (pu/pd) without an obvious etiology. Therefore healthy dogs of 15 different breeds entered the study (median age 3 years ± 1.9; mean weight 16.1 kg ± 9.3). Eight dogs, all of different breeds (mean age 3.8 years ± 3.5) with pu/pd without azotemia entered the study.

The PECCT proved to be a valuable diagnostic tool in the work-up of pu/pd-patients, allowing to detect early renal insufficiency in 2 patients without azotemia. In the dogs with normal GFRs additional testing assessed etiologies of pu/pd different from renal insufficiency in 4 out of 5 cases. The PECCT required a 10-hour hospitalisation and was easy to perform, requiring nothing more than accurate sample prevelation and handling. The results of this study suggest that the PECCT is a useful aid in the diagnostic work-up of pu/pd in the absence of an obvious etiology.

19 BACTERIURIA IN FELINE LOWER URINARY TRACT DISORDERS (FLUTD). J Arons1, HS Lund5, R Krontveit1, AV Eggersdottir5. ‘Department of Companion Animal Clinical Sciences and ‘Department of Food Safety and Infection Biology, Norwegian School of Veterinary Science (NSVS), Norway.

A recent study from the US reported that lower urinary tract signs affected approximately 1.5% of cats presented to private companion animal clinics. University-based studies (US and UK) during the last forty years operate with the following numbers: less than 2% of the cats presented with non-obstructive FLUTD have bacterial infection. 2% of the cats with obstructive FLUTD have a combination of uroliths and bacterial infection. In 2003 and 2004, 134 cats presented with signs of lower urinary tract disorders were included in a study at NSVS. The majority of the cases were first opinion cases (>95%). All the cats went through a physical examination, blood samples for haematology and clinical chemistry were collected. The urinary analysis included urine stix, specific gravity, and microscopic examination of sediment as well as culture and sensitivity tests. An ultrasound examination was done in most cases, but a few had x-rays done instead. Information about previous/concurrent disease, feeding regime, the cats’ environment (out-door access etc.), the cats’ temperament and the weather when the signs were observed, was registered through a standardised questionnaire. The cats received individual treatment, depending on clinical findings and diagnosis.

Our results diverge from those of other studies: the most striking finding was the large proportion of cats with bacteriuria. Only samples cultured on the day of collection, with growth >105 CFU/ml urine, were considered indicative of bacterial infection. Of the cats with non-obstructive FLUTD, 22% had bacterial infection. 1% a combination of uroliths and bacterial infection, and 5% a combination of crystals and bacterial infection. Of the cats with obstructive FLUTD, 5% had bacterial infection, 4% a combination of uroliths and bacterial infection, and 5% a combination of crystals and bacterial infection.

In the US and UK the majority of cats taken to the veterinarian are indoor cats, while many of the Norwegian cats are free to roam outdoors. This may influence e.g. diet, degree of exercise and water intake. The owners’ control of the cats’ drinking, eating and urinating habits may also be limited, thus influencing...
The ovarian remnant syndrome (ORS) is a condition that occurs when a neutered queen returns to oestrus behaviour and is a complication of castration. Although ORS is defined by the presence of functional ovarian tissue, the observed oestrus signs may have different underlying causes and a specific diagnosis is crucial to appropriate clinical management. Little information is currently available about ORS and the appropriate diagnostic method. The aims of the present study were to identify those historical, clinical and diagnostic findings which might assist the clinician in making a diagnosis. Case records of cats referred to our laboratory between 1997 and 2002 were reviewed. 147 cats were suitable for inclusion in the study. Estradiol (E2), progesterone (PG) and prolactin (PRL) were assayed. ORS was diagnosed in 70 cats (group A) by hCG stimulation test (Englund, 1997) whereas no stimulation test had been performed on 77 cats (group B). We usually only recommend the hCG stimulation test when E2 is normal in the first sample.

The queens ranged from 1 to 14 y of age (mean 4 ± 2.7 y). 125 were European short-haired. Duration of clinical signs prior to presentation ranged from 1 to 5 weeks (mean 3.5 mo). Clinical signs included typical signs of oestrus (n=14), vocalizations (123), aggression (9), polyuria/polydipsia (7), weight loss (3), polyphagia (8). Age at neutering was documented in 79 cases and the mean age was 2.6 y. Only 21 cats had been neutered before 1 y. In group A, diagnosis was confirmed for 51/70 cats. Ovulation was not induced in 13 cats with hyperTSH (1), hyperE2 (120 ng/mL), hyperPG (6 ng/mL) and hyPOE2 (120 ng/mL). Eight of these animals were diagnosed with anadromous ORS and PG values in 6 cats remained within the normal range suggesting an adrenal origin of oestrus signs. In group A, the frequency of ORS diagnosis was significantly higher in queens neutered after 1 y (p = 0.01). Treatment was documented in 29 cats. 24 cats underwent surgery with 88% success. Medical management had been used in 2 cats and results unexpectedly, signs resolved spontaneously in 3 cats. Diagnosis in group B, was only conclusive for 1277 cats: 5 queens had hyper PG (12 ± 4 mM/L) indicating that ovulation has occurred hence confirming ORS and 7 showed hypopRL (25 ± 5 ng/mL) with low E2 and PG values. This study confirms the suitability of the hCG stimulation test as method of diagnosis and indicates that clinical signs can be seen months to years after castration. It was apparent in our population that late castration increased the risk of ORS. It should be stressed that the clinical signs of ORS cannot always be treated by surgery.


22 THYROID ENLARGEMENT AND ITS RELATIONSHIP TO SE- RUM T4 STATUS IN CLINICALLY SUSPECTED HYPERHY-ROID CATS. FS Boretti, NS Sieber-Ruckstuhl, P Laluha, CE Reusch. Clinic for Small Animal Internal Medicine, Vetsuisse Fac- ulty, University of Zurich, Switzerland.

Thyroid enlargement has been described as typical clinical finding in hyperthyroid cats. However, there is increasing evidence that the presence of a goiter alone is a poor indicator of symptomatic hyperthyroidism. Aim of the present study was, to relate thyroid size to serum T4 status in a group of cats with suspected hyperthyroidism.

The ventral neck of 161 cats presented to the Clinic for Small Animal Internal Medicine, University of Zurich with clinical signs consistent with hyperthyroidism was examined by 2 independent observers experienced with thyroid palpation using a semi-quantitative palpation system. Individual thyroid gland size of each side was scored between 0 (palpable) and a maximum of 6 (nodule >25 mm), with score 1 = 1–3 mm, score 2 = 3–5, score 3 = 5–8, score 4 = 8–12, score 5 = 12–25 mm. Allocation into scores was performed according to the largest detected nodule. In addition, total score sum of all palpable lobes of each cat was calculated. In cases, in which discrepancy between the 2 observers existed, mean score values of the largest lobe and mean score sum values were calculated. Serum T4 concentrations were determined and compared to thyroid palpation results.

In 117 of the 161 cats, one or more thyroid nodules were palpable by both observers with a score of 1 to 4. Based on the results of the T4-measurements, only 17 of the cats were classified as hyperthyroid and 144 as euthyroid. Thus, all of the hyperthyroid and 69.4% of the cats with a normal T4 had an enlarged thyroid gland.

Median (range) palpation scores and score sum of the hyperthyroid cats were 3.5 (1–4) and 4 (1–7) and in the euthyroid 1 (0–4) and 1.5 (0–6), which was statistically significantly different. Evaluation of the cats with a palpable nodule revealed a mild but significant positive correlation between the score of the largest lobe and the T4 concentration and between the score sum of all palpable lobes and the T4 concentration. However, three cats with a normal T4 had a score of 3.5, 3.5 and 4, respectively.

Results of our study show that a considerable number of cats with palpable thyroid glands has a normal T4 concentration. Therefore, no reliable conclusion on functional status of the thyroid can be drawn using its size. However, the likelihood of goiter increases with increasing size of the gland.

23 SOMATIC MUTATIONS OF THE THYROID STIMULATING HORMONE RECEPTOR GENE IN FELINE HYPERTHYROID- ISM. SG Watson, L Blackwood, AD Radford, A Kipar, P Ibar- rola, ‘Small Animal Hospital, Department of Veterinary Clinical Science, and Department of Veterinary Pathology, University of Liverpool, United Kingdom.

The aetopathogenesis of feline hyperthyroidism is complex and multifactorial and has not been fully elucidated. Dietary, environmental and genetic factors have all been implicated, but there has been little work on the genetic lesions associated with hyperthyroidism in cats. In hyperthyroid humans with toxic nodular goitre (TNG) or functional thyroid adenomas, activating mutations of the thyroid stimulating hormone receptor (TSHR) gene are common, but to date similar mutations have not been found in cats. In humans, most of these mutations occur in exon 10 of the transmembrane domain of the TSHR gene. At the amino acid level, the feline TSHR is 92% homologous to the human TSHR, and feline hyperthyroidism and human TNG are clinically and histopathologically similar. Our hypothesis was that similar mutations exist in hyperplastic/adeno- matous nodules in hyperthyroid cats.

Genomic DNA was extracted from 134 hyperplastic/adenomatous nodules (from 50 hyperthyroid cats), and analysed for the presence of mutations in exon 10 of the TSHR gene (cods 399 to 664) by polymerase chain reaction (PCR) amplification and sequencing of PCR products. Eleven different mutations were detected, one silent and 10 missense (three somatic mutations, one germline mutation/polyorphism, and six unknown). Twenty eight of the 50 cats (67/134 nodules) had at least one missense mutation. The missense mutations were coded as: met452thr (25 nodules), ser504ala (two different mutational forms) in two cats (two nodules), val508gln in one cat (three nodules), arg530glu in one cat (two nodules), val557leu in 13 cats (36 nodules), thr631ala or...
Diabetic autonomic neuropathy (DAN) is frequently recognised in human patients with diabetes mellitus. Parasympathetic dysfunction seems to be more profound than sympathetic dysfunction in those affected resulting in decreased heart rate variability and a higher resting heart rate than controls. Those diagnosed with DAN have a calculated five-year mortality rate of 56%. Analysis of heart rate variability in dogs with experimentally induced diabetes mellitus and have failed to demonstrate evidence of DAN.

This study used the neuroscope as an indicator of cardiac vagal tone. The neuroscope provides a real time measure of instantaneous cardiac vagal tone from a high resolution time domain analysis of R-R interval data. The output of the neuroscope is termed the Cardiac Index of Vagal Activity (CIPA), which is measured in units of the linear vagal scale.

The purpose of this study was to look for the existence of differences in autonomic tone between a population of dogs with naturally occurring diabetes mellitus and a population of control dogs. As part of the study, we wished to determine whether there was any correlation between cardiac vagal tone and duration of diabetes and adequacy of glycaemic control.

The demonstration that DAN exists in dogs is a new finding. Future studies will be required to follow canine diabetics from diagnosis through their disease progression, to establish whether DAN has similar effect on survival as in their human counterparts.

Diabetes mellitus (DM) is one of the most common endocrinopathies in cats. Most diabetic cats are routinely treated with insulin. This, however, does not always lead to normalization of the metabolic situation. Because pancreatic beta cells produce not only insulin but also amylin, it was recognized, that DM is a disease of insulin deficiency. Amylin reduces pancreatic glucagon secretion in various species. Hence, lack of amylin may contribute to postprandial hypersecretion of glucagon in diabetic individuals. It was therefore the aim of the present study to establish a dose-response relationship for amylin’s effect to normalize pancreatic glucagon secretion in cats.

Our preclinical study was performed in 12 healthy male cats (2.4 ± 0.23 years; 4.9 ± 0.4 kg). The effect of amylin on blood levels of glucose, insulin and glucagon was measured in an arginine stimulation and meal response test in cats. D Furrer1, F Tschirch2, C Reusch1, TA Lutz1. 1Institute of Veterinary Physiology, Clinic for Small Animal Internal Medicine, Vetsuisse Faculty University of Zurich, Switzerland.

The objective of our study was to investigate the effect of trilostane on cortisol and cortisone concentrations in dogs with pituitary-dependent hyperadrenocorticism. NS Sieber-Ruckstuhl1, F Roretti1, M Wenger1, Ch Maier-Gluth2, Ch Reusch3. 1Clinic for Small Animal Internal Medicine, Vetsuisse Faculty University of Zurich, Zurich, Switzerland; 2Steroid Laboratory, Institute of Pharmacology, Ruprecht-Karls-University, Heidelberg, Germany.

Trilostane is used increasingly as a first line treatment for dogs with pituitary-dependent hyperadrenocorticism. From studies in human medicine assumptions about its mechanism of action in dogs have been drawn, but have not been confirmed. Recently we have proven that trilostane has an inhibitory effect on the 3β-hydroxysteroid dehydrogenase enzyme system (3β-HSD) in dogs. In this preliminary study we hypothesized that trilostane inhibits the 3β-HSD incompletely and has an additional effect more distal in the enzyme cascade, either on the 11β-hydroxylase or on the 11β-hydroxysteroid dehydrogenase (11β-HSD).

The objective of our study was to investigate the effect of trilostane on cortisol and cortisone concentrations and on the cortisol-cortisone-ratio in dogs with PDH.

Cortisol and cortisone concentrations were evaluated in 8 dogs before and 1 hour after injection of synthetic ACTH on day 0 (10), weeks 1–2 (11), and weeks 3–7 (12) of trilostane treatment.

Serum cortisol and cortisone concentrations before ACTH stimulation did not change significantly during trilostane treatment. Blood samples collected after ACTH stimulation were significantly decreased at t1 and t2.

Cortisol values decreased about 70.5–98.5% (median: 92.9) at t1 and about 85–90% at t2. Cortisone values were decreased about 80.5–98% (median: 92.9) at t1 and about 90–98% at t2.
28

TRH-INDUCED GH SECRETION IN DOGS WITH PRIMARY HYPOTHYROIDISM (PH). MM Diaz Espinera, CMM Schellens, HS Kooistra. Department of Clinical Sciences of Companion Animals, Faculty of Veterinary Medicine, Utrecht University, Utrecht, The Netherlands.

Primary hypothyroidism (PH) in dogs is associated with elevated basal GH secretion (Lee et al. 2001). This elevated GH secretion has endocrine significance as illustrated by elevated plasma IGF-I levels and some physical changes mimicking acromegaly (Lee et al. 2001). In PH the hypothalamic release of TRH is increased. In addition, expression of TRH receptor mRNA has been demonstrated to be increased in the PH thyroid gland (Lee et al. 2001). We therefore tested the hypothesis that the increased GH release in PH dogs is due to a paradoxical response of the somatotrophs to TRH. In 10 dogs with ages ranging from 3 to 11 years, the diagnosis PH was based upon clinical signs, a plasma T4 concentration below 2 nmol/l, a plasma TSH concentration above 1.0 mU/l (range 1.6 to 9.5 mU/l), and insufficient uptake of the radioactive isotope of pertechnetate on thyroid scintigraphy. Blood samples for the determination of the plasma GH concentration were collected on thyroid scintigraphy. Blood samples for the determination of the plasma GH concentration were collected – 15, 5, 10, 20, 30, and 45 minutes after the i.v. administration of 10 µg TRH/kg body weight. The mean basal GH concentration in the hypothyroid dogs was relatively high (2.5 µg/l) compared with that in healthy dogs. In 8 of the 10 hypothyroid dogs TRH-administration resulted in a significant rise in plasma GH concentration (Figure 1). In healthy dogs, TRH administration does not result in changes of the plasma GH concentration (Rutteman et al. 1987). The results of this study indicate that the elevated plasma GH levels in dogs with PH may in part at least be due to the increased release of TRH.

29

FELINE HYPERTENSION: THE ASSOCIATIONS BETWEEN LONG TERM BLOOD PRESSURE CONTROL AND SURVIVAL. RE Jepson1, HM Syme1, J Elliott2. 1The Royal Veterinary College, London, UK. 2Hidefield Veterinary Hospital, Glasgow, UK.

Feline hypertension has frequently been documented in association with chronic renal failure and hyperthyroidism. To date, a large-scale study examining the association between hypertension, the adequacy of systolic blood pressure (SBP) control and survival is lacking. The aim of this study was to examine whether there is a relationship between adequacy of blood pressure control and long-term survival in a large population of naturally occurring hypertensive cats.

Hypertensive cats, from geriatric feline clinics held at two central London first opinion practices since 1998, were selected retrospectively. SBP was measured using the Doppler technique and hypertension diagnosed as SBP>170 mmHg on two occasions or once but in association with clinical manifestations of hypertension. Initial investigations where possible included full physical examination, indirect fundus examination, plasma biochemistry, urinalysis and total T4 if findings were indicative of hyperthyroidism. All hypertensive cats were treated with amiodipine besylate (initially 0.625 mg SID) and the majority of cats were subsequently examined and SBP measured at approximately 6±12 week intervals. The values for RI and PI of arcuate arteries were significantly higher in sedated Beagles (P = 0.016). There was 290, 286.5, 244, and 132.5 days. These data showed an apparent relationship between time averaged SBP and SBP at diagnosis (P = 0.013) and between time averaged SBP and SBP at diagnosis (P = 0.001). The mean blood pressure in dogs with PH was 190.6 mmHg (Q1, 191 mmHg (Q2), 191.2 mmHg (Q3) and 208 mmHg (Q4) and the time averaged median SBP for each quartile was 139.8 mmHg (Q1), 151.5 mmHg (Q2), 159.9 mmHg (Q3), 174.4 mmHg (Q4). The median survival time for Q1 to Q4 respectively was 290, 286.5, 244, and 132.5 days. These data showed an apparent relationship between time averaged SBP and SBP at diagnosis (P = 0.013) and between time averaged SBP and SBP at diagnosis (P = 0.013). The mean blood pressure in dogs with PH was 190.6 mmHg (Q1, 191 mmHg (Q2), 191.2 mmHg (Q3) and 208 mmHg (Q4) and the time averaged median SBP for each quartile was 139.8 mmHg (Q1), 151.5 mmHg (Q2), 159.9 mmHg (Q3), 174.4 mmHg (Q4). The median survival time for Q1 to Q4 respectively was 290, 286.5, 244, and 132.5 days. These data showed an apparent relationship between time averaged SBP and SBP at diagnosis (P = 0.013) and between time averaged SBP and SBP at diagnosis (P = 0.013). The mean blood pressure in dogs with PH was 190.6 mmHg (Q1, 191 mmHg (Q2), 191.2 mmHg (Q3) and 208 mmHg (Q4) and the time averaged median SBP for each quartile was 139.8 mmHg (Q1), 151.5 mmHg (Q2), 159.9 mmHg (Q3), 174.4 mmHg (Q4). The median survival time for Q1 to Q4 respectively was 290, 286.5, 244, and 132.5 days. These data showed an apparent relationship between time averaged SBP and SBP at diagnosis (P = 0.013) and between time averaged SBP and SBP at diagnosis (P = 0.013). The mean blood pressure in dogs with PH was 190.6 mmHg (Q1, 191 mmHg (Q2), 191.2 mmHg (Q3) and 208 mmHg (Q4) and the time averaged median SBP for each quartile was 139.8 mmHg (Q1), 151.5 mmHg (Q2), 159.9 mmHg (Q3), 174.4 mmHg (Q4). The median survival time for Q1 to Q4 respectively was 290, 286.5, 244, and 132.5 days. These data showed an apparent relationship between time averaged SBP and SBP at diagnosis (P = 0.013) and between time averaged SBP and SBP at diagnosis (P = 0.013). The mean blood pressure in dogs with PH was 190.6 mmHg (Q1, 191 mmHg (Q2), 191.2 mmHg (Q3) and 208 mmHg (Q4) and the time averaged median SBP for each quartile was 139.8 mmHg (Q1), 151.5 mmHg (Q2), 159.9 mmHg (Q3), 174.4 mmHg (Q4). The median survival time for Q1 to Q4 respectively was 290, 286.5, 244, and 132.5 days. These data showed an apparent relationship between time averaged SBP and SBP at diagnosis (P = 0.013) and between time averaged SBP and SBP at diagnosis (P = 0.013).
32 IS THERE A DIFFERENCE BETWEEN ENERGY BALANCE OF CATS HOSPITALISED FOR SURGICAL OR FOR MEDICAL REASONS? E Lheureux1, S Claeys2, A Gabriel1, J Detilleux1, M Balligand1, L Issac2, M Debe2. 1Animal Nutrition Unit, Faculty of Veterinary Medicine, U.Lg. 2Small Animal Medicine Unit, Faculty of Veterinary Medicine, U.Lg. Belgium, 3Quantitative Genetic Unit, Faculty of Veterinary Medicine, U.Lg. Belgium.

During hospitalisation, most cats are thought to be in negative energy balance (NEB). NEB can induce hepatic lipodystrophy, increase morbidity, mortality and has the potential of influencing proper medical or surgical management of hospitalised cases. The objectives of this study were to estimate percentage of cats in NEB and, to determine the reasons of NEB, to observe if there was a difference between medicine and surgery units. We included 75 cats (29 from medicine and 46 from surgery) hospitalised at the veterinary school of Liège for at least 2 days from November 2003 to March 2004 and from November 2004 to March 2005 into equivalent conditions (same room and same medical staff). For each cat, breed, gender, age, length of hospitalisation, body condition score (BCS) on a 6-point scale and physical status score (PSS; from 1 = normal patient to 5 = moribund) were recorded. Their energy requirement (ER) during hospitalisation ER was calculated using the equation BW x 70 kcal1 multiplied by a factor of (1.2 to 1.6) to derive the illness ER (IER). When 80% of IER was covered by spontaneous feeding or by nasoenteropageal tube at day 2, patient was considered in positive energy balance. Correlation analyses were performed using the SAS system.

Domestic Shorthair cats represented 81% of patients and 57% were female in the 2 units. The average age was 6.2 years in medicine and 4.3 years in surgery. BCS averaged 3 in the 2 populations, and it was associated with gender (males were heavier) and correlated positively with age (P < 0.05). The cats had an equal average hospital stay of 5 days for the 2 units. A significant relationship between PSS (mean PSS = 2.6 in medicine vs 1.6 in surgery) and unit (P < 0.05) was observed and was reflected by the mortality rate (10.3% in medicine and 4.3% in surgery).

Energy balance was negative in 52% of cases without difference between medicine and surgery: 30% were due to lack of compliance with written feeding rate (10.3% in medicine and 4.3% in surgery).

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33 THE EFFECT OF DIETARY FISH OIL ON PUPPY TRAINABILITY. R Kelley1, A Lepine1, J Burr1, M Shyan-Norwalt1, G Reinhart1, P Ribiczey1, J Jakus1, P Ribiczey1, J Jakus1, M Mezes3, V Kunos1, D Szecsenyi1, C Szentirmai1, T Guall1. 1Dep Internal Medicine, Faculty of Vet Sci, Szent Istvan University, Budapest, Hungary; 2Bio-oxidation Group, Institute of Chemistry, Chemical Research Center, Hung Ac Sci, Budapest; 3Dep Nutrition, Faculty of Agricultural Environmental Sci, Szent Istvan University, Gödöllö, Hungary.

The long chain polyunsaturated fatty acid (LCPUFA), docosahexaenoic acid (DHA) is known to support correct neural development in mammalian species. A study was undertaken to examine the effect of DHA in the canine species. The objective was to determine the effect of maternal and post-weaning dietary fish oil on trainability and DHA status in Beagle puppies.

Twenty-eight (28) Beagle bitches (parity 2–3) were randomly assigned to 3 treatments (TRT) diets at breeding and maintained on those diets through lactation and weaning. Puppies selected from these litters (58 total puppies) were maintained on the respective dam diets from weaning through to 16 weeks of age for trainability assessment by Discrimination Task Testing using a Two-Arm T-maze. In the acquisition phase, all puppies were received daily socialisation exposure to the testing environment which concluded with 5 days of pre-test T-maze training at 9 weeks of age. Trainability testing was conducted from 10–16 weeks of age with puppies participating in 2 sessions/day of 7–10 trials/session, 5 days/week for 30 days. A success criterion was achieved when a puppy achieved an average score in at least 80% of trials for 2 consecutive sessions.

In addition, all puppies were assessed for fatty acid (FA) status at 7, 11, and 15 weeks of age based on RBC membrane FA profiles. TRT diets were poultry and fish oil. BCS and body fat were measured from blood samples taken before and during COPA therapy (cyclophosphamide, vincristine, prednisolone, doxorubicin). Following parameters were measured: reduced glutathione concentration (GSH) in blood plasma and red blood cell hemolysate (RBC-hem), oxidized glutathione concentration (GSSG) in plasma and RBC-hem, GSH/GSSG in plasma and RBC-hem.
glutathione-peroxidase activity (GSH-Px) in RBC-hem, malondialdehyde concentration (MDA, thiobarbituric-acid-reactive substance indicating lipid peroxidation) in RBC-hem, superoxide-dismutase activity (SOD) in RBC-hem, total antioxidant status based on the reduction of iron (FRAP) and vitamin-C concentration in plasma.

Values of the members of the antioxidant defence system in plasma showed a decrease from the basal values in various periods of the treatment then they started to raise until the end of the radiation course. Radiation therapy is an effective treatment for pituitary tumors, which could be used successfully in cats giving extended survival and control of both direct mass effect and para-neoplastic signs.

How to improve the target/non-target ratio—Locoregional application of radiopharmaceuticals. L Balogh, D Mihály, G Andócs, J Thüröczy, E Perger, A Polyák, K Király, GA Jánóki, 'National “FIC” Institute for Radiobiology and Radiohygiene, Budapest, Hungary; Department of Obstetrics and Reproduction, Semot István University, Faculty of Veterinary Science, Budapest, Hungary.

There are several systemic radiopharmaceutical treatment methods in humans and veterinary oncology where the goal of further development is to improve the tumor target dose and parallelly to avoid the normal organs from radiation. In this present study we aimed to find the idealistic application route of \(^{90}\)Re-colloid in multiple liver malignancies and \(^{90}\)Re-HEIDP, \(^{90}\)Lu-EĐTEMĐ in osteosarcoma dog patients and in normal Beagle dogs.

Altogether 8 normal Beagle dogs (4 to liver perfusion studies, 2 for foreleg and 2 for hindleg perfusion studies) and 2 multiple liver tumors (HSCs) and 5 osteosarcoma bearing (3 in forelegs, 2 in hindlegs) were used in locoregional treatments. Livers were injected by a. hepatica after laparatomie, forelegs were perfused via a. brachialis, and hindlegs were injected via a. femoralis while veins from the organs (v. hepatica, v. brachialis and v. femoralis) were clipped for a 30 minutes hypoxia period. A dynamic study during hypoxia period (0–30 min), than 30 min, 1, 2, 4, 6 hs, 1, 2, 5 ds and 1, 2 ws after application whole-body scans were taken. Regions of interest were drawn around the tumor, contralateral sites, visualized parenchymal and excreational organs, then mean residency times and internal dosimetric data were calculated using dedicated softwares (MIRDOSE and OLINDA).

The absorbed dose by the liver is 44–65% higher when \(^{90}\)Re-colloid is applied via a. hepatica compared to the liver dose achieved by the conventional intravenous application. Due to the existing vein anastomoses in the fore- and hindlegs, perfusion studies did not result very high overdose in the perfused region. Only 7–22% higher doses could be reached when radioisotopic is applied via arteries compared to the data of systemic intravenous injection.

Locoregional application of radioisotopic results a higher target/non-target ratio only if blood supply of the perfused organ is discrete. Thirty minutes hypoxic period in liver, fore- and hind leg was found to be well-tolerable for the dogs. Further studies need to elucidate the right place of intraarteric application and the vein-clips in the fore and hind legs in the dogs.

HMGA2 EXPRESSION IN CANINE PROSTATIC TISSUES—A POTENTIAL DIAGNOSTIC TOOL? S Winkler, H Munra Escobar, B Meyer, N Eberle, D Simon, I Buttenduf, I Neche, J Bullerzeller, Centre for Human Genetics, University of Bremen, Germany; Small Animal Clinic, School of Veterinary Medicine, Hanover, Germany.

Beside humans the dog is the only known mammalian species that spontaneously develops prostate cancer. Both species show striking similarities in the development and progress of the disease. In both species adenocarcinomas show the same histopathology and represent a locally invasive disease affecting mainly older subjects. Also in both species, the tumors are likely to metastasize to the same distant regions by blood or lymphatic system and akin to their human counterparts canine prostatic cancers vary with respect to their clinical behaviour. Based on the histology of the lesions alone it is often not possible to recognize sufficiently the malignant potential of the tumour in terms of local invasiveness and metastatic spread. Thus, molecular indicators allowing for a valid prognosis of these cancers are of considerable interest.

In humans, HMGA2 overexpression was recognized to be associated with a highly malignant phenotype of prostate cancer and is therefore considered a molecular marker in prostate cancer diagnosis. HMGA2 is a member of the High-Mobility Group Protein family, which is highly expressed in most normal adult tissues. Re-expression was detected in a variety of the human and canine proteins are highly conserved during evolution. HMGA1 is a member of the High-Mobility Group Protein family comprised of HMGA1a, HMGA1b, and HMGA2. All three proteins show a high amino acid sequence homology and the human and canine proteins are highly conserved during evolution. HMGA proteins are abundantly expressed during embryogenesis and are almost undetectable in most normal adult tissues. Re-expression was detected in a variety of human malignancies with correlation of the expression level with the degree of neoplastic cell transformation and metastatic tumor progression.

In this study we report the HMGA2 expression patterns as determined by real-time quantitative RT-PCR in prostate tissue from 16 dogs with histological findings comprising 4 samples with no abnormality detected, 3 hyperplasias, 3 cysts, and 6 carcinomas. The results show that expression of HMGA2...
is low in tissues with no abnormality detected, rises in benign neoplasms with intermediate values for cysts and hyperplasias and increases at least 19-fold in carcinomas. In our study all malignant neoplasms showed expression levels beyond an assumed threshold of 50,000 transcripts per 250 ng total RNA, whereas none of the non-malignant tissues showed expression levels beyond this threshold. These results indicate that HMGA2 expression analysis using real-time quantitative RT-PCR may provide a potential tool for better differentiation between varying degrees of malignancy in prostate carcinomas.

Preliminary results of the use of carprofen in canine mammary tumors. G Dank1, A Aung2, S Yudelevitch1. 1 Koret School of Veterinary Medicine, Hebrew University, Rehovot, Israel; 2 Patho Vet Laboratories, Rehovot, Israel.

Mammary gland tumors are the most common type of tumor in female dogs, constituting up to 52% of all neoplasms. The reported frequency of malignancy is 50%. Over expression of COX-2 has been reported in various tumors both in humans and animals and COX-2 over expression has been found to be more frequent and more intense in malignant canine mammary tumors as compared to benign mammary tumors. The aim of this study was to investigate the possible role of COX-2 inhibitors in the treatment of canine mammary tumors. We evaluated the effect of treatment with carprofen in dogs with mammary tumors on the expression of COX-2, tumor size and histopathology results.

Seven dogs with malignant tumors were included in this study. At the time of diagnosis a complete blood count, biochemistry panel, 3 view thoracic radiographs, digital imaging of the tumor, measurement of the tumor and an incisional biopsy were performed. If the biopsy revealed a malignant tumor, treatment was initiated with carprofen (2 mg/kg q 12 hours) for 4 weeks. Two weeks following this biopsy, biopsies were removed and the tumor was remeasured. Four weeks following the initial presentation the tumor was measured, imaged, and surgically removed. All samples were sent for histology and immunohistochemistry for expression of COX-2.

Of the 7 dogs with malignant tumors, one tumor reduced slightly in size over the treatment period. Results of the incisional biopsies revealed anaplastic carcinoma (1), mixed adenocarcinoma (1), and adenocarcinoma (4). The histopathologic diagnosis after 4 weeks treatment with the COX-2 inhibitor did not reveal a change in six of the tumors. One tumor previously diagnosed as an adenocarcinoma was diagnosed as a mammary adenoma with hyperplasia and granulomatous inflammation on the excisional biopsy. Immunohistochemistry for COX-2 was performed on all tumors; four tumors had increased COX-2 expression, one had stable COX-2 expression, and two tumors showed a slight increase in COX-2 expression.

These preliminary results are interesting, as a decrease in COX-2 expression was noted in the majority of dogs. These results warrant further evaluation of the role of COX-2 inhibitors in the treatment of canine malignant mammary tumors.

Development of quantitative real-time RT-PCR assay for detection of metastatic disease in canine melanoma. H von Euler1, S Asaad Maroud2, L Lenner1, B Kågedal1. 1 Department of Small Animal Clinical Sciences, Swedish University of Agricultural Sciences, Uppsala; 2 Division of Clinical Chemistry, Faculty of Health Sciences, Linköping University, Sweden.

Canine melanoma (CM) is the most common group of malignant tumors of the oral cavity and digits. Cutaneous CMs are usually benign, while oropharyngeal, uveal, and mucocutaneous neoplasms are often aggressive and have metastatic potential. Oral malignant CMs are characterized by rapid growth and local invasion. Metastatic rate is high, but metastases are not frequently observed until late during the course of the disease. As in humans, the prognosis is guarded if the diagnosis is late or if the cancer has disseminated. Correct staging and early discovery of residual disease is crucial for treatment plan and prognosis.

The aim of this study was to develop a quantitative real-time reverse transcriptase-PCR (RT-PCR) method for the detection of transcripts of canine melanoma associated antigens (MAAs) in primary tumors, regional metastases and cell cultures and to compare these techniques with similar analyses in man.

Biopsies from two canine oral melanomas with regional lymph node metastases were sampled at surgery and placed in RNA stabilization buffer. Specimens for primary culture were taken to cell medium and processed to cell lines. The protocol for human transcripts was used (Johansson et al. Melanoma Res 2000; 10:213–22). RT-PCR was performed using primers and probes for Melan-A and TRP-2. Analysis indicated that the inserted element was the reverse complement of a SINE insertion at this location in the gp100 sequence. A single amplicon of the expected size (792 bp) was generated from genomic DNA prepared from a control blood sample, whereas two amplicons (one of ~792 bp plus an additional larger amplicon) were detected from the melanoma sample. Cloning and sequencing of the larger amplicons yielded a product of 1034 bp. Sequence analysis indicated that the inserted element was the reverse complement of a SINE with short direct nucleotide repeats at each end, a poly-A repeat, multiple CT repeats and potential internal RNA polymerase III transcriptional control regions.

The RNA and protein sequences of the melanoma markers are frequently repeated and dispersed throughout the dog genome, approximately 5–8·3 Kb. There are ~360,000–600,000 copies in total, representing ~1·8–3% of the diploid dog genome. SINEs are thought to arise via transposition, where RNA polymerase III transcripts (rRNA and 5SrRNA) may become integrated into regions of the genome via an integrase enzyme. Staggered breakage and subsequent repair of the recipient chromosome results in flanking direct repeat sequences. In this tumour, the SINE was inserted within exon 11 of the gp100 gene. Quantiﬁcation of the studied markers in tumors and even metastatic disease in melanoma patients. Melan-A and gp100 expression analysis using real-time quantitative RT-PCR may provide a potential tool for better differentiation between varying degrees of malignancy in prostate carcinomas.

Identification of a short interspersed nuclear element (SINE) insertion in the gp100 gene of a poorly pigmented canine oral melanoma. G Dank1, A Aung2, S Yudelevitch1. 1 Koret School of Veterinary Medicine, Hebrew University, Rehovot, Israel; 2 Patho Vet Laboratories, Rehovot, Israel.

Glycoprotein (gp)100 plays a key role in melanin synthesis in melanocytes and melanoma tumors. Whilst investigating gp100 mRNA expression in canine malignant melanomas using RT-PCR, we identified an insertion mutation in the coding region of gp100 in a biopsy from a poorly pigmented oral melanoma. Analysis of the tumour genomic DNA revealed this to be a short interspersed nuclear element (SINE).

RNA was extracted from the tumour biopsy and cDNA synthesised. PCR was performed using primers designed to amplify the full-length coding region of canine gp100. The amplicon produced was cloned and sequenced, which revealed a single amplicon of the expected size (~792 bp) from genomic DNA prepared from a control blood sample, whereas two amplicons (one of ~792 bp plus an additional larger amplicon) were detected from the melanoma sample. Cloning and sequencing of the larger amplicon yielded a product of 1034 bp. Sequence analysis indicated that the inserted element was the reverse complement of a SINE with short direct nucleotide repeats at each end, a poly-A repeat, multiple CT repeats and potential internal RNA polymerase III transcriptional control regions.

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Long term remission of a primary malignant lymphoma of the urinary bladder in a dog—A case report. M Keseler1, R Lindel1, S Pflügerha1. 1 Tierärztliche Klinik für Kleintiere, Höhheim, Germany; 2Praxis für Tierpathologie Dr. D. v. Bomhard, München, Germany.

A 3 year old female spayed mixed breed dog was presented to Höhheim Animal Hospital with a 5 week history of hematuria and pollakisuria. A large infiltrative mass encompassing 2/3 of the bladder was identified by ultrasound. The bladder lumen was almost lost and the bladder wall thickness measured up to 1·8 cm with loss of its typical layered sonographic appearance. Mild distention of both ureters was present. Abdominal computed tomography, ultrasound, chest radiography and blood work revealed no other abnormalities. Full remission of the bladder biopsy was performed and was diagnosed as malignant lymphoma. On immunohistochemistry a CD79a positive and CD3 negative reaction was present which identified the mass as a B-cell lymphoma.

3 sessions of radiation therapy with 5 Gy each using a “Cobalt teletherapy unit” were administered to the urinary bladder with 1 fraction per week and resulted in complete remission of the tumor. No side effects of the radiation treatment were noted. The radiation was followed by a polychemotherapy protocol using four cycles of Vinristin, Asparaginase, Cyclophosphamide, Cytosin Arabinoside, and Doxorubicin. There were mild and self limiting gastrointestinal side effects following each Doxorubicin application.

Follow up examinations by ultrasonography and blood work were performed on a regular basis and revealed no recurrence of the neoplasia or any other signs.
of systemic spread. At present, the dog is in complete remission and free of symptoms for 24 months.

Primary malignant lymphoma of the urinary bladder is extremely rare in the dog and very few clinical data are available. In humans, less than 100 cases have been reported worldwide and most are B-cell lymphomas. Hematuria is reportedly the most common clinical symptom and the tumors usually respond well to therapy and have a favourable long term prognosis. It seems from this case that canine malignant lymphoma of the bladder resembles its human counterpart in both clinical symptoms and response to therapy.

**Diagnosis**

Primary PULMONARY HEMANGIOSARCOMA (HSA) in a GERMAN SHEPHERD DOG WITH SPONTANEOUS PNEUMOTHORAX (SP). L Tabar, F Garcia, R Rubanal*, Y Espara, X Roura, R Ruiz de Goyene, Veterinary Teaching Hospital, Autonomous University of Barcelona, Spain; *Veterinary Pathology Diagnostic Service, Veterinary Faculty, Autonomous University of Barcelona, Spain.

A 6-year-old male German Shepherd dog was referred for evaluation of dyspnea. The referring veterinarian noticed foamy, left, severe pleural effusion and slightly decreased heart sounds bilaterally. Thoracic radiographs showed multiple large, irregular, white, lucent lesions, consistent with pulmonary HSA. Other primary sites were excluded. The owners refused chemotherapy, but 5 months later, the dog showed no evidence of progressive disease. The referring veterinarian noticed fever, left shift neutrophilic leukocytosis and thrombocytopenia. Therapy prescribed included an antibiotic and an anti-inflammatory. Abnormalities from physical exam were congestive oral mucous membranes, rapid capillary refill time, laboured breathing, tachycardia and abdominal distention. Radiology and ultrasound showed bilateral pneumothorax with collapse of caudal pulmonary lobes, hepatomegaly and splenomegaly. Liver and spleen cytologic findings were consistent with systemic inflammatory response syndrome. Presumptive diagnosis was distrubutive shock and SP secondary to infection, unresponsive to neoplastic pulmonary disease. Conservative medical treatment initially instated was broad-spectrum antibiotic therapy, low molecular weight heparin and thoracic drainage via thoracostomy tube. Clinical condition slightly improved. Treatment initially instated was broad-spectrum antibiotherapy, low molecular weight heparin and thoracic drainage via thoracostomy tube. Clinical condition slightly improved. However, and despite incomplete surgical excision and no chemotherapy, the dog showed no evidence of progressive disease. The treatment initially instated was broad-spectrum antibiotherapy, low molecular weight heparin and thoracic drainage via thoracostomy tube. Clinical condition slightly improved.

In this case SIRS was also encountered. Sepsis due to secondary pneumonia could not be ruled out as blood cultures were not performed.

To the authors' knowledge there are no prognostic studies about primary pulmonary HSA. However, and despite incomplete surgical excision and no chemotherapy, survival time of this case is surprisingly above the ones reported for dogs with HSA.

**REFERENCES**


No information exists on the relative proportions, incidences or outcomes of diagnosis and treatment of feline cancer in South Africa, barring one survey of histopathology reports. Standard texts of veterinary oncology quote data from the Northern hemisphere, and geographic differences are apparent even within these figures.

In this retrospective analysis, the medical database of the Onderstepoort Veterinary Academic Hospital (OVAH) was analysed for details of feline cancer patients admitted for period 1998–2004 (n = 73 including 1 duplicate record for 2 different neoplasms on 1 patient out of N = 4274 feline admissions, or 1.71% of total feline admissions). Patients were categorised according to the method of referral (histopathology, cytology or presumed); tumour location and metastasis; survival (where known); whether or not any medical or surgical treatments were performed; and colouration (white/not white) or any medical or surgical treatments were performed and colouration (white/not white). Lymphoma was the second most common diagnosis (19%) followed by various carcinomas and adenocarcinomas (10% combined, excluding 3% various mammary tumors). Only one putative case of vaccine-associated sarcoma was recorded, and this was based on a cytologic diagnosis. A large proportion (55%) of patients received some form of treatment, but only 68% of neoplasms were confirmed by cytology or histopathology.

The average age of feline cancer patients was 9.5 years at presentation, and feline cancer represented 1.71% of the caseload at the OVAH. Squamous cell carcinoma, followed by lymphoma, form the majority of feline cancers diagnosed in South Africa (74% combined). SCC should reasonably form the major South African feline oncology research focus.


47 PATTERN RECOGNITION RECEPTOR mRNA EXPRESSION AND RESPONSE TO STIMULATION IN A CANINE MACROPHAGE CELL LINE. A House, SP Gregory, S Hutchinson, B Catchpole. Royal Veterinary College, University of London, London, UK.

In order to provide a first line of defence against invading organisms, the innate immune system has evolved to recognise conserved molecules (pathogen-associated molecular patterns—PAMPs) that are expressed by foreign organisms. This recognition process is facilitated by pattern recognition receptors (PRRs) which each recognise specific PAMPs. The predominant PRRs are Toll-like receptors (TLRs) or nucleotide-binding oligomerisation domain (NOD) proteins. Defects in PRR function can lead to disease (e.g. NOD2 in Crohn’s disease). The aim of this project was to characterise PRR expression and function in a canine macrophage cell line (DH82) as a model system for studying canine innate immunity.

DH82 cell expression of the PRRs; NOD1, NOD2, TLR1, TLR2, TLR4, TLR3, TLR6 and TLR9 mRNA was determined by RT-PCR using canine-specific primers. Cultured DH82 cells were exposed to various PAMPs including muramyl dipeptide (NOD2), peptidoglycan (TLR-2), bacterial lipopeptide PAM3CSK4 (TLR1/TLR2), lipopolysaccharide (TLR-4) and CpG DNA (TLR-9). The response to stimulation was determined by PCR using canine cytokine-specific primers for IL-1 beta, IL-6 and Tnf-alpha with GAPDH used as a housekeeping gene control. Tnf-alpha cytokine mRNA expression was quantified by performing real-time PCR (DyNaMo SYBR Green PCR Kit; Opticon 2 DNA engine, MJ Research). The Tnf-alpha concentration in the culture medium supernatants was measured using the L929 Tnf-alpha biosay.

DH82 cells expressed NOD1, NOD2, TLR1, TLR2, TLR4, TLR3 and TLR6 mRNA but TLR5 and TLR9 mRNA could not be detected. Cytokine mRNA expression was rapidly upregulated in DH82 cells following stimulation with peptidoglycan, PAM3CSK4 and LPS. Cells did not respond to CpG DNA, consistent with a lack of expression of TLR9. Real-time PCR demonstrated peak levels of Tnf-alpha mRNA expression after 2 hours of stimulation. Tnf-alpha protein concentrations were found to be highest in the cell supernatant after 4 hours of stimulation.

Characterisation of DH82 cell expression of PRRs and their response to spe-
cific PAMPs has proved to be a useful model for studying innate immune activity of canine macrophages. Future work will focus on adapting these techniques for use with blood-derived monocyte/macrophages isolated from dogs with suspected innate immune deficiency.

48 SEROLOGIC RESPONSES OF DOGS AFTER VACCINATION WITH A COMMERCIAL LEPTOSPIOSIS VACCINE. SC Barr, PL McDonough, R Scipioni-Balti, J Starr. ‘Clinical Sciences; Integrative Medicine and Diagnostic Sciences. College of Veterinary Medicine, Cornell University; ‘Marshall Farms, North Rose, NY, USA.

The microscopic agglutination test (MAT) is the standard specific serologic test of choice for diagnosing leptospirosis. Vaccines against leptospirosis are either be whole or subunit. The only vaccines available against current serovars causing most disease in dogs in the US (Leptospira interrogans serovars pomona and autumnalis, and Leptospira kirschneri serovar grippotyphosa) are subunit vaccines against pomona and grippotyphosa. Little is known about whether these vaccines cause elevation in MAT titers and if so, are these elevations serovar specific as has been reported for whole-cell vaccines against other serovars (canicola and icterohaemorrhagiae). Such vaccine-induced elevations in MAT titers often confuse veterinarians trying to diagnose active clinical disease based on MAT titers. The objective of this study was to measure MAT titers against serovars pomona, autumnalis, and grippotyphosa in puppies and mature dogs given a commercial vaccine against serovars pomona and grippotyphosa.

Forty 12-week-old beagle dogs were administered a commercial vaccine against serovars pomona, autumnalis, and grippotyphosa at 12 weeks of age, then boosted 3 weeks later. Twenty mature beagle dogs were administered the vaccine once. Serum MAT titers against serovars pomona, autumnalis, and grippotyphosa were measured pre-vaccination and at 2, 4, 6, 10, and 16 weeks after the first or only vaccination.

Of the 40 puppies vaccinated, 40 (100%), 0 (0%), and 40 (100%) developed MAT titers >100 after vaccination, to serovars pomona, grippotyphosa, and autumnalis, respectively. MAT titers against serovar autumnalis reached higher levels and persisted for some dogs for 16 weeks (6 weeks longer than for titers to serovar pomona). Of the 20 mature dogs, 13 (65%), 5 (25%), and 20 (100%) developed MAT titers >100 2 weeks after vaccination, to serovars pomona, grippotyphosa, and autumnalis, respectively. Titters against serovar pomona reached higher levels and persisted in some dogs beyond 16 weeks post-vaccination while those for serovar pomona and grippotyphosa persisted for 10 and 6 weeks, respectively. These results show that subunit vaccines against serovars pomona and grippotyphosa induce MAT titers not only to homologous antigens, but also to serovar autumnalis which could lead to a miss-diagnosis of leptospirosis due to serovar autumnalis. Further, a failure of puppies and most mature dogs to generate MAT titers to grippotyphosa in this study varies considerably from results published by the vaccine company.

49 PREVALENCE OF ANAPLASMA PHAGOCYTOPHILUM IN DOGS IN GERMANY. J Jensen, D Simon, H Murua Escobar, JT Soeller, J Bullerdiek, P Beelitz, E Pflister, J Nolte. ‘Small Animal Clinic, School of Veterinary Medicine Hannover, Germany; 2Center for Human Genetics, University Bremen, Germany; 3Institute of Comparative Tropical Medicine and Parasitology, Ludwig Maximilian University, Munich, Germany.

Anaplasma phagocytophilum is a gram-negative intracellular organism transmitted by ixodid ticks. The purpose of this study was to evaluate prevalence and significance of Anaplasma phagocytophilum infections in dogs in Germany.

A total number of 112 dogs were included in the study. 49 showed symptoms that could be attributed to Anaplasma phagocytophilum infection such as fever, lethargy, leucopenia, thrombocytopenia, reluctance to move, or shifting lameness (asymptomatic dogs). 63 dogs were without clinical signs (asymptomatic dogs). Exclusion criteria for both groups were a stay abroad and previous treatment with retroviral infections or pretreatment with immunosuppressive drugs. They were more frequent in certain areas (South, West) of Switzerland. A tendency of increased Mnov prevalence in spring could be noted. Sequencing of the 16S rRNA genes from several isolates were sequenced. Plasma samples were tested for FeLV and FIV infection by ELISA. The case histories and laboratory parameters of ill cats with suspected innate immune deficiency.

PCR results very high antibody titers were detected. There was no significant correlation of overall positives or antibody titers to age, breed, sex, or usage of the dogs as family or working dogs. Dogs with no or very low tick infestation were significantly less often infected with Anaplasma phagocytophilum than those with high tick infestation.

In conclusion there seems to be a high risk of infection with Anaplasma phagocytophilum in Germany. Results of this study suggest that severe illness solely caused by Anaplasma phagocytophilum is possible. Subclinical infection was seen frequently in this group of dogs.

50 PREVALENCE AND CLINICAL IMPORTANCE OF THE TWO KNOWN AND A THIRD NOVEL FELINE HAEMOPLASMA SPECIES IN CATS IN SWITZERLAND. B Baumgartner, T Tasker, B Wengen, M Meili, H Latte, CE Resch; R Hofmann-Lehmann. ‘Clinical Laboratory and ‘Clinic for Small Animal Internal Medicine, Vetsuisse Faculty, University of Zurich, Switzerland; ‘School of Clinical Veterinary Science, University of Bristol, Bristol, UK.

Haemobartonella felis, the causative agent of Feline Infectious Anaemia, has been reclassified within the group of haemotrophic Mycoplasma (aka haemoplasmas). Two different species have been recognized: Mycoplasma haemofelis (Mhf) and ‘Candidatus M. haemominutum’ (CMhm). Recently, we identified a third novel feline haemoplasma isolate (‘Mnov’) in a cat with severe anemia and thrombocytopenia. Experimental transmission of ‘Mnov’ resulted in anaemia in two SPF cats. Phylogenetic analyses of the 16S rRNA gene revealed close relationship of ‘Mnov’ to rodent haemoplasma species (Willi et al. 2005, JCM 43, in press).

The goal of the present study was to investigate the prevalence, clinical manifestation and risk factors for infections with the three feline haemoplasmas in representative Swiss cat populations. Blood samples from 586 ill and 86 healthy cats were collected over one year. DNA extracted from 200 μl of blood was analyzed using three newly designed specific TaqMan PCR assays. The 16S rRNA genes from several isolates were sequenced. Plasma samples were tested for FeLV and FIV infection by ELISA. The case histories and laboratory parameters of ill cats with suspected innate immune deficiency.

CfMhm infection was detected in 6.9% and 8.7%, and Mhf in 2.3% and 0.2% of healthy and ill cats, respectively. ‘Mnov was detected only in ill cats (1.1%); 4 out of 7 of these cats were co-infected with CFhm. CFhm infection was associated with male gender, outdoor access and old age. CFhm-infected ill cats had higher blood levels of BUN, creatinine, protein, and lipase, and were more frequently diagnosed with renal insufficiency, than CFhm-uninfected ill cats. Haematological parameters were not different in haemoplasma-infected compared to uninfected ill cats. Haemoplasma infections were not associated with either retroviral infections or pretreatment with immunosuppressive drugs. They were more frequent in certain areas (South, West) of Switzerland. A tendency of increased Mnov prevalence in spring could be noted. Sequencing of the 16S rRNA gene of representative Swiss isolates (n = 14) revealed >98% similarity with published sequences.

In conclusion, we have demonstrated all three feline haemoplasmas in Swit- zerland. CFhm was the most prevalent. Co-infection with CFhm and CFhm could be explained by a similar way of transmission. The association observed between CFhm infection and signs of renal insufficiency could be causal. However, it could also be accounted for by, e.g., the increased age of infected cats. Our study demonstrates the usefulness of the newly developed TaqMan assays in detecting infections with all three feline haemoplasmas.

51 REVIEW OF CLINICO-PATHOLOGICAL FINDINGS AND COAGULATION DISORDERS IN 45 CASES OF CANINE BABESIOSIS. R Ruiz de Gopegui, R Peñalba, LE Fidalgo, Y Espada, A Goucoa, I Espino. ‘Facultad de Veterinaria de Barcelona; ‘Facultad de Veterinaria de Lugo, HCV Rof-Codina, USC.

Canine babesiosis is a tick-borne disease caused by the hemoprotozoan parasite Babesia. The haematological and biochemical findings vary on geographic locations according to the virulence of parasite strains. The occurrence of hypergammaglobulinaemia and haemostatic disorders, particularly disseminated intravascular coagulation (DIC) are also reported, but the information related to those is highly unreliable.

B. canis canis in Spain. 45 dogs of both sex and of different breeds were examined during the period January 2003 to October 2004. The diagnosis of babesiosis was always confirmed by direct observation of the protozoa in a blood smear. All dogs were presented with a l to 3 day history of lethargy (40/45), fever (27/45) and mucosal membrane pallor (27/45). Hematemia (15/45) was also observed. The primary haematological abnormalities included a mild (PCV = 30.8 ± 11.1%) normochromic (MCV = 65.6 ± 3.6 fl), nonormochromic (35.3 ± 1.75 g/dl) anaemia and thrombocytopenia. All dogs (45/45) had the platelet counts below the lower reference range (175 104). Co-infection with B. gibsoni was also found in 2.3% of the cases. The CBC and coagulation profile of those dogs with high tick infestation. Some of the dogs had symptoms attributable to Anaplasma phagocytophilum infection than those with low or no antibody titers (≥1:64, n = 70). In all dogs with positive
evation in serum urea nitrogen (BUN) was observed in 21 of 45 dogs. Hae-
molysis, as it occurs in canine babesiosis, may cause non-renal elevations in
serum urea, possibly due to the ammonia loading of Hyperglycaemia (~120 mg/ dl)
was common (21/45) but was never severe. The highest blood glucose con-
centration was 221.6 mg/dL. We observed an increase in fibrinogen in 33 of 45
dogs (74%). DIC syndrome was diagnosed (based on PT, aPTT, TT, D-dimers, 
Fibrinogen level of 45 dogs (20%).

The clinico-pathological finding observed in 45/45 cases was a moderate 
thyrotoxicosis. In the absence of thyrotoxicosis, babesiosis is an unlikely
diagnosis. In a context of hypermetabolic illness such as babesiosis, hypergly-
caeemia was not a surprising finding. Hyperglycaemia in critical illness most often
is caused by increased glucose mobilization and stress, and can be markedly
increased by increased cortisol secretion. The results also indicated that B. canis
infection may impair haemostasis suggesting induction of DIC, and that
treat dogs in an early stage of infection might potentially avoid the possibility of
 developing and acute and uncompensated DIC.

HUMAN DANDER AS A POTENTIAL ALLERGEN SOURCE IN
ATOPIC DOGS—ALLERGEN CHARACTERIZATION AND IgE
PROFILING. N Resk1, A Hoffmann1, N Bauer1, A Moritz1. Small Animal
Practice 31, Rieker und Bohnenberger, Ravensburg, Germany; 
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Perennal indoor allergens are known to be important inducers of IgE-mediated
hypersensitivity reactions in dogs. Human dander (HD), an important component
of house dust, may also be a major cause of canine atopy with a reported prev-
ance of up to 68% (intradermal test data). As yet HD has been poorly char-
acterized as an allergenic source. The aim of this study was to differentiate
the clinical relevance of HD in dogs with atopic disease.

Several HD extracts were evaluated regarding their biological potency and
protein patterns using a recently developed mediator release assay (RBL-test)
and SDS-PAGE, respectively. Suitable extracts were included in a panel of com-
mon indoor (n = 8) and outdoor (n = 4) allergens to identify dogs with high
HD-specific IgE levels. Four groups of dogs were tested including “multi-
source” atopic dogs (n = 88), dogs with other skin diseases (n = 20), healthy
“household” (n = 30) and healthy “kennelled” dogs (n = 40) using a solid-
phase assay based on the ‘‘grid-blot’’ device. The reactions were graded semi-
quantitatively (0, 1, 2, 3) according to the concentration of allergen-specific serum
IgE.

Whereas grade 1 reactions were common in all groups of dogs, grade 3 re-
actions were not seen in any of the dogs tested and grade 2 reactions were also
an infrequent finding (‘‘multi-source’’ atopic dogs: 15%; dogs with other skin
diseases: 0%; healthy household dogs: 3%; healthy ‘‘kennelled’’ dogs: 10%).
Only sera with high HD-specific IgE levels (grade 2) were selected as probes to
identify IgE-binding proteins by western blot. Six sera from the ‘‘multi-source’’
atopic dogs demonstrated IgE-binding to a protein with an apparent molecular
weight of 11 kDa identified as human cystatin A by N-terminal microsequencing.

Human cystatin A is a cysteine protease inhibitor belonging to the superfamily
of cystatins. These include the recently identified allergen Fel d 3, an IgE reactive
protein for house dust mites which supports the assumption that human cystatin A
may be of relevance for the development of canine atopy. Considering the so-
far reported high prevalence of HD-related atopy detected in intradermal tests,
the low sensitization rate of dogs to HD in our study was remarkable. This may
be due to the presence of cell-bound IgE, which is only detectable in an intra-
dermal test but not in in-vitro assay.

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First European Report of B. gibsoni (Asian Genotype) Infection in Two American Pit Bull Terriers (APBT) without travel history, were published in USA 1999 (Bir-

In 2002 and 2004 we diagnosed Babesiosis in two APBT, without travel his-
tory to endemic areas. Both dogs had a severe haemolytic anaemia, thrombo-
cytopenia, leukocytosis, hyperproteinemia and haemoglobinuria. Small intra-
erythrocytic piroplasms were seen on blood smears. The final diagnosis was
made by PCR, amplifying a partial region of the 18S rDNA gene and
sequencing. The sequenced 480bp PCR-products were 100% identical to Babesia gibsoni (Asian genotype) sequence in GenBank (accession numbers AF175300

The other four APBTs of the same owners were PCR negative.

One dog was treated with Imidocarb, Doxycycline and Phenamidine and was
subsequently PCR negative on three occasions over a period of two years. The
other dog was treated with Imidocarb, Doxycycline and Buparvaquone and
while all clinical parameter improved markedly, PCR remained positive.

The source of infection in these two dogs remains unknown.
Overall, lymphoid tissue was predominantly found in caudal areas (A and B) and it was more developed in puppies than in adult dogs. These areas contained lymph follicles (LF), diffuse aggregates of lymphocytes (DA) and scattered lymphoid cells (SC) in the lamina propria (LP) of the epithelium. In contrast, only small DA and SC were found in the LP of areas C and D. Mast cells were mostly found immediately beneath the epithelium but they were also scattered deeper in the LP. MHC class II+ cells were mostly localized within LF and DA. These cells had the morphology of macrophages, B cells or dendritic cells. MHCII+ cells with a dendritic morphology were also present within and immediately beneath the basement membrane of the respiratory epithelium. The number of MHCII+ cells was higher in puppies than in adult dogs. Polymorphonuclear L1+ cells were found in very low numbers throughout the LP. CD3+ cells represented the vast majority of the lymphocytic cells. There were significantly more CD3+ cells in areas A and B in puppies than in adult dogs. IgA+ and IgM+ plasma cells were mostly associated with the glandular tissue but were also scattered in LF and DA or immediately beneath the respiratory epithelium. IgG+ cells were sparse. Overall, the number of plasma cells was higher in adult dogs than in puppies.

This study has demonstrated the presence of organized lymphoid tissue in the canine nasopharynx. The immunohistochemical findings of the present study will enable comparisons to be made with similar studies conducted in dogs suffering from nasopharyngeal diseases or in dogs intra-nasally vaccinated.

**FREQUENCIES OF FELINE BLOOD TYPES IN CATS AT THE ROYAL VETERINARY COLLEGE, LONDON, UK: Y Forcada1, G Gibson1, 1Royal Veterinary College, London, UK.**

There have been three different feline blood types described: A, B, and AB. The importance of blood typing in cats before a transfusion cannot be overemphasised. The presence of naturally occurring pre-formed alloantibodies in cats requires typing of the donor and recipient blood to prevent a potentially fatal transfusion reaction. The prevalence of different blood types is known to have geographical and breed variation. Various breeds are known to show a higher prevalence of type B (i.e. British Shorthair cats); others type A (Siamese 100% type A); whereas the majority of non-pedigree cats are known to be predominantly type A. Previous reports have suggested the prevalence of blood type B in non-pedigree cats in the north of the UK and Scotland to be 7.9%. The highest percentage of type B non-pedigree cats in Europe was found in France with a prevalence of 14.9%. It was our aim to establish the prevalence of blood types in our hospital population and to compare the frequencies of blood types between pedigree and non-pedigree cats.

Medical records of all cats blood typed in the Queen Mother Hospital for Animals between January 2000 and November 2004 were reviewed. Blood typing had been performed using commercial feline blood typing cards (previously validated for feline blood typing) and saline agglutination test. Cats showing autoagglutination were eliminated from the study.

156 cats were included in the study. Four cats were not included due to saline autoagglutination of red blood cells, which prevented accurate determination of blood groups. A and B cats were predominantly classed as non-pedigree. Of the 51 pedigree cats, the prevalence of blood types was as follows: Type A, n = 42 (82.4%); Type B, 7 (13.7%); Type AB, 2 (3.9%). Of the 105 non-pedigree cats, the prevalence of blood types was as follows: Type A, n = 71 (67.6%); Type B, 32 (30.5%); Type AB, 2 (1.9%).

The results of this study have demonstrated that the prevalence of blood type B in non-pedigree cats is much higher than in other previous studies in the UK. This apparent increased prevalence of blood type B in non-pedigree cats is likely a reflection of geographic variation of feline blood types in the United Kingdom. The clinical implication of this finding is that the increased percentage of type B cats in the non-pedigree cat population increases the risk of transfusion incompatibility if donor and recipient cats are not appropriately blood typed prior to administration of blood products.

**EFFECTIVITY AND SAFETY OF DIFFERENT DOSAGES OF LOW MOLECULAR WEIGHT HEPARIN IN DOGS SUFFERING FROM GASTRIC VOLVULUS/DILATION COMPLEX: R Mischke, C West. Small Animal Clinic, University Hannover, Germany.**

The objective of this study was to evaluate the effectiveness and safety of low molecular weight heparin (LMWH) in dogs suffering from gastric volvulus/dilation syndrome (GVDS), a disease which is often associated with disseminated intravascular coagulation (DIC).

The study included 60 dogs suffering from GVDS. 19 dogs served as a control group (without anticoagulatory treatment). In the remaining groups, treatment with subcutaneous injections of dalteparin-sodium every 8 hours for 7 days started after surgery. The individual dosage consisted of 75 anti-FXaU/kg BM (n = 21, group 2), 100 anti-FXaU/kg BM (n = 14, group 3), and 150 anti-FXaU/kg BM (n = 25, group 4). At defined times, clinical examination and measurements of heparin plasma activity, platelet count, prothrombin time, activated partial thromboplastin time (APTT), factor V activity, antithrombin activity, soluble fibrin as well as a haematological and biochemical profile were performed.

All dogs of group 4 developed severe bleeding complications (mainly in the area of the surgical wound). Due to this fact, only 6 animals were treated with 150 anti-FXaU/kg BM 3 times daily (group 4) and a modified protocol with 100 anti-FXaU/kg BM 3 times daily was added to the study design (group 3). 6 of 14 dogs of group 3, 3 of 21 dogs of group 2 and none of the control dogs showed significant bleeding episodes. The mean plasma heparin activity in groups 2, 3, and 4 reached maximum values of 0.430.17 anti-FXaU/ml, 0.60 ± 0.21 anti-FXaU/ml and 1.01 ± 0.39 anti-FXaU/ml.

Other clinical parameters and measurements of soluble fibrin, factor V activity and prothrombin time did not show significant differences between groups. APTT values were significantly higher in dogs of group 4 when compared to all other groups. In addition, a dose-dependent decrease of antithrombin activity was observed.

The results of this study indicate that LMWH does not have a positive influence on the clinical outcome and consumption coagulopathy in dogs suffering from GVDS. Prolongation of APTT and decrease of antithrombin in dogs treated with LMWH might be due to heparin dosage. Other LMWH dosages were not investigated.

Cats suffering from gastrointestinal symptoms are often treated with LMWH. However, LMWH dosage has not been found in the literature. This study has demonstrated the presence of organized lymphoid tissue in the canine nasopharynx. The immunohistochemical findings of the present study will enable comparisons to be made with similar studies conducted in dogs suffering from nasopharyngeal diseases or in dogs intra-nasally vaccinated.

**QUANTIFICATION OF mRNA ENCODING CYTOKINES AND CHEMOKINES IN NASAL BIOPSIES FROM DOGS WITH SINOS-AS-NASAL ASPERGILLOSIS: D Peeters, IR Peters1, C Clercx, MJ Day1, University of Liege, Belgium; University of Bristol, UK.**

Cats suffering from sino-nasal aspergillosis are characterised by localised invasion of the nasal cavity and frontal sinus and is mostly caused by Aspergillus fumigatus. The pathogenesis of the condition is poorly understood but this disease occurs in otherwise healthy and apparently immunocompetent individuals. In the present study, we have used quantitative RT-PCR to investigate the nature of the local immune response mounted in the upper respiratory mucosa of affected dogs.

Quantitative RT-PCR was carried out on RNA isolated from nasal biopsies from diseased and control dogs, using specific assays designed to amplify mRNA encoding a panel of cytokines (IL-4, IL-5, IL-6, IL-10, IL-12p40, IL-13, IL-18, IFN-γ, TNF-α and TGF-β) and chemokines (IL-8, MCP-1, -2, -3 and -4, and eotaxin-2 and -3). For each molecule, relative copy numbers obtained in nasal tissue from control dogs and dogs with sino-nasal aspergillosis were compared to those of the Wilcoxton-two-samples test using the NPAR1WAY procedure (PROC GLM, SAS Institute, Cary, NC). Cats suffering from sino-nasal aspergillosis were associated with significantly increased expression of mRNA encoding MCP-1, -2, -3 and -4, IL-8, IL-10, IL-18 and TNF-α relative to controls (P < 0.01) but there was no difference between groups with respect to IL-4, IL-5, IL-6, IL-12, TGF-β, and eotaxin-2 and -3. Samples from individual diseased dogs showed elevation of IFN-γ transcript above the upper range of normal, but overall there was no significant difference between diseased and control tissues.

Our results suggest that proinflammatory cytokines (IL-18 and TNF-α and chemokines in phagocytic cells (MCP-1, -2, -3 and -4) might account for the restricted localisation of the infection to the upper respiratory tract in dogs. IL-12 and IFN-γ are known to be essential for clearing fungal infections in mammals, so the lack of up-regulation of mRNA encoding Th1 cytokines (IL-12 and IFN-γ) in diseased tissues might account for the fact that infected dogs are generally unable to clear Aspergillus spontaneously. Failure to express mRNA encoding Th1 cytokines in nasal tissue from dogs with sino-nasal aspergillosis might be due to the up-regulation of mRNA encoding the immunomodulatory cytokine IL-10. IL-10 production and regulatory T cells might also be important in limiting the extent of local tissue destruction.

This study was funded by the ECVIM Clinical Studies Fund 2002.
leishmaniosi at diagnosis and over a year follow-up treatment. Diagnosis of canine leishmaniosi was made by serum titers of anti-Leishmania antibodies (ELISA) and identification of the parasite either by visualization in bone marrow cytology, skin biopsy or detection of DNA by polymerase chain reaction technique in dogs with compatible clinical signs and laboratory abnormalities. In all cases general biochemistry analyses and complete urinalysis (including urine protein/creatinine ratio and microalbuminuria detection test) were performed. Because the measurement of microalbuminuria was qualitative in nature, logistic regression was used to evaluate the results.

The prevalence of microalbuminuria in dogs with patent leishmaniosi at diagnosis was 73.8%. In these dogs, the prevalence of the different levels of microalbuminuria was 6.6% (one +), 13.1% (two +) and 54.3% (three +). The prevalence of microalbuminuria in dogs in follow-up during anti-Leishmania treatment was 66.7% (at the diagnosis), 55.6% (one month later), 76.5% (six months later) and 86.6% (one year later). Urine protein/creatinine ratio was not statistically significantly different between dogs with and without microalbuminuria at six and twelve months of follow-up.

Based on these results, it is clear that the likelihood of microalbuminuria is high in dogs with patent leishmaniosi and, although the prevalence of microalbuminuria decreases during the anti-Leishmania treatment, it is still higher than 50% during all the follow-up. It also seems reasonable to conclude that microalbuminuria detection could be a good test to evaluate renal involvement in dogs with leishmaniosi at the diagnosis and during the follow-up. Further is required to determine if increased prevalence will decrease with the addition of specific therapy for proteinuria to the anti-Leishmania treatment.

60 CLINICAL PRESENTATION AND LONG-TERM OUTCOME IN 21 CATS WITH THIRD DEGREE ATRIOVENTRICULAR BLOCK (1997-2004). KB Bellum, RL Stepien. Department of Medical Sciences, University of Wisconsin School of Veterinary Medicine, Madison, Wisconsin, USA.

The impact of third degree atrioventricular block (3AVB) on clinical findings and long-term outcome in cats is unknown. Presentation, clinical findings and clinical outcome of 21 cats with 3AVB were studied retrospectively. Median age of cats studied was 14 years (range 7-19 years). Typical presenting signs included respiratory distress or collapse, but 29% were presented with no abnormal clinical signs. Most cats had concurrent systemic diseases common in this age group, including diabetes mellitus, hyperthyroidism and renal disease. Eight cats (38%) had congestive heart failure (CHF) at the time of diagnosis. Heart rates overall ranged from 80-140 bpm (median 120 bpm) and there was no difference in heart rate between cats with CHF and those without CHF. Eleven of 18 (61%) cats had cardiac lesions consistent with their systemic disease and 1 cat had no abnormalities noted. No atrioventricular nodal lesions were noted by echocardiography. One cat had histologic atrioventricular nodal lesions. Median survival of 14 cats that died or were euthanized was 386 days (range 1-2013 days). Survival did not differ between cats with or without CHF or between cats with or without structural cardiac disease (P < 0.05). One cat underwent successful pacemaker implantation. Third degree heart block in cats is often not immediately life-threatening. Survival in these 21 cats was not affected by the presence of underlying heart disease or congestive heart failure at the time of presentation. Even cats with overt clinical signs of collapse may survive >1 year without pacemaker implantation.

61 PREVALENCE OF ENTERAL VIRUSES IN 936 DOGS WITH ACUTE HAEMORRHAGIC DIARRHEA. B Schulz1, C Strauch1, U Trayn1, K Hartmann1,2. Medizinische Kleinr Tierklinik der Ludwig-Maximilians-Universität München, Munich, Germany;1 Institut für Tierhygiene und Öffentliches Veterinärwesen, Leipzig, Germany.

Acute haemorrhagic diarrhea is one of the most common reasons for presentation of dogs in veterinary practice. Among the viruses suspected to be involved in naturally occurring diarrhea in dogs are paroviruses, coronaviruses, para- myxoviruses, and rotaviruses. Not much is known about the prevalence of these viruses in dogs with diarrhea. In the field as well as concerning differences in clinical and laboratory signs and outcome associated with different viral agents. Moreover, the pathogenicity of some enteral viruses is matter of discussion, since clinically healthy dogs may be asymptomatic carriers of these viruses. Aim of this retrospective study was to evaluate prevalence and difference of intestinal viruses in dogs with acute bloody diarrhea and to compare signalment, clinical signs, and laboratory abnormalities among groups of dogs infected with different viruses to those that tested virus-negative.

In a retrospective study, using 936 client-owned dogs were included that were presented for acute bloody diarrhea to the Veterinary Teaching Hospital of the Munich University over a period of 11 years. Virus detection was performed by electron microscopy from freshly collected fecal samples in each of these patients. Signalment, clinical signs, and laboratory abnormalities among different virus groups were evaluated statistically and compared to the parameters of the negative dogs.

Viruses was detected in 55.2% of the dogs presented with acute bloody diarrhea. The highest prevalence was demonstrated for paroviruses (16.6%), followed by coronavirus (11.9%), and paramyxoviruses (9.3%). In none of the fecal samples, rotavirus was detected. Two or three virus species were demonstrated in 6.5% of all fecal samples. Dogs with parovirus infection were statistically significantly younger when compared to dogs infected with other enteral viruses or dogs that tested negative. No significant differences were demonstrated concerning sex, breed, or clinical parameters among groups. Parovirus-infected patients showed significantly lower leukocyte and erythrocyte counts as well as hematocrit and total protein and albumin levels compared to all other groups.

Paroviruses still seem to be the most prevalent viral agents involved in acute diarrhea in dogs in Germany. Besides the young age, parovirus infection is strongly associated with typical changes in laboratory parameters. Rotaviruses were not identified in dogs with haemorrhagic diarrhea. Coronavirus and paramyxoviruses were present in these cases of haemorrhagic enteritis or their potential role as secondary invaders, however, still has to be clarified.

62 REPRODUCIBILITY OF ENDOSCOPIC COLLECTION OF DUODENAL JUICE AND EVALUATION OF SAMPLE STABILITY FOR 16S rDNA ANALYSIS OF THE DUODENAL MICROFLORA IN DOGS. JS Suchodolski1, CG Ruax1, RL Stepien1, L Granaly2, T Egelund3, DA Williams4. Gastrointestinal Laboratory, Department of Small Animal Clinical Sciences, Texas A&M University, College Station, TX, USA;2 Karlslunde Dyreklinik, Karlslunde, Denmark.

It is now recognized that many bacteria escape identification using standard bacterial culture. 16S rDNA analysis has revealed previously uncharacterized bacteria present in fecal samples. The aim of this study was to evaluate the reproducibility of endoscopic collection of duodenal juice, to evaluate sample stability, and to describe the canine duodenal microflora in healthy dogs by direct 16S rDNA analysis.

Duodenal juice was collected endoscopically in duplicate using sterile disposable cytology brushes from 5 dogs. Duodenal juice was also collected by needle aspiration from healthy dogs, euthanized for unrelated studies. Samples were divided into 4 aliquots: 3 aliquots were stored in liquid nitrogen, at −80°C, and −20°C, respectively. The fourth aliquot was stored at 4°C for 72 hours, aiming to simulate storage conditions for overnight shipment of cooled samples. Each sample was processed independently. Bacterial DNA was extracted and 16S rDNA amplified. The reproducibility between duplicates collected endoscopically (method reproducibility) and aliquots stored (sample stability) was determined by comparing the similarity of their banding patterns following denaturing gradient gel electrophoresis (Dice coefficient, 100% = complete identity). Bacteria present in duodenal juice from healthy dogs were identified by sequencing of cloned 16S rDNA amplicons.

The mean (±SD) similarity between duplicate samples collected endoscopically was 93.7% (±3.0). Mean (±SD) similarity between the different storage conditions from each dog were: 92.2% (±5.5), 93.0% (±7.9), 91.1% (±3.1), 94.2% (±5.2), 91.7% (±4.5) and 90.0% (±8.1), with no significant difference between different storage conditions (ANOVA, P = 0.893). Sequencing data revealed 47 individual 16S rDNA sequences, 20 (42.6%) were lower than 98% sequence similarity to 16S rDNA sequences of previously described microorganisms as identified in public databases (GenBank, Ribosomal Database Project), suggesting the presence of previously uncharacterized bacterial species in the canine duodenum. The majority of sequences identified belonged to the orders of Clostridiales (40%), Lactobacillales (21%), and Enterobacteriales (19%).

The canine duodenum harbors a complex microflora. Endoscopic collection of duodenal juice using a disposable cytology brush is a rapid and reproducible sampling technique and the specimens obtained show high storage stability. Direct 16S rDNA analysis revealed previously uncharacterized bacterial species in the duodenum of healthy dogs.

63 DETERMINATION OF INTESTINAL PERMEABILITY AND MUCOSAL ABSORPTIVE CAPACITY IN NORWEGIAN LUNDEHUNDS USING A FOUR-SUGAR BLOOD TEST. N Berghoff, CG Ruaxx, JL Steinert, A Stoll, B Shah, H Rodriguez, and DA Williams. Gastrointestinal Laboratory, Texas A&M University, College Station, TX, USA.

Many Norwegian Lundehunds are affected by gastrointestinal disease. Clinical signs may include diarrhea, vomiting, anorexia, weight loss, and in many cases gastrointestinal protein loss. The aim of this study was to evaluate intestinal permeability and absorptive capacity in Norwegian Lundehunds in the USA and Canada, in order to further characterize these intestinal abnormalities.

A four-sugar blood test was performed in 11 Norwegian Lundehunds (mean age 3.4 years; range 0.5-7.5 years), none were exhibiting clinical signs of gas-
trientestinal disease at the time of the test, and in eight healthy control dogs of various breeds (mean age 4.6 years; range 1.5–8.0 years). Food was withheld from the dogs for a period of 15 hours before sampling was obtained from each dog immediately prior to oral administration of the sugar solution. The sugar solution contained 5.0 g/L methylglucose, 10.0 g/L rhamnose, 10.0 g/L xylose, and 10.0 g/L lactulose. Dogs of less than 10 kg body weight received 100 ml, dogs weighing 10–20 kg received 200 ml, dogs weighing more than 20 kg received 400 ml of the solution. Subsequent blood samples were collected at 60, 90, and 120 minutes after sugar administration. Serum concentrations of the four sugars were determined by HPLC with pulsed amperometric detection. Serum concentrations of all sugars at all time points were analyzed in control dogs and Norwegian Lundehunds using the Friedman test. Median ratios of serum lactulose to rhamnose concentrations (L/R ratio) and xylose to methylglucose concentrations (X/M ratio) were compared between control dogs and Norwegian Lundehunds at the 90 minutes post-dosing (Mann Whitney test).

Oral administration of the sugar solution lead to significant changes in serum concentrations of all sugars at all time points compared to baseline (p ≤ 0.0015 for all). Five healthy laboratory Beagle dogs with a permanent jejunal fistula located 60 cm distally from the pylorus were included into this study. Food was withheld for 5 days, but normal tap water was given ad libitum. Jejunal juice samples were collected daily for 5 days via the fistula using a sterile cytology brush before, during and after food deprivation. Thorough clinical examination was performed and serum biochemistry and hematological values were evaluated daily during food deprivation. Bacterial DNA was purified, the variable V6-V8 region of 16S rDNA was amplified with universal bacterial primers, and PCR amplicons were subsequently separated by denaturing gradient gel electrophoresis (DGGE). Variation in the jejunal microbiota before, during and after food deprivation was evaluated by comparing similarity indices (Dice coefficient; 100% represents complete identity) of DGGE profiles using gel analysis software. Friedman two-way analysis of variance followed by multiple comparison test was used to test for the differences in the similarity percentages between the three phases.

No clinical, biochemical or hematological abnormalities were observed over the duration of the study. Mean (±SD) similarity indices of DGGE profiles before, during and after food deprivation were 76.9 (±3.1), 57.0 (±7.6), and 64.1 (±4.6), respectively. Food deprivation led to significant changes (p < 0.05) in DGGE profiles when compared to profiles before the food deprivation period. However, the jejunal microbiota remained relatively stable during the food deprivation period, and significant daily changes were not observed. After food deprivation, jejunal microbiota returned close to the baseline determined before food deprivation.

This is the first study to show that there is a complex bacterial population in the canine jejenum during food deprivation. Also, the jejunal microbiota before, during and after food deprivation. While the composition of the small intestinal microbiota is reported to have a complex bacterial population in a group of healthy control dogs. High L/R ratios suggest an alteration in small intestinal permeability, possibly reflecting disturbed tight junctions and decreased intestinal surface area. The increased X/M ratios seen in this group of Norwegian Lundehunds were predominantly due to lower serum methylglucose concentrations, suggesting decreased mucosal absorptive capacity for this analyte.

DYNAMICS OF JEJUNAL MICROBIOTA DURING FOOD DEPRIVATION. JA Harmoinen1, JS Suchodolski2, CG Ruau2, JM Steiner3, E Westermarck4, DA Williams1. 1Department of Clinical Veterinary Sciences, Helsinki University, Helsinki, Finland; 2Gastrointestinal Laboratory, Department of Small Animal Clinical Sciences, Texas A&M University, College Station, TX, USA.

While the composition of the small intestinal microbiota is reported to have a significant impact on the health status of an animal, and food deprivation is a commonly recommended remedy in acute gastrointestinal disturbances in dogs, no data are available about the dynamics of the canine small intestinal microbiota during an undated fast. The aim of this study was to evaluate the dynamics of the jejunal microbiota before, during and after food deprivation.

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Dynamics of the Jejunal Microbiota during Food Deprivation. J.H. Harmoinen1, J.S. Suchodolski2, C.G. Ruau2, J.M. Steiner3, E. Westermarck4, D.A. Williams1. 1Department of Clinical Veterinary Sciences, Helsinki University, Helsinki, Finland; 2Gastrointestinal Laboratory, Department of Small Animal Clinical Sciences, Texas A&M University, College Station, TX, USA.

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56 REAL TIME-PCR QUANTIFICATION AND GENETIC IDENTIFICATION OF HELICOBACTER STRAINS IN A GROUP OF DOGS WITH DIGESTIVE DISORDERS. O. Dossin1, J. Coillard2, C. Bouscraut-Baralon3, ‘Internal Medicine’, National Veterinary School, Toulouse, France; 2Scanelis, Toulouse, France.

Helicobacter spp. are frequently found in the dog stomach but the association between gastritis and Helicobacter infection is not fully understood. While infection with Helicobacter spp. is a feature of gastritis and Helicobacter spp infection was assessed by silver staining, urease test and PCR (abstract in JVM 2003, 17:447). The aims of this further study were 1. quantification of Helicobacter infection with Real Time-PCR and assessment of its relationship with microscopic quantification and gastritis, 2. characterization of Helicobacter species infecting this group of dogs.

Thirty-six dogs with either chronic vomiting (n = 14), diarrhea (n = 21) or no clinical signs (n = 6) were included in the study. Gastric endoscopic biopsies were performed in all dogs. The gastritis was histologically scored from absence to mild, moderate or severe. Helicobacter spp. infection was assessed by urease test, Warthin Starry staining and Helicobacter specific Real Time-PCR of 16S RNA gene on specimens from 3 gastric parts (fundus, body and antrum). Cloning and sequencing of ampiclon of the urease gene were performed to further characterize Helicobacter species as compared with reference sequences from GenBank Database.

All dogs were infected with Helicobacter spp. The Real Time-PCR quantification was significantly lower in antrum when compared with fundus or body. There was no correlation between silver stain and Real Time-PCR quantification and no relationship between PCR quantification and histological gastritis scoring. As revealed by sequencing analysis performed in 31/36 dogs, 23/31 dogs were infected with more than one Helicobacter spp. strain. The prevalence of the identified Helicobacter spp. strain was H. heilmannii in 25/31 dogs, H. helicobacter in 18/31 dogs and H. salomonis in 13/31 dogs. Helicobacter pylori was identified in one single dog. In 19/31 dogs, the sequences were not matching with the Helicobacter spp. sequences available on GenBank (maximal homology of 83% with H. felis, h. cincorhini or h. heilmannii) but were clearly Helicobacter spp. as shown by 16S RNA. These unknown sequences are relatively homogenous with phylogenetic analysis. Specific sequence distribution was observed for the different Helicobacter strains but H. pylori was found only in the body and H. salomonis only in the fundus and body. No definitive relationship was observed between Helicobacter species infection and gastritis.

This study did not show any relationship between Helicobacter spp. infection and gastritis in dogs. Moreover, none of the Helicobacter species characterized has been specifically related to gastric inflammation in any part of the stomach in our group of dogs.

57 NO CHANGES IN HISTOLOGICAL SCORING, TOTAL NUMBER OF INFILTRATING CELLS AND NUMBER OF T CELLS AFTER TREATMENT IN DOGS WITH CHRONIC ENTEROPAT- HIES. N. Schreiner1, K. Allenbach1, S. Sauteur2, A. Gorne2, F. Gaschen1. 1Department of Clinical Veterinary Medicine, Vetsuisse Faculty, University of Bern, Switzerland; 2Institute of Veterinary Pathology, Faculty of Veterinary Medicine, University of Hannover, Germany.

Histology remains the mainstay of diagnosis in dogs with chronic enteropathies (CE). In human beings with inflammatory bowel disease (IBD), histology has been shown to be useful for assessing clinical remission and correlates well with clinical activity of disease. In dogs with CE, the main infiltrate in the lamina propria consists of plasma cells and lymphocytes. T cells and production of proinflammatory cytokines have been implicated in the pathogenesis of the canine disease. However, no studies have been undertaken so far to show if histological
The prevalence of chronic pancreatitis (CP) in dogs is unknown. Previous studies have focussed on acute pancreatitis and/or have used a highly biased and selected population of second opinion and critical care cases. This study aimed to assess the prevalence of chronic pancreatitis in an unselected population of first opinion dogs.

Sections were obtained from 100 consecutive canine post mortem presented to Glasgow Veterinary School from surrounding first opinion practices. Most dogs were assessed as middle-aged to old. In each case, 3 sections of pancreas were taken: one from each limb and one from the body. Sections were preserved in formalin and stained with H&E and Sirius red. They were examined histologically, blind to signalment. Clinical details were not available. The dogs were grouped according to histological findings: (a) sections too autolysed to interpret; (b) no abnormalities visible; (c) non-specific insignificant changes; (d) chronic or acute-on-acute chronic pancreatitis; (e) acute pancreatitis with no chronic changes and (f) other disease. Prevalence of each group was calculated and relative risk of CP and autolysis were calculated for different breeds.

Prevalence of autolysis was 27% and a shortcoming of this type of study. Autolysis was common in large breed dogs due to slow cooling of core temperature (e.g. German shepherd dogs relative risk of autolysis 3.8) so relative risk of CP could not be calculated in large breeds. The pancreas had no histological abnormalities in only 13% of dogs. Non specific changes were observed in 24% of dogs. Acute pancreatitis had a low prevalence of 2%. The prevalence of CP was 29% and breeds with a high relative risk included Cavalier King Charles spains (CKCS): relative risk 4.1 and Jack Russell terriers: relative risk 2.4. 6/6 CKCS had CP, with 1/6 having end stage disease and 1/6 having concurrent pancreatic neoplasia. The lesions observed are likely to correlate with clinical disease: one out of four published cases of end stage CP with exocrine pancreatic insufficiency (EPI) was a CKCS and a further 27 biopsy-confirmed cases of chronic pancreatitis seen at the Queen’s Veterinary School Hospital since 2002 were CKCS, both of which had EPI and one of which had diabetes mellitus. We conclude that CP is common in the first opinion dog population and in the liver, end stage disease can be considered as a distinct clinically significant entity. There are strong breed-associations in CKCS and JRT, suggesting a possible genetic basis to the disease in these breeds.

COIL-EMBOLIZATION OF EXTRAREGIONAL PORTOSYSTEMIC SHUNT IN DOGS. M. Schneider, S. Scheit, M. Plassmann, Small Animal Clinic (Internal Medicine), Justus-Liebig-University, Giessen, Germany.

The coil embolization of extrahepatic shunts showed a relatively low survival rate (47/71 patients) and one of the four surviving dogs developed acquired extrahepatic shunts (Levyville et al., 2003). Up to now no study proved the combination of coil embolization with antithrombotic treatment in dogs with a congenital extrahepatic portosystemic shunt.

Between July 2000 and October 2003 we diagnosed a congenital extrahepatic portosystemic shunt in 22 dogs. 3 dogs were managed only with dietary treat-
71 DETECTION OF FELINE AUTOSOMAL-DOMINANT POLYCYSTIC KIDNEY DISEASE (AD-PKD) BY REAL-TIME PCR ASSAYS. CR Helps*, S Tasker, S Wills, LJ Lyons1, TJ Gruffydd-Jones1. †School of Clinical Veterinary Science, University of Bristol, Bristol, UK. ‡School of Veterinary Medicine, University of California, Davis, USA.

AD-PKD is the most prevalent inherited genetic disease of cats, particularly affecting Persians. Until now AD-PKD testing in the USA has been hindered by limited diagnostic tools for haemoplasma infection since they allow development of a single nucleotide polymorphism (SNP) in PKD 1 results in the production of an abnormal truncated protein. A conventional polymerase chain reaction (PCR) and restriction endonuclease analysis (REPA) assay were previously developed for the detection of ADV-PKD ultrasound screening at the University of Bristol were obtained. DNA was extracted from 1001 of blood using a Nucleospin Blood kit (Macherey-Nagel) and subjected to real-time PCR and the conventional PCR-RFLP assay previously described (the latter was run both at the Universities of Bristol and California, Davis). Of the 72 UK cats tested, 29 were positive for the PKD 1 SNP (all heterozygous) and 43 were negative (all homozygous wild type); no homozygous mutant PKD cats were identified. All 72 samples showed 100% agreement between the conventional PCR-RFLP assay previously described. All 29 cats which generated positive AD-PKD PCR results were positive for PKD 1 on ultrasonography. Of the 45 cats which generated negative AD-PKD PCR results, 41 were negative and 2 were equivocal by ultrasonography.

AD-PKD in Persians and Exotic Shorthair cats in the UK appears to be caused by the same SNP in PKD 1 identified in the USA. PCR detection of AD-PKD negates the need for ultrasonography for definitive diagnosis and relies on easily performed blood sampling that can be done in cats less than 10 months old. Additionally, compared to the conventional PCR-RFLP assay, real-time PCR genotyping is quicker (90 minutes vs. 5 hours), less labour intensive and significantly reduces the risk of false positives due to amplicon contamination.

72 REAL-TIME QUANTITATIVE PCR ASSAYS FOR THE DIAGNOSIS OF THREE HAEMOPLASMA SPECIES IN FELINE BLOOD SAMPLES. IR Peters1, CR Helps1, B Willi2, R Hofmann-Wellenhof1, S Tasker1. 1School of Clinical Veterinary Science, University of Bristol, Bristol, UK. 2School of Veterinary Medicine, University of California, Davis, USA.

Two distinct species of feline haemoplasma are recognised in the UK: Mycoplasma haemofelis (Mhf) and Candidatus Mycoplasma haemominutum (CMhm). These species differ in pathogenicity as Mhf infection often results in a severe haemolytic anaemia whilst CMhm infection usually results in few clinical signs. A novel feline species of haemoplasma (Mnov), most closely related to rodent haemoplasmas, has recently been reported in Switzerland and has been associated with anaemia.

Real-time quantitative PCR (QPCR) assays are excellent diagnostic tools for haemoplasma infection since they allow development of highly specific and quantitative assays. The purpose of this study was to develop Taqman QPCR assays for detection of all three feline haemoplasma species, together with an internal control. To confirm the specificity of the new assays, and assess whether Mnov was present in UK cats, the newly developed assays were applied to 60 stored blood samples previously tested at the School of Clinical Veterinary Science for the presence of Mhf and CMhm infection by a previously described QPCR assay.

Primers and Taqman probes were designed against published 16S rDNA sequences. Each of the three haemoplasma assays were combined with a feline 28S rDNA-specific assay to produce three duplex assays, thus allowing confirmation of the presence of cat DNA in the PCR (as an internal control). The three assays were sensitive enough to detect 1-2 copies of a sequence-specific plasmid per PCR in the duplex reaction. None of the assays showed cross-reactivity with the other haemoplasma species when tested with 1 x 10^7 copies of the sequence-specific plasmids. DNA was isolated from 60 samples (100 µl) using the Macherey-Nagel Nucleospin Blood kit. QPCR was performed on each sample with Qiagen HotStarTaq Master Mix using a Bio-Rad i-Cycler IQ.

All samples were positive for feline 28S rDNA in all assays performed. Of the samples which had previously tested haemoplasma positive [Mhf (n = 2) and CMhm (n = 25)], all were positive for the new assays, except one CMhm sample. Additionally, one sample was positive for CMhm with the new assay but had previously been negative. Both discordant samples were at the limit of detectability of the assays. One sample was dual positive for both CMhm and Mhf. The remaining samples were negative for all three haemoplasma species on both assays (n = 30). The results of this study demonstrate that the new duplex PCR assays for the detection of haemoplasma infection and that the recently described Mnov species is present in the UK cat population. Further studies are required to determine the prevalence of, and risk factors for, this species in the UK.

73 QUANTITATIVE REAL-TIME RT-PCR FOR FELINE CORONAVIRUS RNA IN THE BLOOD AND TISSUES OF CATS. IR Peters, C Drye, SG Siddell, S Tasker. School of Clinical Veterinary Science, University of Bristol, Bristol, UK.

The aim of the study was to compare the levels of feline coronavirus (FCoV) RNA in the blood and organs of cats with and without feline infectious peritonitis (FIP). Serum and whole blood (collected ante mortem), ascitic fluid (when present: 5 cats) and tissue samples (collected at post mortem into “RNA later”) were taken from six cats that had clinical signs and laboratory data suggestive of FCoV RNA was isolated from the fluids (100 µl) and tissues (30 mg) using the Macherey-Nagel Nucleospin RNA II kit, which includes a DNase step. From five of the 72 cats, RNA was also isolated from whole blood using the QIAgen “PAXgene” Blood RNA kit which includes an RNA stabilisation reagent. Histopathology ruled out a diagnosis of FIP in two of the cats whilst the remaining four cats were confirmed as having FIP.

A two-step quantitative real-time RT-PCR was performed using random hexamer primed cDNA and a FCoV specific Taqman based real-time RT-PCR assay. The assay was designed around the highly conserved MN gene junction from five different FCoV isolates. A feline G3PDH real-time PCR assay was performed before and after reverse transcription on each RNA sample to confirm successful cDNA synthesis. Threshold cycle (Ct) values were calculated and used to compare relative amounts of FCoV RNA in the samples.

The four cats with confirmed FIP all had high levels (Ct < 30) of FCoV RNA in the stomach, intestines, liver, spleen, kidney, body wall and intra-abdominal lymph nodes. Lower levels (Ct > 30) were found in the eye and blood. FCoV RNA levels in the tonsil, heart, lung, brain, bone marrow, peripheral lymph nodes and ascitic fluid varied considerably between cats. The non-FIP cats were both negative for FCoV RNA in the majority of organs tested but extremely low (Ct > 35) FCoV RNA levels were found in occasional samples.

In conclusion, high levels of FCoV RNA were found throughout the abdominal organs of cats with FIP compared with negative or extremely low levels in the non-FIP cats. Given the problems of interpreting FCoV antibody titres in suspected FIP patients, these findings may have diagnostic relevance in helping to distinguish cats with FIP from those with non-FIP disease. Unfortunately, blood, the most accessible body sample to obtain for diagnostic purposes, was not found to be the most useful in this respect. These data represent a small preliminary study and more samples, particularly in the non-FIP group, are being collected to confirm and extend our findings. It will also be interesting to compare the results of “effusive FIP” cats with those of cats with “dry (non-effusive)” FIP disease.


Feline immunodeficiency virus (FIV) is a lentivirus that shares many properties with human immunodeficiency virus (HIV). FIV infection occurs in approximately 6% of healthy cats and 19% of sick cats in the UK, and is a significant cause of feline morbidity. Humans infected with HIV have been shown to have reduced serum tryptophan concentrations mediated by up-regulation of the -interferon-induced enzyme indoleamine dioxygenase. Tryptophan concentrations progressively fall as HIV-associated disease develops, and treatments aimed at reversing the tryptophan depletion have decreased HIV-associated disease progression and improved survival in man.

The aim of this study was to measure the serum tryptophan and kynurenine (a major metabolite of tryptophan) concentrations in HIV-infected and uninfected cats, to see if the tryptophan changes reported in HIV-infected humans are mirrored by similar changes in FIV-infected cats.

Surplus feline serum and plasma subjected to commercial diagnostic laboratories was used for this study. All samples were tested for FIV antibody using an enzyme-linked immunosorbent assay (PetCheck, Idexx, UK). Samples from 69 FIV-negative and 76 FIV-positive cats were analysed by high performance liquid chromatography to determine tryptophan and kynurenine concentrations. No attempt was made to match samples with respect to age, sex or breed.
Sera were initially treated by the addition of an equal volume of 2M trichloroacetic acid to precipitate proteins and to allow subsequent clarification of the sample. FIV-negative and FIV-positive cats were statistically compared using t-tests, following demonstration of a normal distribution of logged data.

FIV-infected cats had significantly lower serum tryptophan concentrations compared to uninfected cats (39.6 μM vs. 50.5 μM, P = 0.012). FIV-infected cats had significantly higher kynurenine concentrations compared to uninfected cats (21.9 μM vs. 10.2 μM, P < 0.0001). The [kynurenine]:[tryptophan] ratios of FIV-infected cats were significantly higher than those of uninfected cats (0.55 vs 0.20, P < 0.0001).

These findings show that tryptophan metabolism is altered in FIV-infected cats, paralleling the situation with HIV infection. Pharmacological manipulation of tryptophan levels may be a feasible means of ameliorating clinical signs and disease progression in cats infected with FIV.

75 SEROPREVALENCE OF FIV AND FELV INFECTION AND DETERMINATION OF FIV SUBTYPES IN SICK DOMESTIC CATS IN SOUTH AFRICA. JP Schoeman1, R Kahn2, J Meers2, J Seddon2, T Schoeman3, M van Vuuren4. 1Department of Companion Animal Clinical Studies and 4Veterinary Tropical Diseases, Faculty of Veterinary Science, University of Pretoria, Onderstepoort, South Africa; 2School of Veterinary Science, University of Queensland, Australia and 3Cape Animal Medical Centre, Cape Town, South Africa.

The prevalences of Feline leukaemia virus (FeLV) and Feline immunodeficiency virus (FIV) infections have been shown across the world to be higher in sick than in healthy cats and to vary widely from study to study and from country to country. The prevalence of FIV and FeLV infections, as well as the subtypes of FIV infecting domestic cats in South Africa is currently unknown.

The aim of this study was to determine the prevalence of these viral infections in sick domestic cats and to determine the subtype(s) of FIV virus that exist in South Africa.

Serum was collected from 454 sick cats presenting to the Onderstepoort Veterinary Academic Hospital over a 7-year period from 1998 to 2004. The serum was submitted for detection of specific antibodies directed to FIV gag (group antigen) or env (envelope) gp40 proteins and the group-specific p27 core antigen of FeLV by ELISA. In addition, heparinised whole blood was collected from FIV positive cats, consisting of 11 of the above cats and a further 20 cats from 3 different centres in the country, viz Cape Town (12), Durban (4) and Johannesburg (4). The whole blood samples were subjected to polymerase chain reaction (PCR) amplification and sequences were determined for the V3-V5 region of the env gene of FIV.

Fifty-six out of 454 (12.3%) samples were positive for FeLV antigen and 101 out of 454 (22.2%) samples were positive for FIV antibody. Sixteen out of 454 cats (3.5%) were co-infected with both viruses. Twenty-two out of 31 (71%) samples revealed FIV subtype A and 9 out of 31 (29%) samples revealed FIV subtype C.

The prevalence rates of these two viruses in sick cats in South Africa is in line with prevalences encountered in the rest of the world, with the FIV prevalence rating amongst the highest in the world, closely resembling that found in two separate studies of cats in Australia and Italy and only slightly higher than rates from the UK and France. The prevalence of FIV is approximately twice that of FeLV in sick cats in South Africa. Although early literature suggested that each FIV subtype was limited in its geographical distribution, the data presented here add to growing evidence that many FIV subtypes are widely distributed around the world. This study thus provides supportive data that the introduction of the FIV vaccine containing subtype A, should protect the majority of cats in South Africa.