



# Oral Research

## Communications of the 21<sup>st</sup> ECVIM-CA Congress

Sevilla, Spain, 8th to 10th September 2011

Number	Day	Time	First Author Last Name	Title
ESVC - European Society of Veterinary Cardiology				
CA-O-1	Thursday 8 September	14.25- 14.40	Waxman	THE ROLE OF MECHANICAL STRAIN AND TRANSFORMING GROWTH FACTOR-BETA1 IN THE PATHOGENESIS OF CANINE MYXOMATOUS MITRAL VALVE DEGENERATION
CA-O-2	Thursday 8 September	14.40- 14.55	Estrada	ATRIAL-BASED PACING FOR SINUS NODE DYSFUNCTION
CA-O-3	Thursday 8 September	14.55- 15.10	Riesen	MYOCARDIAL EXPRESSION OF HYPERPOLARIZATION-ACTIVATED, CYCLIC NUCLEOTIDE-GATED PROTEINS IN HEALTHY CATS AND CATS WITH HYPERTROPHIC CARDIOMYOPATHY
CA-O-4	Thursday 8 September	15.10- 15.25	Payne	RISK FACTORS FOR SUDDEN DEATH IN FELINE HYPERTROPHIC CARDIOMYOPATHY
CA-O-5	Thursday 8 September	15.25- 15.40	Estrada	SINGLE VERSUS DUAL CHAMBER PACING IN DOGS: A COMPARISON OF IMPLANTATION TIMES AND COMPLICATION RATES (2005–2009)
CA-O-6	Thursday 8 September	15.40- 15.55	Ohad	DOES THE ADDITION OF CONSTANT RATE INFUSION TO INTERMITTENT BOLUS ADMINISTRATION OF INTRAVENOUS FUROSEMIDE IMPROVE MEDICAL OUTCOME OF PETS WITH LEFT-SIDED CONGESTIVE HEART FAILURE?
CA-O-7	Thursday 8 September	15.55- 16.10	Tidholm	LEFT ATRIAL EJECTION FRACTION ASSESSED BY 3-DIMENSIONAL ECHOCARDIOGRAPHY IN DOGS WITH AND WITHOUT MYXOMATOUS MITRAL VALVE DISEASE
CA-O-8	Thursday 8 September	16.10- 16.25	Stepien	ACCURACY OF AUSCULTATION ALONE TO IDENTIFY MITRAL INSUFFICIENCY IN ADULT WHIPPETS
CA-O-9	Thursday 8 September	16.25- 16.40	Bongrand	THREE-DIMENSIONAL ECHOCARDIOGRAPHY IMPROVES NONINVASIVE ESTIMATE OF CARDIAC OUTPUT IN HEALTHY DOGS
CA-O-10	Friday 9 September	9.00- 9.15	Damoiseaux	ADIPONECTIN PLASMA CONCENTRATION IN HEALTHY AND HEART FAILURE DOGS
CA-O-11	Friday 9 September	9.15- 9.30	Streitberger	MEASUREMENT OF PULMONARY TRANSIT TIME IN CATS BY USE OF THE ULTRASOUND CONTRAST MEDIA “SONOVUE®”. FEASIBILITY, REPRODUCIBILITY, AND NORMAL VALUES

CA-O-12	Friday 9 September	9.30-9.45	Trehou-Sechi	COMPARATIVE BREED-SPECIFIC FEATURES OF FELINE HYPERTROPHIC CARDIOMYOPATHY: A RETROSPECTIVE STUDY OF 344 CASES (2001-2011)
CA-O-13	Friday 9 September	9.45-10.00	Ferasin	CONGESTIVE HEART FAILURE IS NOT A PRIMARY CAUSE OF COUGHING IN DOGS WITH CHRONIC DEGENERATIVE MITRAL VALVE DISEASE (MMVD)
CA-O-14	Friday 9 September	10.00-10.15	Georgiev	SELF-REPORTED TWO-DIMENSIONAL ECHOCARDIOGRAPHIC ESTIMATES OF LEFT ATRIAL SIZE BY CARDIOLOGISTS: COMPARISON WITH MEASURED VARIABLES
CA-O-15	Friday 9 September	10.15-10.30	Chervier	A PROPOSED MECHANISM OF INHERITANCE OF HYPERTROPHIC CARDIOMYOPATHY IN A FAMILY OF BRITISH SHORTHAIRED CATS
CA-O-16	Friday 9 September	11.20-11.35	Georgiev	COMPARISON OF TWO ECHOCARDIOGRAPHIC ESTIMATES OF AORTIC VALVE DIAMETER IN DOGS AND CATS
CA-O-17	Friday 9 September	11.35-11.50	Baron Toaldo	ECHOCARDIOGRAPHIC ASSESSMENT OF REGIONAL LEFT ATRIAL DEFORMATION PROPERTIES USING TISSUE DOPPLER IMAGING: A FEASIBILITY STUDY ON HEALTHY DOGS
CA-O-18	Friday 9 September	11.50-12.05	Hambrook	THE EFFECT OF PIMOBENDAN ON THE CLINICAL OUTCOME AND SURVIVAL OF CATS WITH NON TAURINE RESPONSIVE DILATED CARDIOMYOPATHY
CA-O-19	Friday 9 September	12.05-12.20	Carnabuci	2D-SPECKLE TRACKING ECHOCARDIOGRAPHY (STE) FOR ASSESSMENT OF GLOBAL AND REGIONAL LEFT VENTRICULAR PEAK SYSTOLIC STRAIN AND STRAIN RATE IN HEALTHY LABRADOR RETRIEVER DOGS
CA-O-20	Friday 9 September	12.20-12.35	Zois	CHANGES IN RADIAL AND LONGITUDINAL SEGMENTAL MYOCARDIAL DEFORMATION IN DOGS WITH MYXOMATOUS MITRAL VALVE DISEASE DETECTED BY USE OF TWO-DIMENSIONAL SPECKLE TRACKING ECHOCARDIOGRAPHY
CA-O-21	Friday 9 September	12.35-12.50	Kraus	RELATION OF VITAMIN D STATUS TO CONGESTIVE HEART FAILURE AND INCIDENT CARDIOVASCULAR EVENTS IN DOGS

ESCG - European Society of Comparative Gastroenterology

GA-O-1	Thursday 8 September	14.25-14.40	Dandrieux	SHOULD CHLORAMBUCIL-PREDNISOLONE COMBINATION THERAPY BE USED IN PREFERENCE TO AZATHIOPRINE-PREDNISOLONE FOR THE TREATMENT OF CHRONIC ENTEROPATHIES WITH CONCURRENT PROTEIN LOSING ENTEROPATHY?
GA-O-2	Thursday 8 September	14.40-14.55	Gaschen	EVALUATION OF THE EFFECTS OF METOCLOPRAMIDE AND CISAPRIDE ON CANINE GASTRIC, SMALL BOWEL AND COLONIC MOTILITY USING A WIRELESS MOTILITY CAPSULE
GA-O-3	Thursday 8 September	14.55-15.10	Lynch	DECREASE IN TOLL- AND NOD-LIKE RECEPTOR EXPRESSION IN POST-TREATMENT DUODENAL MUCOSAL TISSUE FROM DOGS WITH CHRONIC ENTEROPATHY
GA-O-4	Thursday 8 September	15.10-15.25	Lynch	DECREASE IN CYTOKINE GENE EXPRESSION IN POST-TREATMENT DUODENAL MUCOSAL TISSUE FROM DOGS WITH CHRONIC ENTEROPATHY
GA-O-5	Thursday 8 September	15.25-15.40	Gostelow	PHYLOGENETIC COMPARISON OF DUODENAL BACTERIA IN CATS WITH FOOD RESPONSIVE ENTEROPATHY AND INFLAMMATORY BOWEL DISEASE USING 16S rRNA GENE PYROSEQUENCING

GA-O-6	Thursday 8 September	15.40- 15.55	Heilmann	SERUM CALGRANULIN CONCENTRATIONS IN DOGS WITH INFLAMMATORY BOWEL DISEASE
GA-O-7	Thursday 8 September	15.55- 16.10	Grellet	FECAL CALPROTECTIN CONCENTRATION IN ADULT DOGS WITH AND WITHOUT DIGESTIVE TROUBLES
GA-O-8	Friday 9 September	16.30- 16.45	Procoli	SEROREACTIVITY AGAINST BACTERIAL FLAGELLIN IN DOGS WITH INFLAMMATORY BOWEL DISEASE: PRELIMINARY FINDINGS
GA-O-9	Friday 9 September	16.45- 17.00	Lobetti	TREATMENT OF OESOPHAGEAL SPIROCERCOSIS IN 20 DOGS WITH ORAL DORAMECTIN
GA-O-10	Friday 9 September	17.00- 17.15	Schmitz	INTERLEUKIN-17A AND INTERLEUKIN-22 MRNA EXPRESSION IS LOW IN DUODENAL TISSUE FROM DOGS WITH INFLAMMATORY BOWEL DISEASE
GA-O-11	Friday 9 September	17.15- 17.30	Jones	UTILITY OF THE SERUM PANCREATIC ELASTASE-1 IN THE DIAGNOSIS OF CANINE PANCREATITIS
GA-O-12	Friday 9 September	17.30- 17.45	Xenoulis	SERUM TRIGLYCERIDE AND CHOLESTEROL CONCENTRATIONS AND LIPOPROTEIN PROFILES IN DOGS WITH NATURALLY OCCURRING PANCREATITIS AND HEALTHY CONTROL DOGS
GA-O-13	Friday 9 September	17.45- 18.00	Kilpinen	TYLOSIN ADMINISTRATION INCREASES THE LEVELS OF ENTEROCOCCUS SPP. IN THE FECES OF DOGS WITH TYLOSIN-RESPONSIVE DIARRHEA

## ESVE - European Society of Veterinary Endocrinology

EN-O-1	Thursday 8 September	9.00- 9.15	van der Helm	INVOLVEMENT OF THE GH-IGF PATHWAY IN CANINE CORTISOL-SECRETING ADRENOCORTICAL TUMORS
EN-O-2	Thursday 8 September	9.15- 9.30	Smets	LONG-TERM FOLLOW-UP OF RENAL FUNCTION IN DOGS WITH CUSHING'S DISEASE BEFORE AND AFTER TREATMENT
EN-O-3	Thursday 8 September	9.30- 9.45	Niessen	ROUTINE SCREENING OF DIABETIC CATS FOR ACROMEGALY: OVERDUE OR OVERKILL?
EN-O-4	Thursday 8 September	9.45- 10.00	Müller	TSH STIMULATION TEST WITH RECOMBINANT HUMAN TSH (RH-TSH) FOR THE DIAGNOSIS OF FELINE HYPERTHYROIDISM
EN-O-5	Thursday 8 September	10.00- 10.15	Williams	INDICES OF URINARY CAUXIN AND N-ACETYL-?-D-GLUCOSAMINIDASE (NAG) ARE NOT PREDICTORS OF SURVIVAL OR THE DEVELOPMENT OF AZOTAEMIA IN HYPERTHYROID CATS
EN-O-6	Thursday 8 September	10.15- 10.30	Williams	CHANGES IN PARATHYROID HORMONE (PTH) CONCENTRATIONS AFTER TREATMENT OF HYPERTHYROID CATS WITH VARIABLE RENAL FUNCTION.
EN-O-7	Thursday 8 September	11.20- 11.35	Forcada Atienza	A MISSENSE MUTATION IN THE CODING SEQUENCE OF MC4R (MC4R:C.92 C>T) IS PRIMARILY ASSOCIATED WITH SUSCEPTIBILITY TO DIABETES MELLITUS IN OBESE DSH CATS
EN-O-8	Thursday 8 September	11.35- 11.50	Zini	PANCREATIC ENZYMES ACTIVITY AND ULTRASONOGRAPHIC FINDINGS IN DIABETIC CATS AT DIAGNOSIS AND DURING FOLLOW-UP
EN-O-9	Thursday 8 September	11.50- 12.05	Callegari	PROGNOSTIC FACTORS IN CATS WITH NEWLY DIAGNOSED DIABETES MELLITUS
EN-O-10	Friday 9 September	14.25- 14.40	Zoia	RELATIVE FIBRINOGEN AND HAPTOGLOBIN DEFICENCY IN DOGS WITH NATURAL OCCURRING ADDISON DISEASE

EN-O-11	Friday 9 September	14.40-14.55	Arenas	LONG-TERM SURVIVAL OF DOGS WITH ADRENAL-DEPENDENT HYPERADRENOCORTICISM TREATED WITH MITOTANE VERSUS TRILOSTANE
EN-O-12	Friday 9 September	14.55-15.10	Hafner	INTENSIVE INTRAVENOUS INSULIN THERAPY IN DIABETIC CATS
EN-O-13	Friday 9 September	15.10-15.25	Bresciani	ACCURACY OF CAPILLARY BLOOD 3- $\beta$ -HYDROXYBUTYRATE DETERMINATION FOR THE DETECTION AND TREATMENT OF CANINE DIABETIC KETOACIDOSIS
EN-O-14	Friday 9 September	15.25-15.40	Scott-Moncrieff	ACCURACY OF SERUM FREE THYROXINE CONCENTRATIONS DETERMINED BY A NEW VETERINARY CHEMILUMINESCENT IMMUNOASSAY IN EUTHYROID AND HYPOTHYROID DOGS
EN-O-15	Friday 9 September	15.40-15.55	Shiel	QUALITATIVE AND SEMI-QUANTITATIVE ASSESSMENT OF THYROXINE BINDING GLOBULIN IN THE GREYHOUND AND OTHER DOG BREEDS

#### ESCH - European Society of Comparative Hepatology

HE-O-1	Saturday 10 September	9.00-9.15	Weisse	ENDOVASCULAR TREATMENT AND/OR EVALUATION OF CANINE INTRAHEPATIC PORTOSYSTEMIC SHUNTS: SHORT- AND LONG-TERM EXPERIENCE IN 100 DOGS.
HE-O-2	Saturday 10 September	9.15-9.30	Oudry	EVALUATION OF HEPATIC DISEASES IN IN VIVO DOGS WITH FIBROSCAN <sup>®</sup> DEVICE: A FEASIBILITY STUDY
HE-O-3	Saturday 10 September	9.30-9.45	Fieten	OPTIMIZING DIAGNOSIS AND TREATMENT OF COPPER ASSOCIATED HEPATITIS IN THE LABRADOR
HE-O-4	Saturday 10 September	9.45-10.00	Kook	COMPARATIVE BACTERIOLOGIC AND MOLECULAR EXAMINATIONS OF GALLBLADDER BILE OF HEALTHY DOGS.
HE-O-5	Saturday 10 September	10.00-10.15	Stosic	EMBOLIZATION OF EXTRAHEPATIC PORTOSYSTEMIC SHUNTS IN DOGS WITH A SINGLE COIL

#### ESVONC - European Society of Veterinary Oncology

ON-O-1	Friday 9 September	14.25-14.40	Berlato	IS MINICHROMOSOME MAINTENANCE PROTEIN 7 (MCM7) BETTER THAN HISTOLOGICAL GRADE, MITOTIC INDEX, OR KI-67 IN PREDICTING SURVIVAL IN DOGS WITH CUTANEOUS MAST CELL TUMOUR?
ON-O-2	Friday 9 September	14.40-14.55	Gallay Lepoutre	EVALUATION OF DOPPLER ECHOCARDIOGRAPHY, TISSUE DOPPLER IMAGING AND BIOMARKERS MEASUREMENT FOR THE DETECTION OF DOXORUBICIN-INDUCED CARDIOTOXICITY IN DOGS: A PILOT STUDY
ON-O-3	Friday 9 September	14.55-15.10	Langner	ASSESSMENT OF ANTIGEN RECEPTOR REARRANGEMENT IN CANINE LYMPHOMA PATIENTS BY A NOVEL SYBRGREEN <sup>®</sup> REAL-TIME POLYMERASE CHAIN REACTION
ON-O-4	Friday 9 September	15.10-15.25	Floch	CANINE AGGRESSIVE LARGE GRANULAR LYMPHOCYTE LYMPHOMAS: A CLINICAL AND MORPHOLOGICAL RETROSPECTIVE STUDY OF 80 CASES
ON-O-5	Friday 9 September	15.25-15.40	Laberke	EFFICACY OF MESNA AND FUROSEMIDE IN THE PREVENTION OF STERILE HAEMORRHAGIC CYSTITIS IN DOGS WITH MALIGNANT LYMPHOMA RECEIVING A CYCLOPHOSPHAMIDE-CONTAINING CHEMOTHERAPY PROTOCOL – A RETROSPECTIVE STUDY OF 131 DOGS (1997-2009)

ON-O-6	Friday 9 September	15.40- 15.55	Santos	ERBB2 ONCOGENE EXTRACELLULAR DOMAIN DNA SEQUENCE VARIANTS, RNA EXPRESSION AND IN SILICO PROTEIN ANALYSIS IN CAT MAMMARY LESIONS: CORRELATION WITH CLINICOPATHOLOGICAL FEATURES
ON-O-7	Friday 9 September	16.30- 16.45	Tater	ASSESSMENT OF CARDIAC TROPONIN I (CTNI) AND TISSUE VELOCITY IMAGING (TVI) IN 14 DOGS WITH MALIGNANT LYMPHOMA UNDERGOING DOXORUBICIN TREATMENT - FIRST RESULTS
ON-O-8	Friday 9 September	16.45- 17.00	Polton	FELINE INJECTION SITE SARCOMA – RESULTS FOLLOWING NEOADJUVANT AND ADJUVANT CHEMOTHERAPY COMBINED WITH COMPARTMENTAL TUMOUR RESECTION
ON-O-9	Friday 9 September	17.00- 17.15	Milner	NASAL AND ORAL TUMOURS IN SMALL ANIMALS TREATED WITH STEREOTACTIC RADIOSURGERY
ON-O-10	Friday 9 September	17.15- 17.30	Finck	COMPARISON OF COMPUTED TOMOGRAPHY AND RHINOSCOPY FOR DIAGNOSIS OF CANINE AND FELINE NASAL TUMORS: RETROSPECTIVE STUDY OF 22 CASES (2007-2010)
ON-O-11	Friday 9 September	17.30- 17.45	Milner	THE IMMUNE RESPONSE IN DOGS WITH MELANOMA TO DISIALOGANGLIOSIDE GD3 VACCINATION: A PHASE I CLINICAL TRIAL
ON-O-12	Friday 9 September	17.45- 18.00	Meier	PATUPILEONE (EPOTHILONE B) IN CANINE REFRACTORY LYMPHOMA AND ADVANCED SOLID TUMORS: A PHASE I/II CLINICAL STUDY

\*ESVONC ABSTRACTS APPEAR IN *VETERINARY CLINICAL ONCOLOGY*

ESVCN - European Society of Veterinary Clinical Nutrition

NU-O-1	Friday 9 September	14.25- 14.40	van de Velde	CHRONIC OBESITY DOES NOT ALTER INFLAMMATORY CYTOKINES BUT EVOKES AN IMPAIRED LYMPHOCYTE FUNCTION IN ADULT HEALTHY DOGS
NU-O-2	Friday 9 September	14.40- 14.55	Acke	CAMPYLOBACTER SPECIES AND MULTILOCUS SEQUENCE TYPES FROM COMMERCIAL RAW MEAT DIETS FOR PETS
NU-O-3	Friday 9 September	14.55- 15.10	Tvarijonaviciute	EFFECTS OF WEIGHT LOSS IN OBESE CATS ON BIOCHEMICAL ANALYTES RELATED TO INFLAMMATION AND GLUCOSE HOMEOSTASIS
NU-O-4	Friday 9 September	15.10- 15.25	Lutz	IMPACT OF TREATMENT WITH RECOMBINANT FELINE INTERFERON OMEGA ON RESOLUTION OF ANOREXIA OF UNKNOWN ORIGIN IN CATS
NU-O-5	Friday 9 September	15.25- 15.40	Iff	WEIGHT LOSS IMPROVES OXYGENATION IN RELATION TO FAT LOSS DURING SEDATION IN OBESE PET DOGS
NU-O-6	Friday 9 September	15.40- 15.55	Queau	EFFECT OF THE ADDITION OF SOLUBLE FIBERS TO DRY EXPANDED DIETS ON FECAL AND URINE WATER CONTENT IN HEALTHY CATS
NU-O-7	Friday 9 September	16.30- 16.45	Serisier	WEIGHT LOSS AND POST WEIGHT LOSS MAINTENANCE ENERGY REQUIREMENT OF OBESE COLONY CATS
NU-O-8	Friday 9 September	16.45- 17.00	German	ADJUNCTIVE DIRLOTAPIDE THERAPY FOR DURING DIET-BASED WEIGHT MANAGEMENT – A PILOT STUDY

ISFM - International Society of Feline Medicine

FE-O-1	Saturday 10 September	16.30- 16.45	Verjans	SCREENING OF APPARENTLY HEALTHY MIDDLE-AGED AND OLDER CATS
FE-O-2	Saturday 10 September	16.45- 17.00	Lobetti	PREVALENCE OF INFECTIOUS AGENTS IN CATS IN SOUTH AFRICA
FE-O-3	Saturday 10 September	17.00- 17.15	Gowan	RETROSPECTIVE ANALYSIS OF THE EFFECT OF LONG-TERM USE OF MELOXICAM ON LONGEVITY OF CATS WITH CHRONIC PAIN AND CHRONIC KIDNEY DISEASE

VBPS - Veterinary Blood Pressure Society

*NO ABSTRACTS RECEIVED*

ESVIM - European Society of Veterinary Internal Medicine

IM-O-1	Thursday 8 September	9.45- 10.00	Schuller	IDENTIFICATION OF ANTIGENS OF LEPTOSPIRA REACTIVE WITH SERA FROM DOGS WITH ACUTE LEPTOSPIROSIS WITH AND WITHOUT PULMONARY HAEMORRHAGE
IM-O-2	Thursday 8 September	10.00- 10.15	Lilja-Maula	NEW POTENTIAL BIOMARKERS FOR CANINE IDIOPATHIC PULMONARY FIBROSIS
IM-O-3	Thursday 8 September	10.15- 10.30	O'Brien	COMPUTED TOMOGRAPHIC IMAGING OF DOGS WITH LARYNGEAL OR TRACHEAL AIRWAY OBSTRUCTION
IM-O-4	Thursday 8 September	11.20- 11.35	Krafft	HIGH SERUM TRANSFORMING GROWTH FACTOR- BETA 1 CONCENTRATION IN WEST HIGHLAND WHITE TERRIERS: A KEY TO THE BREED PREDISPOSITION IN CANINE IDIOPATHIC PULMONARY FIBROSIS ?
IM-O-5	Thursday 8 September	11.35- 11.50	Heikkilä	MATRIX METALLOPROTEINASE -2 AND -9 IN BRONCHOALVEOLAR LAVAGE FLUID OF DOGS WITH IDIOPATHIC PULMONARY FIBROSIS AND CHRONIC BRONCHITIS
IM-O-6	Thursday 8 September	11.50- 12.05	Hugonnard	EVALUATION OF POLYMERASE CHAIN REACTION IN THE DIAGNOSIS OF CANINE LEPTOSPIROSIS: COMPARISON WITH SEROLOGIC TESTING IN 33 DOGS
IM-O-7	Thursday 8 September	12.05- 12.20	Smith	CYTOKINES IN STORED ERYTHROCYTE CONCENTRATES
IM-O-8	Thursday 8 September	12.20- 12.35	Shibly	EFFECTS OF NEBULISED N-ACETYLCYSTEINE ON PLETHYSMOGRAPHIC FINDINGS IN CATS
IM-O-9	Thursday 8 September	14.25- 14.40	Weber	PREVALENCE OF MYCOPLASMA SPP. IN BRONCHOALVEOLAR AND NASAL LAVAGE IN CATS WITH CHRONIC BRONCHIAL DISEASE
IM-O-10	Thursday 8 September	14.40- 14.55	Gomez Ochoa	EFFICACY OF DOMPERIDONE FOR THE TREATMENT OF MILD AND MODERATE CASES OF CANINE LEISHMANIOSIS: CLINICAL AND IMMUNOLOGICAL SHORT-TERM FOLLOW-UP
IM-O-11	Thursday 8 September	14.55- 15.10	Balog	COMPARISON OF THE EFFECT OF HUMAN INTRAVENOUS IMMUNOGLOBULIN VERSUS VINCRISTINE ON PLATELET RECOVERY TIME IN DOGS WITH SEVERE IDIOPATHIC IMMUNE-MEDIATED THROMBOCYTOPENIA
IM-O-12	Thursday 8 September	15.10- 15.25	Lalor	INVESTIGATION OF RELATIONSHIP BETWEEN VITAMIN D STATUS AND MYCOBACTERIAL INFECTIONS IN CATS

IM-O-13	Thursday 8 September	15.25-15.40	Fischer	EFFECT OF PROPENTOFYLLINE ON THE SURVIVAL TIME AND QUALITY OF LIFE OF CATS WITH FELINE INFECTIOUS PERITONITIS
IM-O-14	Thursday 8 September	15.40-15.55	Galler	EFFICACY AND SYSTEMIC EFFECTS OF INHALED GLUCOCORTICOIDS IN NATURALLY OCCURRING FELINE ASTHMA AND CHRONIC BRONCHITIS
IM-O-15	Thursday 8 September	15.55-16.10	Tamborini	A COMPARISON OF THE PACKED CELL VOLUME (PCV) AND TOTAL PLASMA PROTEIN (TPP) VALUES GAINED FROM CANINE LITHIUM HEPARIN AND EDTA BLOOD SAMPLES USING MICROHAEMATOCRIT AND REFRACTOMETRY METHODS
IM-O-16	Thursday 8 September	16.10-16.25	Zoia	A NEW APPROACH TO PLEURAL EFFUSION IN DOGS: MARKERS TO DISCRIMINATE BETWEEN TRANSUDATES AND EXUDATES
IM-O-17	Thursday 8 September	16.25-16.40	Held	ACCURACY OF DIAGNOSTIC TESTS FOR FELINE INFECTIOUS PERITONITIS (FIP) IN CATS WITH BODYCAVITY EFFUSION

ESVNU - European Society of Veterinary Nephrology and Urology

RE-O-1	Friday 9 September	17.15-17.30	Berent	URETERAL STENTING FOR FELINE URETERAL OBSTRUCTIONS: TECHNICAL AND CLINICAL OUTCOMES: 74 URETERS (2006-2011).
RE-O-2	Friday 9 September	17.30-17.45	Berent	THE USE OF A SUBCUTANEOUS URETERAL BYPASS DEVICE FOR URETERAL OBSTRUCTIONS IN CATS
RE-O-3	Friday 9 September	17.45-18.00	Canonne-Guibert	RETROSPECTIVE STUDY OF URETERAL ECTOPIAIN MALE DOGS: TEN CASES (2002-2011)
RE-O-4	Saturday 10 September	9.00-9.15	Francey	REGIONAL CITRATE ANTICOAGULATION FOR EXTRACORPOREAL BLOOD PURIFICATION TECHNIQUES IN DOGS
RE-O-5	Saturday 10 September	9.15-9.30	Schweighauser	INTOXICATION WITH GRAPES OR RAISINS CAUSING SEVERE ACUTE KIDNEY INJURY AND NEUROLOGICAL SIGNS IN DOGS.
RE-O-6	Saturday 10 September	9.30-9.45	Hoglund	EXCITEMENT DURING EXAMINATION AT A CLINIC AFFECTS RENAL FUNCTION IN HEALTHY DOGS
RE-O-7	Saturday 10 September	9.45-10.00	Defauw	ASSESSMENT OF RENAL DYSFUNCTION USING URINARY MARKERS IN SOUTH AFRICAN CANINE BABESIOSIS
RE-O-8	Saturday 10 September	10.00-10.15	Martinez Padua	DOGS WITH VISCERAL LEISHMANIASIS AND SOME CORRELATIONS BETWEEN RENAL DAMAGE INDICATORS
RE-O-9	Saturday 10 September	10.15-10.30	Pomba	DISSEMINATION OF THE HUMAN PANDEMIC ST131-O25B CLONE AMONG UROPATHOGENIC ESCHERICHIA COLI ISOLATES FROM COMPANION ANIMALS IN PORTUGAL
RE-O-10	Saturday 10 September	11.20-11.35	Dorsch	IDENTIFICATION OF BACTERIAL DNA IN THE URINE OF CATS WITH IDIOPATHIC CYSTITIS
RE-O-11	Saturday 10 September	11.35-11.50	Zeza	INTRAVESICAL LIDOCAINE TREATMENT OF CATS WITH OBSTRUCTIVE LOWER URINARY TRACT DISEASE (LUTD)
RE-O-12	Saturday 10 September	11.50-12.05	van Hoek	LARGER DAILY URINARY VOLUMES IN HEALTHY CATS INDUCE MORE FREQUENT MICTURITIONS OF SMALLER VOLUMES

ESVCP - European Society of Veterinary Clinical Pathology ABSTRACTS PUBLISHED ELSEWHERE\*

CP-O-1	Saturday 10 September	11.20-11.35	Mukorera	VASCULAR ENDOTHELIAL GROWTH FACTOR AS A MARKER FOR NEOPLASTIC TRANSFORMATION IN CANINE SPIROCERCOSIS
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CP-O-2	Saturday 10 September	11.35- 11.50	Monti	INITIAL EVALUATION OF URINARY CYSTATIN C MEASUREMENT BY IMMUNOTURBIDIMETRY (PETIA) IN DOGS AS A MARKER OF RENAL TUBULAR DAMAGE.
CP-O-3	Saturday 10 September	11.50- 12.05	Spodsberg	DOGS WITH DISEASES PREDISPOSING FOR THROMBOSIS HAVE SIGNIFICANTLY LOWER FIBRINOLYSIS POTENTIAL COMPARED TO CLINICALLY HEALTHY DOGS IN TISSUE PLASMINOGEN ACTIVATOR (TPA) MODIFIED TEG ANALYSES
CP-O-4	Saturday 10 September	12.05- 12.20	Nagel	THROMBOELASTOGRAPHIC EVALUATION OF HAEMOSTATIC FUNCTION IN DOGS WITH NATURAL ENVENOMATION BY SOUTH AFRICAN SNAKES
CP-O-5	Saturday 10 September	12.20- 12.35	Hunter	MEASUREMENT OF THE TOTAL PROANP PRODUCT IN MAMMALS BY PROCESSING INDEPENDENT ANALYSIS
CP-O-6	Saturday 10 September	12.35- 12.50	Zoia	MEAN PLATELET COMPONENT CONCENTRATION IN DOGS WITH IMMUNE-MEDIATED HAEMOLYTIC ANAEMIA

*\*ESVCP ABSTRACTS APPEAR IN VETERINARY CLINICAL PATHOLOGY*

**CA-O-1**

**THE ROLE OF MECHANICAL STRAIN AND TRANSFORMING GROWTH FACTOR-BETA1 IN THE PATHOGENESIS OF CANINE MYXOMATOUS MITRAL VALVE DEGENERATION.** A. Waxman<sup>1</sup>, B.G. Kornreich<sup>1</sup>, N.S. Moise<sup>1</sup>, J.T. Butcher<sup>2</sup>. <sup>1</sup>Cornell University College of Veterinary Medicine, ITHACA, NY, United States of America, <sup>2</sup>Cornell University, Biomedical Engineering, ITHACA, NY, United States of America

Myxomatous valve degeneration (MVD) is the leading cause of cardiac-related death in small breed dogs, but the pathogenic mechanisms are poorly understood. Recent evidence suggests that transforming growth factor-beta1 (TGF-B1) induces myofibroblastic activation in mitral valve interstitial cells (MVIC). TGF-B1 is also regulated by mechanical strain, which is prevalent in the valve leaflet. The interactions between mechanical strain and TGF-B1 in canine MVD is unknown. We hypothesized that TGF-B1 induces a myxomatous phenotype in MVIC that is potentiated by cyclic mechanical strain. To test this, we developed a novel three-dimensional (3D) in vitro culture assay.

Canine MVIC were isolated from anterior mitral valve leaflets by collagenase digestion and cultured in DMEM with 10% FBS. Cells were encapsulated in 3D collagen hydrogels and cultured for 7 days (n=6, normal and moderate MVD) with or without 5 ng/ml TGF-B1. Gel compaction area was calculated daily. MVIC in 3D collagen culture were also exposed to 15% cyclic equibiaxial strain at 1Hz for 48 h (n=4, mild MVD). Static cultures served as controls. Cell phenotype was then determined using immunofluorescence to vimentin and  $\alpha$ -SMA. ANOVA with repeated measures was performed with day, treatment and disease state as covariants. A Mann-Whitney U test was performed to compare static and strained construct phenotypes.

Our results show that collagen compaction was significantly greater (1) with TGF-B1 treatment for both the normal and MVD valves compared to no treatment ( $p < 0.001$ ) and (2) in MVD valves with and without TGF-B1 treatment compared to normal valves ( $p < 0.001$ ). Strained constructs showed a significant increase in vimentin expression ( $p < 0.05$ ) while no difference was observed in  $\alpha$ -SMA.

Collectively, these results suggest that MVIC differentiate towards myxomatous phenotypes via TGF-B1 in vitro regardless of MVD state. Increased collagen compaction from diseased MVIC may be due to elevated endogenous TGF-B1 signaling. Cyclic equibiaxial strain applied here protected against a myxomatous phenotype, but alternate strain profiles such as may be observed in hearts with MVD may differently regulate the MVIC phenotype, both in concert and independent of TGF-B1 signaling. Studies are underway to assess different strain patterns, rates, and signaling molecules on MVIC phenotype in canine MVD.

**CA-O-2**

**A TRIAL-BASED PACING FOR SINUS NODE DYSFUNCTION.** A.H. Estrada<sup>1</sup>, N.S. Moise<sup>2</sup>, R. Pariaut<sup>3</sup>, B.J. Horowitz<sup>1</sup>, S. Hemsley<sup>2</sup>, M. Powell<sup>1</sup>. <sup>1</sup>University of Florida, GAINESVILLE, United States of America, <sup>2</sup>Cornell University, ITHACA, United States of America, <sup>3</sup>Louisiana State University, BATON ROUGE, United States of America

Atrial pacing of dogs with sick sinus syndrome (SSS) and adequate atrioventricular (AV) node conduction permits physiological depolarization and contraction of the ventricles; thereby decreasing the possibility of pacemaker syndrome. This complication is more likely in dogs with AV valve regurgitation, which is common in the small breeds affected with SSS. This retrospective study examined atrial pacing (AAI) in dogs with SSS with the hypothesis that it is feasible and not different in complication frequency to ventricular pacing.

Medical records of patients with SSS treated with AAI pacing were evaluated. Sixteen dogs with SSS and AAI pacing were identified. Follow up time ranged from 45-1,227 days (mean 436  $\pm$  344). One dog developed complete AV block 27 days post-operatively requiring ventricular pacing. The remainder of the dogs had occasional 2nd degree AV block detected on either

ECG or Holter evaluation, either immediately post-operatively or only during times of sleep and rest on recheck evaluations. Lead dislodgment occurred in 3/16 dogs 1, 19 and 27 days post-operatively. All lead dislodgements occurred with actively fixed, bipolar leads. Lead perforation into the pericardial space occurred in 2/16 dogs 79 and 120 days post-operatively, both with passively fixed, bipolar leads. Lead migration occurred in 3/16 dogs 57, 192, and 1,016 days after implantation. None of these dogs had complete loss of capture but all required higher thresholds for pacing. Two of these dogs likely had microdislodgement of the lead tip and 1 dog had a stable lead tip with a loop of lead located in the right ventricle on follow up. Two of these were active fixation leads and 1 was a passive fixation lead. All 3 were bipolar leads. Oversensing, consisting of refractory sensed events and noise reversion pacing was seen in 5/5 unipolar leads and 4/11 bipolar leads.

Clinically important AV block does not appear to occur in the long term for dogs with SSS. Lead perforation, dislodgement and migration appear to be risks, which are greater when compared to early published experiences with ventricular pacing in veterinary cardiology and atrial pacing in people. Leads with reduced torque at the lead tip, higher flexibility during atrial contraction and increased lead-tip surface of contact with the endocardium may reduce these complications. Because of the potential long-term benefits of AAI pacing in dogs with SSS, veterinary cardiologists should continue to optimize AAI pacing and compare this method to ventricular pacing for dogs with SSS.

**CA-O-3**

**MYOCARDIAL EXPRESSION OF HYPERPOLARIZATION-ACTIVATED, CYCLIC NUCLEOTIDE-GATED PROTEINS IN HEALTHY CATS AND CATS WITH HYPERTROPHIC CARDIOMYOPATHY.** S.C. Riesen<sup>1</sup>, K.E. Schober<sup>2</sup>, R. Terentiev<sup>2</sup>, D. Terentiev<sup>2</sup>, J.D. Bonagura<sup>2</sup>, C.A. Carnes<sup>2</sup>. <sup>1</sup>Vetmeduni Vienna, WIEN, Austria, <sup>2</sup>The Ohio State University, OHIO, United States of America

Cardiac pacemaking is the result of the electrical activity of pacemaker cells, determined by the interplay of several ionic currents, pumps, and exchangers. Of the cellular and molecular mechanisms involved, the funny current (If) plays an important role. The molecular correlates of native funny channels are the hyperpolarization-activated, cyclic nucleotide-gated (HCN) channels, of which four isoforms are known (HCN1 to 4). Of these isoforms, HCN4 is the most abundant and predominantly expressed in the sinoatrial node. It has been previously shown in humans and rodents that the density of HCN channels in the heart does change during cardiac development and may be altered with myocardial disease and in response to hormonal stimuli.

The aim of this prospective study was to evaluate the expression of different HCN isoforms in myocardial tissue from healthy control cats and cats with hypertrophic cardiomyopathy (HCM) and to compare expression of HCN between kittens and adult healthy cats.

Myocardial tissue samples of the right atrium, left ventricle (LV), and right ventricle (RV) of control cats (adults [n=16] and kittens [n=4]) and cats with HCM (n=8) were collected. Expression of HCN was determined by immunoblot analysis using antibodies against HCN2 and HCN4. Optical densities of Western blot bands were measured and compared subsequently by one-way ANOVA and unpaired t-test.

HCN4 was consistently evaluable by immunoblot analysis while HCN2 was not. HCN4 expression was increased in LV myocardial samples of cats with HCM (P=0.036) compared to control cats, whereas in RV samples only a trend toward upregulation was observed (P=0.055). Myocardial HCN4 expression was not different between kittens and adult cats.

Our results indicate that myocardial HCN4 expression can be evaluated in cats by immunoblot analysis and that HCN4 expression is upregulated in LV myocardial tissue of cats with HCM. The pathophysiological importance of HCN overexpression with regard to myocyte function and altered automaticity deserves further study.

**CA-O-4**

**RISK FACTORS FOR SUDDEN DEATH IN FELINE HYPERTROPHIC CARDIOMYOPATHY.** J. Payne, V. Luis Fuentes, D. Connolly, A. Boswood, D. Brodbelt. Royal Veterinary College, HATFIELD, HERTFORDSHIRE, United Kingdom

Sudden death is poorly described in cats with hypertrophic cardiomyopathy (HCM). Our aim was to evaluate sudden death in a population of cats with HCM.

Clinical and echocardiographic records of feline HCM cases from 2004-2009 were reviewed. Outcome was obtained from clinical records or referring veterinarians/owners. Sudden death was defined as cats with a witnessed death/found dead without obvious cause but apparently well in the preceding 24 hours. Sudden death cats were compared to cats that died in other ways (euthanasia due to congestive heart failure (CHF)/aortic thromboembolism (ATE) or non-cardiac). Symptomatic status was classified as: 'asymptomatic' / 'collapse' / 'CHF/ATE'. Arrhythmias were identified on ECG, during echocardiography or during ambulatory ECG monitoring. Left ventricular outflow tract obstruction (LVOTO) was defined as LVOT velocity >2.5 m/s. Multivariable analysis was performed using binary logistic regression. Model fit was assessed with the Hosmer-Lemeshow test.

Of 289 HCM cats, 145 had died including 15 sudden deaths (5.1% of all cats). Collapse and ventricular tachycardia at presentation were univariable predictors of sudden death ( $p=0.003$  and  $p=0.041$  respectively). Symptomatic status, heart rate, auscultatory arrhythmia or gallop, ventricular tachycardia, regional wall hypokinesis and LVOTO were taken forward for evaluation in the multivariable model. Collapse and LVOTO remained as predictors of sudden death (OR 132.4 (7.2 - 2429.0) and 5.7 (1.1 - 29.5) respectively). Model fit was good (Hosmer-Lemeshow test  $p=0.772$ ).

Sudden death occurs in cats with HCM, and collapse, ventricular tachycardia and/or LVOTO may increase this risk. Results should be interpreted cautiously due to small sample size.

**CA-O-5**

**SINGLE VERSUS DUAL CHAMBER PACING IN DOGS: A COMPARISON OF IMPLANTATION TIMES AND COMPLICATION RATES (2005-2009).** A.H. Estrada, D. Genovese, W. Maisenbacher, B. Heatwole, M. Powell. University of Florida, GAINESVILLE, United States of America

Artificial cardiac pacing is a common procedure performed by veterinary cardiologists. In humans, dual chamber pacing is almost always used in order to allow for physiologic pacing. Despite previous reports demonstrating the feasibility and safety in veterinary medicine, dual chamber pacing is still less commonly used because of concern for longer procedure lengths and increased complication rates related to this. This retrospective study compares implantation times, complication rates and outcome for single and dual chamber pacing at a single institution over a 5 year period.

Medical records of canine patients with clinically significant bradyarrhythmias treated with a single chamber atrial (AAI), single chamber ventricular (VVI), dual chamber single lead (VDD), dual chamber right ventricular apical (DDD-RVA) or dual chamber biventricular (DDD-BiV) pacemakers were reviewed for information regarding pacemaker implantation details, pacemaker type, complications, and survival. Statistical analyses were performed to determine significant differences in procedure time and outcome for dogs divided into groups based on pacing mode (single vs. dual chamber) and lead number (one, two, or three). Fifty four dogs were identified, 28 with single chamber pacemakers (4 AAI, 24 VVI) and 26 with dual chamber pacemakers (5 VDD, 15 DDD-RVA and 6 DDD-BiV). Average procedural time was longer ( $p=0.002$ ) for dual chamber paced patients (135±48 minutes) than single chamber paced patients (96±36 minutes). Mean procedure time lengthened ( $p=0.02$ ) with an increasing number of leads placed (1 lead:

102±50 minutes; 2 leads: 127±38 minutes; 3 leads: 158±9 minutes). Major complications, defined as those considered to be life threatening, causing sudden loss of pacing, or requiring replacement or revision of the pacing system, were not significantly different between pacing modes (log rank  $p=0.90$ ) or between number of pacing leads (hazard ratio = 0.8, 0.26-3.04,  $p=0.85$ ). Minor complications such as seroma formation, higher pacing thresholds, and phrenic nerve stimulation were also not significantly different between pacing mode (log rank  $p=0.13$ ) or number of leads used (hazard ratio= 0.56, 0.15-2.11,  $p=0.39$ ). Mean survival times, including dogs still alive, were 1054 (+/- 705) days and 923 (+/- 506) days for single and dual chamber groups, respectively; this difference was not statistically significant ( $p=0.44$ ).

While dual chamber pacing does increase procedural time, this is not associated with a higher complication rate at our institution. This study supports previous studies which challenge the validity of avoiding dual chamber pacemakers due to an increased complication rate.

**CA-O-6**

**DOES THE ADDITION OF CONSTANT RATE INFUSION TO INTERMITTENT BOLUS ADMINISTRATION OF INTRAVENOUS FUROSEMIDE IMPROVE MEDICAL OUTCOME OF PETS WITH LEFT-SIDED CONGESTIVE HEART FAILURE?** D. Ohad, Y. Segev, S. Klainbart. Koret School of Veterinary Medicine, REHOVOT, Israel

Loop diuretics are a crucial therapeutic component in the management of left-sided congestive heart failure (L-CHF), yet published data about their safety and efficacy in acute L-CHF in veterinary patients are sparse. We sought to investigate whether an addition of constant rate infusion (CRI) to intermittent bolus (IB) administration of intravenous (IV) furosemide has an added benefit to short-term outcome in client-owned pets with acute L-CHF. A retrospective analytic cross-sectional study investigated pets hospitalized between 2003 and 2010. Patients were divided between an IB and an IB+CRI group. Recorded data included serum biochemistry parameters, hospitalization length, mortality, L-CHF recurrence and respiration-related parameters. The IB group consisted of 43 dogs and 16 cats, while the CRI group included 33 dogs and 8 cats. The median cumulative furosemide dose of the CRI group was 0.99 mg/kg/h (range 0.02-3.73,  $n=39$ ), significantly lower than that of the IB group at 1.19 mg/kg/h (range 0.17-7.14,  $n=51$ ) ( $P=0.008$ ). Both treatment groups induced a statistically significantly lower respiratory rate relative to baseline values (from 111.3±44.8 and 104.7±43.6 to 34.9±19.82 and 30.87±12.87 for IB and CRI, respectively;  $p<0.0001$  within each group). The maximal respiratory rates in CRI-dogs were lower compared to IB-dogs ( $p=0.054$ ). The minimal respiratory rate were lower in CRI-cats compared to IB-cats ( $p=0.016$ ). However, no statistically significant difference in respiratory rate parameters was recorded between the two groups, when compared on an hourly basis. Both CRI and IB methods induced minor hypokalemia with no significant difference between groups ( $p=0.564$ ). While serum creatinine increased following both administration methods there was only a borderline significant difference between the two groups in dogs. Serum creatinine following treatment was 1.22±0.4 and 1.5±0.7 mg/dl in the IB and CRI groups, respectively ( $p=0.074$ ). The median length of hospitalization was 2.00 days, and did not differ significantly between dogs and cats or between treatment groups. There was no significant difference in mortality rates between the CRI (14.6%) and the IB (15.3%) groups ( $p=0.93$ ). A significantly higher mortality rate was found for cats (29%) relative to dogs (10.5%,  $p=0.045$ ). Study results could not support the hypothesis that adding CRI to routine IB improves the short-term outcome for pets with L-CHF. Nevertheless, the fact that both treatments reached a similar outcome despite a significantly lower cumulative dose in the CRI group is encouraging. A larger scale, prospective, double-blind random study should be designed with similar cumulative doses in both treatment groups.

**CA-O-7**

**LEFT ATRIAL EJECTION FRACTION ASSESSED BY 3-DIMENSIONAL ECHOCARDIOGRAPHY IN DOGS WITH AND WITHOUT MYXOMATOUS MITRAL VALVE DISEASE.** E.M. Tidholm<sup>1</sup>, A. Bodegård Westling<sup>1</sup>, K. Höglund<sup>2</sup>, I. Ljungvall<sup>3</sup>, J. Häggström<sup>3</sup>. <sup>1</sup>Albano Animal Hospital, DANDE-RYD, Sweden, <sup>2</sup>Dept of Anatomy, Physiology and Biochemistry, UPPSALA, Sweden, <sup>3</sup>Dept of Clinical Sciences, Faculty of Veterinary Medicine, UPPSALA, Sweden

The aim of the study was to assess left atrial ejection fraction (LAEF) in dogs with and without myxomatous mitral valve disease (MMVD) and to correlate LAEF with clinical variables. All dogs underwent clinical examination and 2-dimensional (2D) and real-time 3-dimensional (RT3D) echocardiography. The latter was used to assess left atrial (LA) volumes at atrial end-diastole (LAd) and end-systole (LAS), and the resulting LAEF. A total of 153 dogs, of which 101 dogs had MMVD and 52 were healthy control dogs, were included into the study. Included dogs were of 58 different breeds. Eighty-six dogs (56%) were males. According to the CHIEF classification of congestive heart failure (CHF); 73 dogs were classified without CHF (68 in class BI and 5 in BII) and 28 dogs with CHF (22 in CII and 6 in CIII). Age ranged from 2 months to 18 years in all dogs, with a median of 10 years for MMVD dogs and 4 years for control dogs ( $P < 0.0001$ ). Body weight ranged from 3 to 55 kg in all dogs, with a median of 11 kg for MMVD dogs and 17 kg for control dogs ( $P = 0.009$ ). All dogs were in sinus rhythm, and heart rate (HR) ranged from 67 to 190 beats/min with a median of 120 for both groups.

LAEF ranged from 8 to 74 % in all dogs, with no significant difference between MMVD dogs and control dogs (median 41 and 38%, respectively,  $P = 0.12$ ). LAEF was negatively correlated with increasing LA to aortic ratio (LA/Ao), percentage increase in left ventricular (LV) internal dimension in diastole (LVIDdinc%) and systole (LVIDsinc%) and age for MMVD dogs, and with body weight for control dogs. The final models in the multivariate regression analyses included LVIDdinc% and age for the MMVD dogs, and body weight alone for the control dogs. There was no association between LAEF and gender, HR, mitral E/A ratio or LV ejection fraction assessed by Teichholz method.

In conclusion, LAEF varied widely in both MMVD dogs and in normal control dogs with no significant difference between groups. LAEF decreased with increasing LA and LV volume overload and with increasing age in MMVD dogs, and with increasing body weight in control dogs.

**CA-O-8**

**ACCURACY OF AUSCULTATION ALONE TO IDENTIFY MITRAL INSUFFICIENCY IN ADULT WHIPPETS.** L. Stepien<sup>1</sup>, B. Kellihan<sup>1</sup>, L. Luis Fuentes<sup>2</sup>. <sup>1</sup>University of Wisconsin School of Veterinary Medicine, MADISON, United States of America, <sup>2</sup>Royal Veterinary College, LONDON, United Kingdom

Differentiating between functional murmurs and mitral insufficiency (MI) murmurs by auscultation is challenging and less reliable than echocardiography (ECHO). This study used auscultation and ECHO as a means to identify MI and functional murmurs in a population of dogs predisposed to both. The prevalence of MI and functional murmurs was also recorded.

201 adult whippets (95 male) were examined at dog shows (2005-2009). Median (range) age and weight were 67 (9-209) months and 15 (9.2-22.2) kg. One examiner evaluated murmur intensity (/6) and point of maximal intensity. A different examiner blinded to auscultation findings and dog age recorded the ECHO exam. 'MI' was defined as presence of eccentric or multiple color flow MI jets.

Auscultation identified a heart murmur in 186 dogs (93%) dogs; 58/186 (31%) were left apical systolic (LAS) murmurs (29% overall), and 128/186 (69%) were left basilar systolic murmurs (64% overall). 15 dogs (7%) had no murmur. ECHO identified MI in 76/201 (38%) dogs. The presence of any murmur was highly sensitive (98%) to detect MI, but had low specificity (11%, PPV: 41%, NPV: 93%). Identification of any murmur as LAS was moderately successful at identifying MI (Se: 65%, Sp:

92%, PPV: 84%, NPV 80%). Overall, 49/58 (84%) dogs with LAS murmurs had MI and 39/42 (93%) dogs with grade 3/6 or louder LAS murmurs had MI.

In whippets, absence of a murmur likely indicates absence of MI, but ECHO is required to differentiate mild/moderate MI murmurs from functional murmurs.

**CA-O-9**

**THREE-DIMENSIONAL ECHOCARDIOGRAPHY IMPROVES NONINVASIVE ESTIMATE OF CARDIAC OUTPUT IN HEALTHY DOGS.** Y. Bongrand, M.C. Bélanger. Faculté de Médecine Vétérinaire, University of Montreal, SAINT-HYACINTHE, Canada

Determination of stroke volume and cardiac output (CO) by two-dimensional echocardiography (2DE) is based on assumptions of left ventricular symmetry and geometry. Three-dimensional echocardiography (3DE) overcomes this limitation and its incremental value over 2DE for estimation of stroke volume is well-established in human medicine.

The objective of this study was to compare the accuracy of cardiac output determination by 3DE with the standard 2D method of disk summation (Simpson's rule) in healthy dogs. The lithium dilution cardiac output technique (LiDCO) was used as the reference method for cardiac output measurement.

The study protocol was approved by the Animal Care committee of the University of Montreal. Cardiac output was measured in 20 anesthetized Beagles by use of 3DE, 2DE and the LiDCO technique. Two measures of cardiac output were realized by use of the LiDCO technique immediately prior and following the acquisition of the echocardiogram from the left apical view. Left ventricular stroke volume and heart rate were measured and cardiac output was calculated off-line from 2D images (2DCO) and 3D images (3DCO). The agreement between 2DCO/3DCO and LiDCO was evaluated using Bland-Altman analysis.

The correlation coefficient between 3DCO and LiDCO was 0.76. The mean of the differences of 3DCO minus LiDCO was -0.15 L/min/m<sup>2</sup> (95% limits of agreement: -1.13 to 0.83 L/min/m<sup>2</sup>). Correlation coefficient between 2DCO and LiDCO was 0.73. The mean of the differences of 2DCO minus LiDCO was -0.52 L/min/m<sup>2</sup> (95% limits of agreement: -1.44 to 0.40 L/min/m<sup>2</sup>). The proportion of markedly divergent measures (>20% difference from LiDCO) was significantly lower for 3DE (6/20; 30%) than for 2DE (13/20; 65%) ( $p = 0.039$ ).

Overall, the correlation between the ultrasound-derived estimates of CO and LiDCO was moderate and the limits of agreement were large. 3DE allows in healthy dogs a slight improvement of the accuracy of CO estimate in comparison with the 2D method of disk summation. We predict a heightened benefit of 3DE in presence of left ventricular distortion. Further studies are warranted in canine heart disease to confirm this hypothesis.

**CA-O-10**

**ADIPONECTIN PLASMA CONCENTRATION IN HEALTHY AND HEART FAILURE DOGS.** C. Damoiseaux<sup>1</sup>, N. Gomez<sup>2</sup>, P. Jaspers<sup>2</sup>, A.C. Merveille<sup>1</sup>, C. Clercx<sup>1</sup>, K. Mc Entee<sup>2</sup>. <sup>1</sup>ULg, Faculty of Veterinary Medicine, LIÈGE, Belgium, <sup>2</sup>ULB, Faculty of Medicine, BRUSSELS, Belgium

Adiponectin is an abundant endocrine hormone secreted from adipocytes and paradoxically, inversely correlated to adiposity. By increasing angiogenesis and vasodilation, and by decreasing the cardiac remodeling processes such as hypertrophy, apoptosis, fibrosis and inflammation, adiponectin elicits protective effects in the vasculature and myocardium. In humans, hypoadiponectinemia is associated with coronary artery disease, while hyperadiponectinemia predicts morbidity and mortality in heart failure patients. This paradoxical relationship could be explained by the fact that in overt congestive heart failure, wasting is associated with an increased risk of death.

The present study was designed (1) to determine physiological determinants of adiponectin plasma concentration in dogs and (2) to evaluate adiponectin levels in heart failure dogs. Forty nine adult (range 1-13 years), 19 growing (range 1-11 months) healthy dogs, and 36 dogs in heart failure, classified either according to the etiology or to the ISACHC class (mitral valve disease n=16, dilated cardiomyopathy n=11, other causes n=9; ISACHC1 n = 15, ISACHC2 n = 9, ISACHC3 n = 12) were included. Adiponectin was measured by a canine specific sandwich ELISA kit. The intra-day and inter-day coefficients of variation were respectively 4.4% and 4.1%. Mean adiponectin level in healthy adults was 11.1±1.2 mg/L. Adiponectin plasma concentration decreased linearly with age (-0.6 mg/L per year,  $r=0.37$ ,  $p=0.011$ ). No significant effect of body weight, gender, body score or circadian rhythm was observed. Adiponectin levels were increased in dilated cardiomyopathy (20±4 mg/L  $p<0.01$ ) and tended to decrease in mitral valve disease (6.6±1.4 mg/L,  $p=0.06$ ) compared to healthy adults. Adiponectin was higher in overt heart failure (ISACHC3 17.3±4.1 mg/L  $p<0.05$ ) compared to ISACHC1 and 2 together (10.6±1.8 mg/L). In conclusion, these results demonstrate that, in dogs, adiponectin is age dependent. Moreover, in heart failure dogs, adiponectin levels are disease and stage dependent. Pathophysiological, diagnostic, prognostic and therapeutic implications of these observations should be investigated.

#### CA-O-11

**MEASUREMENT OF PULMONARY TRANSIT TIME IN CATS BY USE OF THE ULTRASOUND CONTRAST MEDIA 'SONOVUE®'. FEASIBILITY, REPRODUCIBILITY, AND NORMAL VALUES.** A. Streitberger, V. Hocke, P. Modler. Traunkreis Vet Clinic Sattledt, SATTLEDT, Austria

Pulmonary transit time (PTT) is a parameter of cardiac performance which has been previously established in dogs and humans. This index is usually normalized to the heart rate (nPTT) according to the formula  $nPTT=PTT/mRR$ , where mRR is the mean RR interval duration. Previous studies in dogs showed that PTT can be measured either by first pass radionuclide technique or echocardiographically by use of the contrast agent SonoVue®.

The objective of this study was to evaluate feasibility of this method in cats, to determine the range of nPTT in healthy cats, and to estimate the reproducibility of nPTT by use of the echocardiographic contrast medium SonoVue®.

This prospective data analysis consisted of 44 cats. All patients underwent complete physical and echocardiographic examination. Each cat received a dose of 0.05-0.06 ml/kg SonoVue® via the cephalic vein. To measure the contrast agent's transit time from the pulmonary artery to the left atrium (PALA) the right parasternal short axis view was used. 52 contrast passage recordings were performed and nPTT was determined independently by 3 examiners of different levels (low, medium, high) in experience. For each cat (n=44) the mean of the nPTT results obtained by the 3 observers was computed and normal values were described by mean +/- standard deviation. Within-day variability was derived from 3 cats in which the contrast passage had been recorded 5 times each, and was described by calculating coefficients of variation for each observer. The inter-observer variability was computed from all measurements (n=52) and was expressed as coefficient of variation. A t-test was used to determine the influence of expertise on nPTT-measurement.

The present study showed that the normal range of nPTT in healthy cats (n=40) is 4.1 +/- 1.02. The inter-observer variability showed that the observer with the lowest experience achieved significantly higher results than the most experienced observer. The median inter observer variation for nPTT (n=45) was 7.7% (interquartile range 4.8% to 14.2%). The within-day variability obtained by the low, medium and high experienced observer of variation was 13.2%, 12.9% and 12.9%, respectively. We did not notice adverse reactions in any of the cats examined.

In summary it is possible to measure nPTT in cats under clinical conditions. Normal values were established. They are comparable to those published in dogs and humans.

#### CA-O-12

**COMPARATIVE BREED-SPECIFIC FEATURES OF FELINE HYPERTROPHIC CARDIOMYOPATHY: A RETROSPECTIVE STUDY OF 344 CASES (2001-2011).** E. Trehiou-Sechi<sup>1</sup>, V. Gouni<sup>1</sup>, R. Tissier<sup>2</sup>, C. Misbach<sup>1</sup>, A. Petit<sup>1</sup>, C. Carlos Sampedrano<sup>1</sup>, D. Balouka<sup>1</sup>, J.L. Pouchelon<sup>3</sup>, V. Chetboul<sup>3</sup>. <sup>1</sup>Unité de Cardiologie d'Alfort, CHUVA, ENVA, MAISONS-ALFORT, France, <sup>2</sup>UMR INSERM U955; Unité de Pharmacie-Toxicologie, ENVA, MAISONS-ALFORT, France, <sup>3</sup>Unité de Cardiologie d'Alfort, CHUVA; UMR INSERM U955, ENVA, MAISONS-ALFORT, France

Primary hypertrophic cardiomyopathy (HCM) is the most common feline heart disease, which has been demonstrated to be inherited in some specific breeds. However, few studies have compared feline HCM phenotypes and survival according to breed.

The aim of this retrospective study was to compare epidemiological characteristics, clinical findings, left ventricular (LV) geometric patterns, and prognosis according to breed in a large population of HCM cats.

The case records of client-owned cats of 5 different breeds (Domestic shorthair (DS), Persian, Maine coon (MC), Sphynx, and Chartreux), diagnosed with primary HCM (2001-2011) at the Cardiology Unit of Alfort, were reviewed.

The study population consisted of 344 cats (mean±SD, age=7.4±4.7 years, body weight=4.9±1.4 kg, 71% males and 29% females) including 239 DS, 41 Persians, 28 MC, 22 Sphynx, and 14 Chartreux.

Age at the time of diagnosis was significantly lower ( $p<0.001$ ) in MC (median age=2.5 years) and Sphynx (3.5 years) compared to other breeds (8.0, 8.0, and 11.0 years for Chartreux, DS and Persians, respectively).

Sphynx with HCM had a significantly higher ( $p=0.043$ ) heart rate (203±31 bpm) compared to the whole study population (189±30 bpm), resulting in a shorter isovolumic relaxation time ( $p=0.018$ ). The prevalence of LV outflow tract obstruction was higher ( $p<0.05$ ) in Persians (23/41; 56%) compared to other breeds (115/303; 38%).

The proportion of cats that underwent a cardiac event (congestive heart failure, aortic thromboembolism (ATE), syncope, or sudden death (SD)) differed significantly between the 5 breeds: 39%, 33%, 12%, 9%, and 7% for MC, DS, Persians, Sphynx, and Chartreux, respectively. The type of cardiac event was also different among breeds ( $p=0.048$ ), with no ATE in MC, Sphynx, and Chartreux cats. Age at the first cardiac event was significantly lower ( $p=0.007$ ) in MC (median age=2.5 years) than in other breeds (7.0 years).

At the time of writing, 124/344 cats are dead. The cause of death differed significantly according to the breed ( $p=0.036$ ) and to the clinical status at the time of diagnosis ( $p<0.0001$ ). Most cats that died and were asymptomatic at the time of diagnosis (n=89) died from non-cardiac causes (71/89, 80%). Sudden death (representing 24% of all cardiac deaths) was only observed in 3 breeds (DS, MC, and Sphynx).

In conclusion, as in human HCM, feline HCM is characterized by marked phenotypic variability with several specific breed-dependent features regarding epidemiology, age at the time of diagnosis and decompensation, and LV geometric patterns.

#### CA-O-13

**CONGESTIVE HEART FAILURE IS NOT A PRIMARY CAUSE OF COUGHING IN DOGS WITH CHRONIC DEGENERATIVE MITRAL VALVE DISEASE (MMVD).** L. Ferasin<sup>1</sup>, L. Crews<sup>2</sup>, D.S. Biller<sup>3</sup>, K.E. Lamb<sup>4</sup>, M. Borgarelli<sup>3</sup>. <sup>1</sup>SVCC Ltd, NEWBURY, United Kingdom, <sup>2</sup>Radiology Vet Consulting, Inc., TRENTON, ONTARIO, Canada, <sup>3</sup>Kansas State University, MANHATTAN, KS, United States of America, <sup>4</sup>University of Florida, GAINESVILLE, FL, United States of America

**Introduction:** Coughing is an important physiological mechanism that maintains healthy respiratory function by removing potentially harmful substances from the airways. Stimulation of

the cough reflex does not occur in deeper airways or respiratory parenchyma (smaller bronchi, bronchioles, alveoli). According to this observation, cardiogenic pulmonary oedema (congestive heart failure; CHF) should not be an expected cause of cough. The aim of our study was to investigate the association between presence of coughing and potential causes of cough, such as CHF, airway disease and cardiomegaly in dogs affected by naturally acquired MMVD.

**Methods:** Clinical records of 204 dogs affected by MMVD that underwent full cardiac evaluation were retrospectively reviewed. Exclusion criteria included incomplete records, equivocal diagnosis, concomitant presence of other cardiac disease, administration of antitussives and/or furosemide in the 24h prior to cardiology consultation. Echocardiographic and radiographic interpretations were reviewed in a blind fashion by 2 boarded cardiologists and 2 boarded radiologists respectively. Univariate and multivariate logistic models were utilised to assess the patient's likelihood of exhibiting coughing. Interaction models were also generated to address the interaction of increased left atrial (LA) size and presence of airway disease.

**Results:** Univariate analyses showed that CHF is not a predictor of coughing (OR = 1.27; 0.66, 2.45), while airway disease (OR = 3.85; 2.15, 6.88) and increased LA size (OR = 3.48; 2.15, 6.88) enhance the risk of coughing in dogs with MMVD. The same risk factors were significant in multivariate analyses. While compelling to suggest that a divergent interaction effect is present, the interaction terms were non-significant for the enlarged LA by airway disease ( $P=0.3992$ ) suggesting that both of these main effects are acting statistically independently.

**Conclusion:** This study supports our hypothesis that CHF is not a primary cause of coughing in dogs with MMVD. Instead, airway disease and cardiomegaly represent important risk factors for the development of cough in these patients. This important finding should be taken into account when considering diagnosis and clinical management of CHF in dogs with MMVD.

**CA-O-14**  
**SELF-REPORTED TWO-DIMENSIONAL ECHOCARDIOGRAPHIC ESTIMATES OF LEFT ATRIAL SIZE BY CARDIOLOGISTS: COMPARISON WITH MEASURED VARIABLES.** R. Georgiev<sup>1</sup>, M. Rishniw<sup>2</sup>, D. Ohad<sup>3</sup>. <sup>1</sup>Central Vet Clinic, SOPHIA, Bulgaria, <sup>2</sup>Cornell University, ITHACA, United States of America, <sup>3</sup>Hebrew University of Jerusalem, REHOVOT, Israel

Two dimensional measurements of left atrial (LA) size are routinely obtained in echocardiographic evaluations, and often normalized to the aortic (AO) dimensions but clinicians provide subjective assessments of LA size in clinical reports. How these assessments compare to objective measurements is not known.

We obtained LA and AO linear and area dimensions for 98 dogs and 20 cats with heart diseases that can result in LA enlargement from 21 cardiologists along with their subjective assessments of LA size (none, mild, moderate, severe). We compared the LA:AO and LAarea:AOarea with the self-reported subjective assessments. Additionally, we examined inter-operator variability in obtaining LA:AO and LAarea:AOarea from a single series of echocardiographic images.

LA:AO differed between subjectively classified groups, except for mild and moderate classifications, which were not different from each other. Overlap existed across all but the most extreme groups. LAarea:AOarea did not differ between normal and mild classifications or moderate and severe classifications. Overlap existed across all groups. LA:AO in cats separated better than in dogs.

Interobserver comparison of measurement of LA:AO and LAarea:AOarea showed that 95% of the LA:AO estimates fell within  $\pm 0.3$  and 95% of the LAarea:AOarea estimates fell within  $\pm 1.2$ , with no evidence of bias.

Our data show that objective measures of LA:AO and LAarea:AOarea differ between subjective classifications of LA size,

but considerable overlap exists. Trained echocardiographers measure the LA and AO similarly from identical images.

**CA-O-15**  
**A PROPOSED MECHANISM OF INHERITANCE OF HYPERTROPHIC CARDIOMYOPATHY IN A FAMILY OF BRITISH SHORTHAIRED CATS.** C. Chervier<sup>1</sup>, T. Ribas<sup>1</sup>, C. Burnichon<sup>1</sup>, A. Thomas<sup>2</sup>, L. Chabanne<sup>1</sup>, J.L. Cadore<sup>1</sup>, I. Bulbot<sup>1</sup>. <sup>1</sup>Veterinary Campus, MARCY L'ÉTOILE, France, <sup>2</sup>Antagene, DARDILLY, France

**Aims of the work:** Hypertrophic cardiomyopathy (HCM) is a common disease in cats. Despite being mainly encountered in European cats, many purebred cats may also be affected (Maine Coon, Ragdoll, Persians, American and British Shorthairs). A genetic basis to the disease with an autosomal dominant mode of inheritance has been shown in the Maine Coon and Ragdoll breeds and 2 different associated genetic mutations in the MyBPC3 gene have been discovered in these breeds. The goal of our study was to determine the mode of inheritance of HCM in a population of British Shorthair cats.

**Material and method:** The group of cats consisted of 7 males and 21 females that were variably related. All cats underwent physical examination including cardiac auscultation and one echocardiographic procedure either in 2006 or in 2009. Four cats that were seen in 2006 underwent a second echocardiographic examination in 2009, owing to equivocal status determined after initial examination. Based on the echocardiographic data, the cats were classified as affected, equivocal or normal (free of HCM).

Considering the status of the cats (affected, equivocal, or normal), the family tree analysis was undertaken in order to propose a mode of inheritance of HCM in this breed.

**Results:** Echocardiographic results were compatible with HCM in 2 males and 4 females; 1 male and 5 females were considered equivocal and 4 males and 12 females were classified normal. Both obstructive and non-obstructive forms of HCM were observed in these cats.

Status of offsprings (affected, equivocal, or normal) and family tree analysis allowed us to exclude a sex-linked as well as an autosomal recessive mode of transmission. An autosomal dominant mode of inheritance of HCM in the British Shorthair was then proposed.

**Conclusion:** The first goal of the present study was to propose a mode of inheritance of HCM in British Shorthair cats. In accordance with those results presented by Putcuypis et al at the ACVIM in 2003 an autosomal dominant mode of inheritance was hypothesized in the British Shorthair cats enrolled in our study.

The next step of this study will be to identify a possible mutation causing HCM in this breed, since the mutations on the MyBPC3 gene discovered in the Maine Coon and Ragdoll cats is known to be not responsible for HCM in the British Shorthair cat.

**CA-O-16**  
**COMPARISON OF TWO ECHOCARDIOGRAPHIC ESTIMATES OF AORTIC VALVE DIAMETER IN DOGS AND CATS.** R. Georgiev<sup>1</sup>, M. Rishniw<sup>2</sup>. <sup>1</sup>Central Vet Clinic, SOPHIA, Bulgaria, <sup>2</sup>Cornell University, ITHACA, United States of America

Measurements of cardiac chambers and walls are often normalized for bodyweight by comparing them to aortic valve measurements, allowing clinicians to develop weight-independent estimates of chamber enlargement. Two-dimensional echocardiographic measurements of the left atrium (LA) and aorta (AO) have been proposed as more accurately reflecting LA size than M-mode measurements. Reference intervals for LA:AO in healthy dogs have been proposed independently by 2 groups of investigators, however, these two groups used slightly different methods to measure the AO and obtained different reference intervals of LA:AO in healthy dogs. To determine if these reference intervals differed because of the different methods used to

measure the AO, we compared these 2D methods of measuring the AO.

We examined right-parasternal short-axis echocardiographic images from 273 dogs and 99 cats with various cardiac diseases. The AO was measured using both methods in the same end-systolic frame. Each measurement was obtained without consideration of the other measurement. The two methods were compared using Limits-of-Agreement analysis.

In dogs and cats, both methods agreed with each other, with regressions of the lines and intercepts not different from zero. Slight heteroscedasticity was apparent in both dogs and cats. 95% limits of agreement were within 2mm for dogs across most measurements and 1mm for cats.

Our data show that 2D echocardiographic estimates of aortic dimension by these methods are similar and interchangeable. Therefore, the differences in LA:AO reference intervals for healthy dogs previously observed cannot be accounted for by differences in AO measurements.

#### CA-O-17

**ECHOCARDIOGRAPHIC ASSESSMENT OF REGIONAL LEFT ATRIAL DEFORMATION PROPERTIES USING TISSUE DOPPLER IMAGING: A FEASIBILITY STUDY ON HEALTHY DOGS.** M. Baron Toaldo<sup>1</sup>, C. Guglielmini<sup>2</sup>, G. Sgreccia<sup>1</sup>, A. Diana<sup>1</sup>, M. Cipone<sup>1</sup>. <sup>1</sup>Faculty of Veterinary Medicine, University of Bologna, OZZANO EMILIA (BO), Italy, <sup>2</sup>Department of Veterinary Clinical Sciences, University of Padova, LEGNARO (PD), Italy

Strain (St) and strain rate (SR) imaging are ultrasound modalities that have been employed to assess the global and regional canine left ventricular mechanical function in normal and pathological conditions. To the best of the author's knowledge, the use of tissue Doppler imaging (TDI) for the study of left atrial (LA) deformation has never been described in the dog.

The aims of the present study were: to assess the feasibility, describe the patterns and determine the reproducibility of measuring TDI-based velocity, St and SR of the LA walls in a group of normal dogs.

Ten different TDI variables (peak velocity, St and SR, during systole, early diastole, and late diastole, and Time-to-peak) were recorded in ten healthy, unsedated dogs of different breeds and sizes. These variables were measured using the left apical 4-chambers and 2-chambers views in four different LA walls: interatrial septum and lateral, anterior and posterior walls. The data obtained from the ten dogs for each atrial segment were expressed as mean values and standard deviations. Test reproducibility was determined by estimating the within-day and between-day intra-observer variability in one dog. Variability was expressed as coefficient of variation (CV) and the absolute value below which the difference between two measurement will lie with 95% probability. The degree of variability was arbitrarily defined as follows: < 5%, very low variability; 5-15%, low variability; 16-25%, moderate variability; >25%, high variability.

The measurement of the ten variables was feasible in all dogs. LA tissue Doppler profiles showed constant patterns in all dogs. Tissue velocity and SR had peak positive values during ventricular systole, and two distinct peaks with negative values during early and late diastole. Atrial St presented peak positive values during systole, positive values during early diastole, and a negative peak during late diastole. The within-day intra-observer variability was very low for 24/40 of the analyzed variables, low for 8/40 variables, moderate for 4/40 variables and high for only 2/40 variables. The between-day intra-observer variability was very low for 26/40 of the analyzed variables, low for 13/40 variables and moderate for only 1/40 variable.

Left atrial TDI-based longitudinal deformation imaging was feasible and reproducible in healthy dogs. These preliminary data need additional validation to be used as reference values. The application of Sr and SR on the canine left atrium might be useful for a better understanding of atrial pathophysiology in different cardiac diseases in dogs.

#### CA-O-18

**THE EFFECT OF PIMOBENDAN ON THE CLINICAL OUTCOME AND SURVIVAL OF CATS WITH NON TAURINE RESPONSIVE DILATED CARDIOMYOPATHY.** L.E. Hambrook, P. Bennett. Melbourne Veterinary Specialist Centre, GLEN WAVERLEY, Australia

Non taurine responsive feline dilated cardiomyopathy (DCM) has a poor prognosis and a reported median survival time (MST) of 11 days. Pimobendan, a positive inotrope, has a beneficial effect on the survival of dogs with DCM, but its effect on the survival of cats with DCM is unknown. This retrospective study was designed to assess the effect of pimobendan on the MST of cats with non taurine responsive DCM.

The medical records of cats diagnosed with DCM at the Melbourne Veterinary Specialist Centre in Australia between 2001 and 2010 were reviewed. Thirty five cats met the inclusion criteria, which consisted of a left ventricular end systolic dimension of >14mm, a fractional shortening of <28% and an intention to treat. Three cats were subsequently excluded because of a clinical and echocardiographic response to taurine (n=2) and a lack of follow up (1).

Presenting signs included respiratory distress (n=28), aortic thromboembolism (ATE, 3) and collapse (1). All 32 cats received standard treatment with frusemide, taurine and benazepril. Pimobendan (Vetmedin, Boehringer Ingelheim, 0.19-0.36mg/kg PO q 12 hours) was administered to 80% of cats (16/20) diagnosed after 2005. Of the 16 cats that did not receive pimobendan, 9 were treated with digoxin. All 32 cats demonstrated recurrent clinical signs and/or echocardiographic evidence of DCM despite treatment. The cats were divided into pimobendan (n=16) and non pimobendan (n=16) groups for analysis of survival data.

All cats in the non pimobendan group died. Twelve were euthanized due to refractory congestive heart failure (8/16) or ATE (4/16) and 4 died suddenly. Of the 16 cats in the pimobendan group 15 died. One cat remained alive (502 days after diagnosis) and it was censored from the outcome analysis. Eight cats were euthanased due to refractory congestive heart failure (n=5), ATE (2) and seizure activity (1). Sudden death was observed in 7 cats.

The MST of the pimobendan group (49 days; range 1 to >502 days) was four times that of the non pimobendan group (12 days; 1 to 244 days). The difference in survival between the two groups was statistically significant (P=0.048). Hypothermia (<37.2°C) and a fractional shortening of <20% were associated with a poor prognosis. No adverse effects of pimobendan therapy were noted.

Pimobendan therapy appears to be well tolerated in cats and it may extend the survival times of cats with DCM. Further investigation of the possible benefits of pimobendan therapy in cats is warranted.

#### CA-O-19

**2D-SPECKLE TRACKING ECHOCARDIOGRAPHY (STE) FOR ASSESSMENT OF GLOBAL AND REGIONAL LEFT VENTRICULAR PEAK SYSTOLIC STRAIN AND STRAIN RATE IN HEALTHY LABRADOR RETRIEVER DOGS.** C. Carnabuci<sup>1</sup>, S. Hanås<sup>2</sup>, I. Ljungvall<sup>1</sup>, A. Tidholm<sup>3</sup>, C. Bussadori<sup>4</sup>, J. Häggström<sup>1</sup>, K. Högglund<sup>1</sup>. <sup>1</sup>Swedish University of Agricultural Sciences, UPPSALA, Sweden, <sup>2</sup>Strömsholm Animal Hospital, STRÖMSHOLM, Sweden, <sup>3</sup>Albano Animal Hospital, DANDERYD, Sweden, <sup>4</sup>Clinica Veterinaria Gran Sasso, MILANO, Italy

Two-dimensional speckle tracking echocardiography (STE) is a new angle-independent echocardiographic modality for quantifying global and regional myocardial motion. The aim of the study was to assess left ventricular (LV) global and regional longitudinal (L), circumferential (C) and radial (R) peak systolic strain (St) and strain rate (Sr) STE variables, including their feasibility, and variability, and LV synchronicity in healthy Labrador retriever dogs. Nineteen clinically healthy Labradors: 15 females and 4 males with (mean ± SD) age 6.6 ± 3.7 years, body-weight 28.6 ± 4.2 kg, and heart rate 107 ± 15 beats per minute were included. Cine-loops were acquired from left apical 4 chamber view and right parasternal short axis view at basal and apical

levels with a 5-2 MHz probe for MyLab30TM Gold system (Esaote, Florence) and analyzed with regards to St and Sr using the XStrainTM software. In addition, synchronicity time index (STI) and strain-derived dyssynchrony index (SDI) were assessed. For all variables, a mean of 3 beats was used for the statistical analysis. 86% of the 912 segments was analyzed: 14% were excluded, all longitudinal, due to echo-drop out or artifacts. A general epicardial to endocardial St gradient was found (all  $P < 0.01$ ). The CSt variables were significantly higher at apex compared to base ( $P < 0.05$ ). Radial and C regional homogeneity was found, except for CSt epicardial segments at base and apex. A LSt base to apex gradient was found. Apical RSt and RSr were positively correlated with heart rate. Including dogs in which all the segments were obtainable ( $n=12$ ), the coefficient of variation was lower for the LSt and LSr variables compared to corresponding C variables. Synchronicity results for STI and SDI were  $35.72 \pm 21.64$  ms and  $16.17 \pm 10.5$  ms, respectively. This study shows that STE assessment of global and regional systolic St and Sr was possible in all dogs for C and R variables, and in a majority of L variables. Longitudinal variables had lower feasibility, but also lower variability. These and other results of this study could be of value for future STE studies.

#### CA-O-20

**CHANGES IN RADIAL AND LONGITUDINAL SEGMENTAL MYOCARDIAL DEFORMATION IN DOGS WITH MYXOMATOUS MITRAL VALVE DISEASE DETECTED BY USE OF TWO-DIMENSIONAL SPECKLE TRACKING ECHOCARDIOGRAPHY.** N.E. Zois<sup>1</sup>, A. Tidholm<sup>2</sup>, K.M. Nägga<sup>1</sup>, S.G. Moesgaard<sup>1</sup>, C.E. Rasmussen<sup>1</sup>, T. Falk<sup>1</sup>, J. Häggström<sup>3</sup>, H.D. Pedersen<sup>4</sup>, L.H. Olsen<sup>1</sup>. <sup>1</sup>University of Copenhagen, FREDERIKSBERG, Denmark, <sup>2</sup>Albano Animal Hospital, DANDERYD, Sweden, <sup>3</sup>Swedish University of Agricultural Sciences, UPPSALA, Sweden, <sup>4</sup>Novo Nordisk A/S, MAALOEV, Denmark

Newer echocardiographic methods such as speckle tracking (2DStrain) provide quantitative assessment of regional systolic and diastolic myocardial deformation. In myxomatous mitral valve disease (MMVD), conventional echocardiographic indices of myocardial function are preserved until late stages. However, dogs often develop congestive heart failure (CHF) and experimental studies have indicated contractile dysfunction. Thus, the aim of this study was to investigate the segmental peak radial (R) and longitudinal (L) strain (S), strain rate in systole (SRs), in early (SR<sub>e</sub>) and late (SR<sub>a</sub>) diastole in dogs with different severities of MMVD using 2DStrain. The study included 102 dogs: 20 Beagles with no/minimal mitral regurgitation (MR), 61 Cavalier King Charles Spaniels (CKCS) with increasing severity of MR, 14 CKCS (CHF-CKCS) and 7 dogs of different breeds (CHF-MIX) with clinical signs of CHF due to MMVD. Using a Vivid-i ultrasound system, all dogs underwent echocardiography including 2D grayscale images for offline analysis of 2DStrain by use of commercially available software (EchoPAC). By use of Mann-Whitney U test, Beagles and CKCS with no/minimal MR were compared, as were CKCS-CHF compared to CHF-MIX, without finding differences in 2DStrain variables between groups. Including all dogs, Spearman's rank correlations with a Bonferroni corrected P-value ( $P < 0.008$ ) were computed between 2DStrain variables and age, heart rate (HR), left atrial-to-aortic root ratio (LA/Ao) and MR. Radial S increased with MR in 5/6 myocardial segments ( $P < 0.002$ ) and with LA/Ao in 4/6 segments ( $P < 0.003$ ). Radial SRs significantly increased with MR in 5/6 segments ( $P < 0.006$ ), however significant increase with HR was also found for these segments ( $P < 0.002$ ). Radial SR<sub>e</sub> increased with LA/Ao in 6/6 segments ( $P < 0.002$ ) and with MR in 4/6 segments ( $P < 0.001$ ). In one segment, SR<sub>a</sub> also increased with HR ( $P = 0.007$ ). Likewise, LS significantly increased with LA/Ao in 3/6 segments, with MR in 2/6 segments (all  $P < 0.0003$ ) and with age in one segment ( $P = 0.006$ ). Longitudinal SRs increased with HR in all segments ( $P < 0.0003$ ) and LA/Ao and MR in one segment ( $P < 0.003$ ). Longitudinal SR<sub>e</sub> and LS<sub>Ra</sub> increased with MR and LA/Ao in 2/6 myocardial segments (all  $P < 0.0008$ ) but also with HR and age ( $P < 0.003$ ) in one of these segments. In conclusion, 35/48 2DStrain variables increased with indices of MMVD.

Systolic radial and longitudinal deformation as well as radial deformation rate in systole and early diastole increased with increasing MR while systolic deformation rates were strongly correlated to HR.

#### CA-O-21

**RELATION OF VITAMIN D STATUS TO CONGESTIVE HEART FAILURE AND INCIDENT CARDIOVASCULAR EVENTS IN DOGS.** S. Kraus<sup>1</sup>, M. Rassnick<sup>1</sup>, J. Wakshlag<sup>1</sup>, R.M. Gelzer<sup>1</sup>, S. Waxman<sup>1</sup>, A. Struble<sup>1</sup>, L. Tian<sup>2</sup>. <sup>1</sup>Cornell University, ITHACA, United States of America, <sup>2</sup>State University of New York at Buffalo, BUFFALO, NY, United States of America

Vitamin D plays a pivotal role in cardiac function and there is increasing evidence that vitamin D deficiency contributes to the etiology and pathogenesis of congestive heart failure (CHF) in people. We examined the association between circulating 25-hydroxyvitamin D [25(OH)D], the hallmark of vitamin D status, and CHF in dogs. A further objective was to determine the impact of 25(OH)D status on clinical outcome in dogs with CHF. Serum 25(OH)D concentrations were examined in 82 client-owned dogs, including 31 with CHF and 51 unaffected controls. The relationship between CHF-cases and controls and 25(OH)D level, age, and body condition score were evaluated using univariate and multivariate analyses. Potential differences in vitamin D oral intake, calculated on the basis of a dietary questionnaire, were also evaluated between groups. The effect of 25(OH)D level on time to another cardiovascular event was evaluated by use of Cox Regression and Kaplan-Meier methods. Values of  $P \leq 0.05$  were considered significant. Mean 25(OH)D concentration ( $100 \pm 44$  nmol/L) in dogs with CHF was significantly lower than that of unaffected dogs ( $123 \pm 42$  nmol/L). The mean calculated vitamin D intake (nutrition) per kg body weight in dogs with CHF was actually significantly higher than that of unaffected dogs ( $0.67 \pm 0.49$   $\mu\text{g}/\text{kg}$  body weight versus  $0.42 \pm 0.27$   $\mu\text{g}/\text{kg}$  body weight, respectively). There was a significant effect of 25(OH)D level on time to another episode of CHF. These findings suggest low levels of 25(OH)D might be a risk factor for CHF in dogs.

#### GA-O-1

**SHOULD CHLORAMBUCIL-PREDNISOLONE COMBINATION THERAPY BE USED IN PREFERENCE TO AZATHIOPRINE-PREDNISOLONE FOR THE TREATMENT OF CHRONIC ENTEROPATHIES WITH CONCURRENT PROTEIN LOSING ENTEROPATHY?** J.R.S. Dandrieux, P.J.M. Noble, A.J. German. University of Liverpool, WILLASTON, United Kingdom

Protein losing enteropathy (PLE), a syndrome characterised by intestinal loss of proteins, can occur secondary to chronic enteropathy (CE; including inflammatory bowel disease), lymphoma or lymphangiectasia. Such cases usually have a guarded prognosis and treatment can be challenging. The aim of this retrospective study was to compare treatment protocols, for CE and concurrent PLE, using prednisolone in conjunction with either azathioprine or chlorambucil.

Dogs presenting over a 3-year period were included, if diagnosed with CE and concurrent hypoalbuminaemia (albumin  $< 18.0$  g/L), after complete gastrointestinal investigations including intestinal biopsy. During this time, cases received either azathioprine-prednisolone (group A,  $n=13$ ), or chlorambucil-prednisolone combination (group C,  $n=14$ ). Response to treatment was assessed by weight gain, increase in circulating albumin, and survival time.

No significant pre-treatment differences were noted, between groups, for any baseline parameter (signalment and weight), for clinicopathological parameters (albumin, cobalamin, and folate concentrations), and for histopathology. After therapy, serum albumin concentration ( $P=0.004$ ) and weight gain ( $P=0.003$ ) were significantly greater in group C. Median survival time for group A dogs was 30 days (confidence interval 15-45 days) and was not

reached for group C dogs. Using Cox's proportional hazards regression, survival was positively associated with the histopathological presence of lacteal dilatation ( $P=0.012$ ) and use of chlorambucil ( $P<0.001$ ).

This study suggests that a chlorambucil-prednisolone treatment protocol conveys a survival advantage for CE and concurrent PLE. Given these findings, a prospective randomised clinical trial is now warranted.

**GA-O-2**  
**EVALUATION OF THE EFFECTS OF METOCLOPRAMIDE AND CISAPRIDE ON CANINE GASTRIC, SMALL BOWEL AND COLONIC MOTILITY USING A WIRELESS MOTILITY CAPSULE.** F.P. Gaschen, C.T. Mole, R.W. Stout. Louisiana State University, BATON ROUGE, United States of America

Prokinetic drugs are commonly used to optimize gastrointestinal motility in dogs. Metoclopramide (M) and cisapride (C) are serotonergic prokinetics which unfold their effects through stimulation/antagonism of different serotonin receptors on post-ganglionic cholinergic neurons. The goal of the present study was to evaluate the effects of therapeutic doses of M and C in healthy dogs using a wireless motility capsule (WMC) equipped with pH, pressure and temperature sensors (SmartPill pHp).

The protocol was approved by the LSU institutional animal care and use committee. Six healthy dogs were used. After baseline tracings were collected dogs were administered M and C orally at 0.5 mg/kg TID for 2 days at prior to undergoing the WMC study, and treatment was continued until the capsule was expelled with feces. Transit times (gastric emptying time (GET), small and large bowel transit times [SBTT, resp. LBTT]) were determined. Parameters evaluated to measure whole gastric, gastric antral, small bowel and colonic motility included contraction frequency (CF) and motility index (MI, which represents area under the curve divided by time) and were obtained using proprietary software. Results were analyzed with ANOVA followed by pairwise testing with appropriate post-hoc corrections.

Neither M nor C had any influence on GET while M significantly increased the gastric antral MI (mean 292 mmHg\*s/min.; 95% CI 212-373) compared to baseline (153; 73-244) and C (146; 65-226),  $p=0.049$  and  $0.038$  resp. SBTT and LBTT were not affected by any drug. Small bowel MI was significantly higher in dogs receiving C (863; 610-1117) compared to baseline (276; 22-529),  $p=0.009$ . Colonic MI remained unchanged after administration of C and M. Finally; neither M nor C significantly changed CF in any studied segment.

Significant increases in antral motility in dogs receiving M and in small bowel motility in dogs administered C were demonstrated in spite of limitations associated with the small group size, and the individual day-to-day variability of GI transit times and motility parameters in dogs. Generally, the wireless motility capsule was useful for non-invasive detailed evaluation of GI motility in healthy dogs.

**GA-O-3**  
**DECREASE IN TOLL- AND NOD-LIKE RECEPTOR EXPRESSION IN POST-TREATMENT DUODENAL MUCOSAL TISSUE FROM DOGS WITH CHRONIC ENTEROPATHY.** A. Lynch, I.R. Peters, E.J. Hall, M.J. Day. University of Bristol, BRISTOL, United Kingdom

Canine chronic enteropathy (CCE) includes a spectrum of disease that may be categorised by response to treatment as food-responsive, antibiotic-responsive or steroid-responsive. CCE is believed to result from failure of normal immunological regulatory mechanisms controlling the maintenance of intestinal homeostasis.

The intestinal microbiota is essential for development of healthy gut-associated lymphoid tissue. Structurally conserved microbial-associated molecular patterns are detected by the pat-

tern-recognition receptors (PRRs) such as Toll-like receptors (TLRs) and NOD-like receptors (NLRs) of intestinal epithelial and dendritic cells. Under conditions of intestinal homeostasis, activation of these PRRs drives a tolerogenic response. In the presence of pathogens, and in the absence of appropriate regulation, activation of the PRRs can stimulate a pro-inflammatory response. Studies in man and dogs indicate that mutations in PRRs, and dysregulation of their interactions with the microbiota, may be implicated in the development of CE.

The aim of this study was to compare the expression of mRNA encoding TLRs 1-10 and NLRs 1 and 2 in endoscopic biopsies of the duodenal mucosa of 12 dogs before and after successful treatment for CCE (food-responsive:  $n = 10$ , antibiotic-responsive:  $n = 1$ , steroid-responsive:  $n = 1$ ), using each dog as its own control. Each dog was scored on the CCE clinical activity index (CCECAI), and for histopathological changes according to WSAVA guidelines. Tissue surplus to diagnostic requirements was used in this work.

Total RNA was isolated from duodenal biopsies and assessed for quality and quantity by automated electrophoresis. TLR 1-10 and NLR 1-2 mRNA expression was measured by quantitative real-time RT-PCR. Expression was normalised using four stably expressed housekeeper genes (GAPDH, SDHA, TBP and YWAZ). For each sample, a relative copy number was calculated for each gene. Expression differences before and after treatment were assessed using Student's paired t test.

Expression of TLRs 2, 4, 5, 9 and 10 and NOD2 was significantly decreased after treatment compared with expression before treatment. These changes were associated with a decrease in the CCECAI and histopathological scores.

Results of this study indicate that up-regulation of expression of genes encoding TLRs 2, 4, 5, 9 and 10 and NOD2 may play a role in the development of inflammation associated with CCE. Further studies are required with greater numbers of dogs to determine whether the CCECAI, histopathological score and PRR gene expression patterns are associated with specific disease phenotypes within the CCE spectrum.

**GA-O-4**  
**DECREASE IN CYTOKINE GENE EXPRESSION IN POST-TREATMENT DUODENAL MUCOSAL TISSUE FROM DOGS WITH CHRONIC ENTEROPATHY.** A. Lynch, I.R. Peters, E.J. Hall, M.J. Day. University of Bristol, BRISTOL, United Kingdom

Canine chronic enteropathy (CCE) includes a spectrum of disease that may be categorised by response to treatment as food-responsive, antibiotic-responsive or steroid-responsive. CCE is believed to be multifactorial in nature and to result from dysregulation of the interactions between the gut-associated lymphoid tissue, the intestinal microbiota and dietary antigens, leading to development and perpetuation of an uncontrolled inflammatory cascade.

In the presence of altered luminal microbiota and weakened intestinal barrier function, and in the absence of appropriate immunological regulation, activation of pattern recognition receptors expressed on intestinal epithelial and dendritic cells can stimulate the innate and adaptive immune systems to switch from the production of tolerogenic cytokines and chemokines to increase the production of those associated with inflammation. Such changes have been implicated in the development of idiopathic chronic enteropathy in man and in experimental rodent models.

The aim of this study was to compare the expression of mRNA encoding selected cytokines and chemokines in endoscopic duodenal mucosal biopsies in 12 dogs before and after successful treatment for CCE (food-responsive:  $n = 10$ , antibiotic-responsive:  $n = 1$ , steroid-responsive:  $n = 1$ ), using each dog as its own control. Each dog was scored on the CCE clinical activity index (CCECAI), and for histopathological changes in samples of duodenal mucosa according to WSAVA guidelines. Tissue surplus to diagnostic requirements was used in this work.

Total RNA was isolated from the biopsies and the quality and quantity of RNA was assessed by automated electrophoresis. Gene expression was measured by quantitative real-time RT-PCR. Expression was normalised using four stably expressed housekeeper genes (GAPDH, SDHA, TBP and YWAZ). For each sample, a relative copy number was calculated for each gene. Expression differences before and after treatment were assessed using Student's paired *t* test.

Expression of IL-5, IL-10, IL-12p35, IL-13, IL-17C, IL-27, RANTES/CCL5, eotaxin-2/CCL24, TGF- $\alpha$  and IFN- $\gamma$  after treatment was significantly decreased compared with expression before treatment. This was associated with a decrease in the CCECAI and histopathological scores.

Results of this study suggest that treatment of CCE may be associated with a change in the expression of genes encoding particular cytokines and chemokines in the duodenal mucosa and supports extension of this work in a larger cohort of patients of defined disease phenotypes.

#### GA-O-5

**PHYLOGENETIC COMPARISON OF DUODENAL BACTERIA IN CATS WITH FOOD RESPONSIVE ENTEROPATHY AND INFLAMMATORY BOWEL DISEASE USING 16S rRNA GENE PYROSEQUENCING.** R. Gostelow<sup>1</sup>, J. Suchodolski<sup>2</sup>, J.M. Steiner<sup>2</sup>, K.Smith<sup>1</sup>, S.E. Dowd<sup>3</sup>, K. Allenspach<sup>1</sup>. <sup>1</sup>The Royal Veterinary College, Hatfield, United Kingdom, <sup>2</sup>Texas A&M University, College Station, United States of America, <sup>3</sup>Research and Testing Laboratory, Lubbock, United States of America

The intestinal microbiome likely plays a role in the pathogenesis of inflammatory bowel disease (IBD) in cats. Molecular methods, such as 16S rRNA gene pyrosequencing, allow better assessment of the intestinal microbiota than culture-based techniques. This is the first study to use pyrosequencing to categorize the small intestinal microbiota in cats.

Cats undergoing gastroduodenoscopy for signs of chronic gastrointestinal disease were recruited between July 2006 and April 2009. A Feline Chronic Enteropathy Activity Index (FCEAI) value was calculated for each cat according to a recently described method. Duodenal mucosal biopsies for histology and duodenal mucosal brush samples for pyrosequencing were collected during endoscopy. Duodenal biopsies were graded by a single pathologist (KS) according to published guidelines. Genomic DNA was extracted and bacterial tag-encoded FLX amplicon pyrosequencing (bTEFAP) based upon the V4-V5 region of the 16S rRNA gene was performed. Bacterial identity was determined by comparing sequences against high confidence 16S rRNA gene sequences from public databases. Results were compiled to provide relative abundance estimations at each taxonomic level (expressed as median percentage of total sequences and range). Cats were classified as having food responsive enteropathy (FRE) or steroid responsive enteropathy (IBD) based on their response to dietary or steroid therapy for clinical signs. Mann-Whitney U test was used to compare variables between FRE and IBD groups ( $P < 0.05$ ). Uni-frac distance metric was used to compare the duodenal microbiome of the two groups.

Ten cats with FRE and 16 cats with IBD were recruited. There was no significant difference in age, weight, FCEAI (FRE: median 6.5, range 4-10; IBD: median 8, range 4-13) or histology score (FRE: median 6.5, range 4-10; IBD: median 8, range 4-13) between groups. The most prevalent bacterial phyla were Proteobacteria (FRE: 53.07%, range 19.3-98.4%; IBD: 42.75%, range 3.1-99.1%), Bacteroidetes (FRE: 22.34%, range 0.0-48.3%; IBD: 3.58%, range 0.0-51.9%), Firmicutes (FRE: 8.2%, range 0.7-36.6%; IBD: 11.6%, range 0.01-96.4%) and Actinobacteria (FRE: 1.73%, range 0.5-11.7%; IBD: 2.11%, range 0.0-58.0%). There was no significant difference between the microbiome of FRE and IBD groups.

Pyrosequencing can successfully be used to classify the intestinal microbiota of cats. Further studies are needed to assess whether the microbiota in cats with FRE and IBD differs from that of healthy cats.

#### GA-O-6

**SERUM CALGRANULIN CONCENTRATIONS IN DOGS WITH INFLAMMATORY BOWEL DISEASE.** R.M. Heilmann<sup>1</sup>, K. Allenspach<sup>2</sup>, F. Procoli<sup>2</sup>, K. Weber<sup>1</sup>, J. Suchodolski<sup>1</sup>, J.M. Steiner<sup>1</sup>. <sup>1</sup>Gastrointestinal Laboratory, COLLEGE STATION, United States of America, <sup>2</sup>Royal Veterinary College, University of London, LONDON, United Kingdom

Idiopathic inflammatory bowel disease (IBD) in dogs often represents a diagnostic challenge. Little is known about the pathogenesis of canine IBD, but mounting evidence suggests a central role of innate immunity. Calprotectin, the S100A8/A9 complex, and S100A12 are believed to function as endogenous damage-associated molecular patterns and to be involved in the immune response in diseases such as IBD in humans. In dogs with IBD, expression of mucosal S100-mRNA was shown to be increased 11-fold, but actual concentrations of S100 proteins such as calprotectin and S100A12 have not been investigated to date in canine IBD. Therefore, this study aimed at measuring serum calprotectin and S100A12 concentrations in dogs with IBD before treatment and evaluating their correlation with the clinical disease activity as determined using a clinical scoring system (CCECAI) and the concentration of C-reactive protein (CRP) in serum.

Serum was collected from 13 dogs with IBD at the time of diagnosis and was used to measure serum calprotectin, S100A12, and CRP. Each dog was assessed using the CCECAI scoring system. Canine calprotectin and S100A12 concentrations were measured using analytically specific in-house immunoassays. A Wilcoxon rank sum test was used to compare serum calprotectin and S100A12 concentrations between dogs with IBD and 110 healthy controls. A Spearman rank sum correlation coefficient ( $\rho$ ) was calculated to evaluate the relationship of both serum calprotectin and S100A12 concentrations with CCECAI and serum CRP, respectively.

Canine calprotectin (median: 24.5 mg/L) and S100A12 concentrations (median: 223.0  $\mu$ g/L) were significantly increased in dogs with IBD compared to healthy controls (median: 4.9 mg/L and 85.2  $\mu$ g/L, respectively; both  $p < 0.0001$ ) but did not correlate with the CCECAI score ( $\rho = 0.318$ ;  $p = 0.2905$  and  $\rho = 0.214$ ;  $p = 0.4826$ , respectively) or with the concentration of CRP in serum ( $\rho = 0.396$ ;  $p = 0.1809$  and  $\rho = 0.308$ ;  $p = 0.3064$ , respectively).

This study showed that canine calprotectin and S100A12 are increased in dogs with IBD and that their concentrations in serum may provide a useful addition to the limited repertoire of inflammatory biomarkers available for use in dogs with IBD. Lack of a correlation between serum calgranulin concentrations and clinical disease activity agrees with the results for other serum markers, and the lack of a correlation with the concentration of CRP in serum could possibly be explained by spatial and/or temporal differences in their expression and/or release into the extracellular space. Further studies are under way to evaluate mucosal and fecal concentrations of calprotectin and S100A12 in canine patients with IBD.

#### GA-O-7

**FECAL CALPROTECTIN CONCENTRATION IN ADULT DOGS WITH AND WITHOUT DIGESTIVE TROUBLES.** A. Grellet<sup>1</sup>, R.M. Heilmann<sup>2</sup>, A. Feugier<sup>3</sup>, P. Lecoindre<sup>4</sup>, J. Hernandez<sup>2</sup>, V. Freiche<sup>6</sup>, D. Peeters<sup>7</sup>, J.S. Suchodolski<sup>2</sup>, D. Grandjean<sup>1</sup>, J.M. Steiner<sup>2</sup>. <sup>1</sup>Ecole Nationale Vétérinaire d'Alfort, MAISONS-ALFORT, France, <sup>2</sup>Gastrointestinal Laboratory, College of Veterinary Medicine and Biomedical Sciences, COLLEGE STATION, TX, United States of America, <sup>3</sup>Royal Canin, AIMARGUES, France, <sup>4</sup>Clinique vétérinaire des Cérissioz, ST PRIEST, France, <sup>5</sup>Frégis, ARCUEUIL, France, <sup>6</sup>Alliance, BORDEAUX, France, <sup>7</sup>Faculté de Médecine Vétérinaire, LIÈGE, Belgium

Calprotectin (CP) is a heterodimeric protein complex of neutrophils and macrophages. To screen patients prior to more invasive procedures, several noninvasive markers of gastrointestinal inflammation have been suggested in humans with inflammatory diseases of the gastrointestinal tract. The concentration

of fecal CP in feces has been shown to be a useful marker for inflammatory bowel disease in humans with higher concentrations in affected humans than in healthy controls. A radioimmunoassay for the quantification of canine calprotectin (cCP) in fecal samples has recently been developed and analytically validated [1]. To our knowledge, fecal cCP concentrations have not been studied extensively in dogs with gastrointestinal diseases. Thus this study aimed at investigating fecal cCP concentrations in dogs with and without digestive troubles.

Fecal samples were collected prospectively from 154 dogs. For each dog, fecal consistency was scored using a 5-point numerical scale. Dogs were separated into three groups: healthy dogs with an optimal fecal score (n = 93), dogs with acute diarrhea (n = 33) and dogs with chronic diarrhea (n = 28). For the 28 dogs with chronic diarrhea, a clinical disease activity index (CCECAI) was calculated [2]. In dogs with chronic diarrhea, intestinal inflammation was evaluated by the realisation of a gastroscopy, duodenoscopy and/or colonoscopy and histopathological evaluation of mucosal biopsies from the stomach, duodenum and/or colon. Fecal cCP concentration was evaluated in all the 154 dogs. Data were not normally distributed therefore nonparametric tests (Kruskal Wallis test, Mann-Whitney U test) were used. Significance was determined by a P-value < 0.05.

Fecal cCP concentrations did not differ between healthy dogs and dogs with acute diarrhea. Dogs with chronic diarrhea had significantly higher fecal cCP concentrations compared to dogs with acute diarrhea (medians: 35.6 vs 4.6 µg/g; P < 0.001). Dogs with a CCECAI > 12 had significantly higher fecal cCP concentrations compared to dogs with a CCECAI < 12 (medians: 71.1 vs 19.8 µg/g; P = 0.032). Dogs with chronic diarrhea and severe intestinal inflammation (based on endoscopy and histology) had significantly higher fecal cCP concentrations compared to dogs with chronic diarrhea and mild intestinal inflammation (medians: 71.1 vs 19.8 µg/g; P = 0.024).

In our study, dogs with intestinal inflammation had increased fecal concentrations of cCP. An association between the disease activity as assessed by endoscopy and histology and fecal cCP concentrations was observed.

1. Heilmann et al. AJVR 2008; 69: 845

2. Allenspach K et al. JVIM 2007; 21: 700

#### GA-O-8

**SEROREACTIVITY AGAINST BACTERIAL FLAGELLIN IN DOGS WITH INFLAMMATORY BOWEL DISEASE: PRELIMINARY FINDINGS.** F. Procoli<sup>1</sup>, J. Elson-Riggins<sup>1</sup>, K. Graham<sup>1</sup>, M.deAmbrogio<sup>1</sup>, S. Schmitz<sup>1</sup>, A. Kathrani<sup>1</sup>, F. Gaschen<sup>3</sup>, K. Simpson<sup>4</sup>, A. Rycroft<sup>2</sup>, K. Allenspach<sup>1</sup>. <sup>1</sup>Department of Veterinary Clinical Sciences, Royal Veterinary College, University of London, United Kingdom, <sup>2</sup> Department of Pathology and Infectious Diseases, Royal Veterinary College, University of London, United Kingdom, <sup>3</sup>Department of Veterinary Clinical Sciences, School of Veterinary Medicine, Louisiana State University, United States of America, <sup>4</sup>Department of Clinical Sciences, College of Veterinary Medicine, Cornell University, United States of America

Inflammatory bowel disease (IBD) is a common cause of chronic diarrhoea in dogs. Diagnosis of canine IBD is often challenging and relies on an exclusion diagnosis with histopathological assessment of intestinal mucosal biopsies. Therefore, non-invasive serological markers may help to simplify diagnosis and possibly predict prognosis in canine IBD. Aberrant immune response towards commensal microorganisms has been shown to play a role in the pathogenesis of this disease; and serum antibodies against bacterial flagellin are found in a subset of people with IBD.

Therefore, the aim of this study was to evaluate the presence of antibodies against different bacterial flagellins in a small cohort of dogs with IBD and healthy dogs. All IBD dogs were selected on the basis of moderate clinical severity (Canine Chronic Enteropathy Clinical Activity Index, score >8).

Sera from six dogs diagnosed with IBD at the Royal Veterinary College, London, and 3 healthy dogs were analysed using Western Blot (WB) to assess seroreactivity against different

flagellins. Two commercially available recombinant flagellins, (*Bacillus subtilis* and *Salmonella typhimurium*), and a purified flagellin extracted from a commensal strain of *Escherichia coli*, which had previously been cultured from a dog with confirmed IBD, were used as antigen in the WB. Sera samples were analyzed at dilutions of 1:200 and 1:500. Commercially available murine monoclonal antibodies against *S. typhimurium* flagellin were used as the positive control. All experiments were performed in duplicates.

All IBD dogs showed seroreactivity against commensal derived *E. coli* flagellin, as indicated by the presence of a band of the expected size (55 kD). One IBD dog showed seroreactivity against *S.typhimurium* flagellin. No seroreactivity was found against *Bacillus subtilis* flagellin in any of the IBD dogs. No antibodies against any of the flagellins were detected in the healthy dogs.

To our knowledge this is the first study demonstrating the presence of seroreactivity against flagellin in dogs with IBD. This important finding could lead to the development of a novel non-invasive marker for the diagnosis and monitoring of canine IBD cases.

#### GA-O-8

**TREATMENT OF OESOPHAGEAL SPIROCERCOSIS IN 20 DOGS WITH ORAL DORAMECTIN.** R. Lobetti. Bryanston Veterinary Hospital, BRYANSTON, South Africa

Spirocercosis represents a significant health problem in dogs in many regions of the world, which has been to be difficult to treat as there is currently no registered drug for use in the dog. Although the cattle anthelmintic doramectin, a macrocyclic lactone, has been extra-labelled and successfully used for the treatment of *S. lupi*, there does not appear to be consensus on dose, route, or frequency of administration.

The purpose of this study was to evaluate the effect of a daily dose of doramectin given orally in dogs with spirocercosis. Twenty naturally infected dogs with endoscopic confirmed spirocercosis were evaluated. All dogs were treated with 0.5 mg/kg doramectin administered orally once daily ranging from 42 to 126 days.

In 13 of the dogs (65%) there was resolution of the nodules after 42 days of therapy; whereas in the other seven dogs (35%) nodules were still evident on oesophagoscopy after 42 days. The doramectin was continued at the same dose for a further 42 days (total of 84 days), which resulted in elimination of the nodules in 5 of the dogs. In the other 2 dogs that still had oesophageal nodules the doramectin was continued at the same dose for a further 42 days (total of 126 days), which finally resulted in complete resolution of the nodules. None of the dogs showed any adverse clinical reaction to the doramectin. There was no sex or age predilection for infection but the German shepherd dog (GSD) was over represented at 45% of the cases with an Odds ratio of 9.35. In addition seven GSD's did not responded to the initial course of therapy, which could imply that the GSD is less sensitive to doramectin and that it requires a longer duration of therapy before there is resolution of the oesophageal nodules.

This study concluded that the daily use of doramectin at 0.5 mg/kg once a day is effective in the elimination of *Spirocera lupi* oesophageal nodules in dogs without any clinical side effects.

#### GA-O-10

**INTERLEUKIN-17A AND INTERLEUKIN-22 MRNA EXPRESSION IS LOW IN DUODENAL TISSUE FROM DOGS WITH INFLAMMATORY BOWEL DISEASE.** S. Schmitz, D. Werling, K. Allenspach. Royal Veterinary College, NORTH MYMMS, United Kingdom

Inflammatory bowel disease (IBD) is the most common cause of chronic gastrointestinal signs in dogs, the pathogenesis of which remains elusive. In people with IBD, Interleukin (IL)-17-

and IL-22-producing CD4(+) helper T cells (Th17-cells) mediate mucosal immunity and are involved in the pathogenesis of the disease. Cytokines specific to the Th17 pathway have so far not been investigated in the context of canine IBD. The aim of this study was therefore to investigate the mRNA expression patterns of key Th17 cytokines (IL-17A and IL-22) in duodenal tissue from dogs with IBD.

Reverse-transcriptase quantitative polymerase chain reaction (RT-qPCR) was performed on archived cDNA samples from duodenal endoscopic biopsies of 18 German Shepherd Dogs (GSD) diagnosed with IBD, 33 dogs of other breeds with IBD and 15 healthy control dogs. Primers were designed based on the predicted canine gene sequence for IL-17A & IL-22. Absolute quantification of gene expression was performed using a standard curve and internal controls. Comparison between groups was performed with Graph-Pad Prism software using one-way ANOVA and Mann-Whitney U tests. Statistical significance was set at  $p < 0.05$ .

There was a significant difference of absolute IL-17A mRNA quantities across groups ( $p = 0.027$ ). IL-17A mRNA concentration was significantly lower in the duodenum of GSD with IBD compared to other breeds of dogs with IBD ( $p = 0.004$ ) and healthy dogs ( $p < 0.0001$ ). There was no difference in absolute IL-22 mRNA expression across groups ( $p = 0.25$ ).

In contrast to the situation in human IBD, canine IL-17A and IL-22 mRNA were not increased in diseased tissue compared to controls. Overall, expression of both genes was low in this pilot study. The exact contribution of Th17 cytokines to the pathogenesis of canine IBD should be further investigated in the future in order to characterize the disease in dogs on a molecular level.

#### GA-O-11

**UTILITY OF THE SERUM PANCREATIC ELASTASE-1 IN THE DIAGNOSIS OF CANINE PANCREATITIS.** B R. Jones<sup>1</sup>, P.D. Watson<sup>2</sup>, C.S. Mansfield<sup>3</sup>. <sup>1</sup>IVABS, PALMERSTON NORTH, New Zealand, <sup>2</sup>University of Cambridge, CAMBRIDGE, United Kingdom, <sup>3</sup>University of Melbourne, MELBOURNE, Australia

Pancreatitis in dogs is a diagnostic challenge for veterinary surgeons due to the variable forms of the disease, a factor which influences the clinicopathological findings. Pancreatic elastase-1 is a pancreas-specific enzyme synthesised in the pancreatic acinar cells that is released into the blood stream as a result of pancreatic inflammation at the same time or immediately after trypsin. It also directly contributes to ongoing inflammation in the disease, and its concentration can be measured in canine serum.

The purpose of this prospective study was to determine the clinical utility of serum canine pancreatic elastase (cPE1) for the diagnosis and differentiation of pancreatic disease in dogs.

61 dogs were included from three veterinary referral centres. 49 had pancreatic disease confirmed on ultrasonography and/or histology (severe acute pancreatitis  $n=23$ , mild acute pancreatitis  $n=11$ , acute on chronic pancreatitis  $n=7$ , chronic pancreatitis  $n=3$  and pancreatic carcinoma  $n=5$ ); whilst 12 had similar clinical signs to pancreatitis, but had normal pancreatic histology. There was a significant difference ( $p=0.005$ ) in serum cPE1 between dogs with all types of pancreatic disease and non-pancreatic disease, and a significant difference in serum cPE1 between acute pancreatitis and non-pancreatic disease ( $p=0.0022$ ). A cut-off value for serum cPE1  $> 17.24$  ng/mL resulted in sensitivity of 61.4% and specificity of 91.7% for diagnosis of all types of pancreatic disease. The sensitivity rose to 65.85% and 78.26% for the diagnosis of all types of acute pancreatitis and severe acute pancreatitis, respectively.

Serum cPE-1 is more sensitive at confirming a diagnosis of severe acute pancreatitis than chronic or mild acute pancreatitis. Dogs with chronic pancreatitis tended to have a low serum cPE1 concentration, suggesting decreased exocrine tissue and function. A diagnosis of severe acute pancreatitis can be made by measurement of serum cPE1 and concurrent imaging would improve the diagnostic sensitivity. Concurrent histopathology is required for confirmation in chronic disease.

#### GA-O-12

**SERUM TRIGLYCERIDE AND CHOLESTEROL CONCENTRATIONS AND LIPOPROTEIN PROFILES IN DOGS WITH NATURALLY OCCURRING PANCREATITIS AND HEALTHY CONTROL DOGS.** P.G. Xenoulis<sup>1</sup>, P.J. Cammarata<sup>2</sup>, K. Wooten<sup>1</sup>, R.M. Heilmann<sup>1</sup>, K. Weber<sup>1</sup>, R. Walzem<sup>3</sup>, R. D. Macfarlane<sup>2</sup>, J.S. Suchodolski<sup>1</sup>, J.M. Steiner<sup>1</sup>. <sup>1</sup>Gastrointestinal Laboratory, Texas A&M University, College Station, TX, USA, <sup>2</sup>Laboratory for Cardiovascular Chemistry, Texas A&M University, College Station, TX, USA, <sup>3</sup>Department of Nutrition, Texas A&M University, College Station, TX, USA

A possible association between hyperlipidemia and pancreatitis is obscure in dogs. Under certain circumstances hypertriglyceridemia might cause pancreatitis, but hyperlipidemia has also been hypothesized to be the result of pancreatitis. Studies specifically investigating the serum lipid status and lipoprotein profiles of dogs with spontaneous pancreatitis have not been reported. The aim of this study was to compare serum triglyceride and cholesterol concentrations and lipoprotein profiles between dogs with naturally occurring pancreatitis and healthy dogs.

Seventeen dogs with pancreatitis (group 1) were enrolled into this study. The diagnosis of pancreatitis was based on the presence of compatible clinical signs and a serum Spec cPL<sup>®</sup> concentration  $\geq 400$   $\mu\text{g/L}$ . Dogs with hyperlipidemia secondary to diseases or medications known to cause hyperlipidemia were excluded from the study. Fifty-three healthy dogs with a normal serum Spec cPL concentration ( $<200$   $\mu\text{g/L}$ ) and no history of diseases or current drug use that affect lipid metabolism were enrolled as controls (group 2). Food was withheld for at least 12 hours prior to blood collection. Lipoprotein profiling was performed using a NaBiEDTA density gradient ultracentrifugation method. Data were analyzed using the Shapiro-Wilk test, t-tests, Mann-Whitney tests, Fisher's exact tests, and sliced inverse regression.

Three of the 17 dogs (18%) in group 1 and 4/53 control dogs (7.5%) in group 2 had hypertriglyceridemia ( $p=0.35$ ). Dogs in group 1 had significantly higher serum triglyceride concentrations (median: 67 mg/dL; range: 48-324 mg/dL) than dogs in group 2 (median: 54 mg/dL; range: 26-257 mg/dL;  $p=0.0026$ ). Four of 17 dogs in group 1 (24%) and 1/53 control dogs in group 2 (1.9%) had hypercholesterolemia ( $p=0.011$ ). However, there was no significant difference in serum cholesterol concentrations between the two groups ( $p=0.565$ ). Lipoprotein profiles were distinctly different between dogs with pancreatitis and healthy dogs ( $p=0.0012$ ). Dogs could be classified in the correct group (healthy versus pancreatitis) with 89% accuracy based on their lipoprotein profiles alone. The most important differences involved LDL2, LDL3, and LDL4.

In the present study, the majority of dogs with pancreatitis ( $>70\%$ ) had normal serum triglyceride and cholesterol concentrations, while some dogs had mild increases in those parameters. The majority of dogs with pancreatitis had distinctly different lipoprotein profiles from healthy dogs, even in cases in which serum concentrations of triglyceride and cholesterol were normal. Further studies are needed to determine the pathophysiology and clinical importance of those changes in lipoprotein profiles.

#### GA-O-13

**TYLOSIN ADMINISTRATION INCREASES THE LEVELS OF ENTEROCOCCUS SPP. IN THE FECES OF DOGS WITH TYLOSIN-RESPONSIVE DIARRHEA.** S. Kilpinen, M. Rantala, T. Spillmann, J. Björkroth, E. Westermarck. University of Helsinki, UNIVERSITY OF HELSINKI, Finland

Tylosin has been shown to be effective in treating diarrheal disorders in dogs and thus the term tylosin-responsive diarrhea (TRD) has been established. In dogs with TRD, the stool remains normal as long as treatment continues, but diarrhea reappears in many dogs within weeks after discontinuation. Tylosin has been reported to increase the proportions of Enterococcus-like organisms in the canine jejunal microbiota of healthy dogs. It was then speculated, that this could be one possible

factor contributing to tylosin's positive effect in ceasing diarrhea in dogs, as some enterococci strains are known to have probiotic characters. The aim of this clinical study was to assess the effects of tylosin administration on the levels of *Enterococcus* spp. and lactic acid bacteria (LAB) as well as levels of tylosin resistant *Enterococcus* spp. and LAB in the feces of dogs with TRD. Eleven client-owned dogs with TRD were enrolled in this prospective clinical trial. At the beginning (time point A) all the dogs were on tylosin treatment and had normal fecal consistency. After the tylosin treatment was discontinued a follow-up period of a maximum of two months commenced to determine the reappearance of diarrhea. After onset of diarrhea (time point B), all dogs received tylosin orally 25 mg/kg q 24h for seven days. On day seven (time point C) the stool was firm again in all dogs. Fecal samples were collected on all three time points. Feces were serially diluted ten-fold in peptone water and plated for enumeration. LAB and enterococci were enumerated on MRS and Slanez-Bartley media, respectively. Tylosin-resistant LAB and enterococci were enumerated on corresponding media supplemented with 100 µg/ml tylosin. The results showed that the total counts of *Enterococcus* spp. and LAB decreased after tylosin discontinuation while the dogs had diarrhea and increased during tylosin administration when the dogs had normal fecal consistency. The changes in the total counts of *Enterococcus* spp. were significant ( $p < 0.05$ ). Almost all LAB and *Enterococcus* spp. were resistant to tylosin when the dogs were on tylosin treatment (timepoint A and C), but resistance against tylosin decreased significantly when the treatment was discontinued. Our findings indicate that tylosin administration results in a significant increase in the levels of *Enterococcus* spp. in the feces of dogs suffering from TRD. These bacteria could have probiotic properties and thus attenuate inflammation on the gut mucosa and normalize the fecal consistency in dogs with diarrhea.

#### EN-O-1

**INVOLVEMENT OF THE GH-IGF PATHWAY IN CANINE CORTISOL-SECRETING ADRENOCORTICAL TUMORS.** N. van derHelm, S. Galac, M.M.J. Kool, J.A. Mol, H.S. Kooistra. Utrecht University, Faculty of Veterinary Medicine, UTRECHT, The Netherlands

Hypercortisolism is a common endocrinological disorder of middle-aged and older dogs. In about 15% of cases the excessive secretion of glucocorticoids is a result of an ACTH-independent benign or malignant adrenocortical tumor (AT). The growth hormone (GH)-insulin-like growth factor (IGF) pathway has been reported to be important in adrenocortical growth and steroidogenesis. Moreover, in about 90% of human cortisol-secreting adrenocortical carcinomas overexpression of IGF-II is described. In case of overexpression of IGF, IGF-receptor blockers may be used to treat this disorder. Therefore, the objective of this study was to investigate whether altered expression of the GH-IGF pathway plays a role in tumorigenesis of cortisol-secreting ATs in dogs.

Gene expression of GH and its receptor (GHR), IGF-I, IGF-II, the IGF-receptors (IGF-IR and IGF-IIR) and six IGF-binding proteins (IGFBP-1 to -6) was performed by quantitative PCR on tissue of 15 normal adrenals, 13 adrenocortical adenomas and 30 carcinomas. The ATs were obtained from dogs with ACTH-independent hypercortisolism that underwent unilateral adrenalectomy at the Department of Clinical Sciences of Companion Animals.

In both normal adrenals and ATs mRNA encoding for all genes except IGFBP-1 was present. Compared to normal adrenals, there was significantly lower abundance of mRNA encoding for IGF-IR in carcinomas ( $p < 0.05$ ) and for IGFBP-5 in both adenomas and carcinomas ( $p < 0.01$ ). The abundance of mRNA encoding for IGFBP-2 in both tumor types was significantly higher ( $p < 0.01$ ) than in normal adrenals. Immunohistochemistry is needed to confirm the results at the protein level.

In conclusion, these results demonstrate that in canine cortisol-secreting ATs there are changes in the gene expression pattern of the GH-IGF pathway and these changes may play a role in the pathogenesis of these tumors.

#### EN-O-2

**LONG-TERM FOLLOW-UP OF RENAL FUNCTION IN DOGS WITH CUSHING'S DISEASE BEFORE AND AFTER TREATMENT.** P.M.Y. Smets<sup>1</sup>, H.P. Lefebvre<sup>2</sup>, B.P. Meij<sup>3</sup>, E. Meyer<sup>1</sup>, Ivan deMaele<sup>1</sup>, S. Daminet<sup>1</sup>. <sup>1</sup>Ghent University, MERELBEKE, Belgium, <sup>2</sup>Ecole Nationale Vétérinaire, TOULOUSE, France, <sup>3</sup>Utrecht University, UTRECHT, The Netherlands

Proteinuria and systemic hypertension are important factors in development and progression of chronic kidney disease in dogs. Forty-six % of dogs with pituitary-dependent hypercortisolism (PDH) have proteinuria and 50 to 80 % have hypertension, which do not always resolve after treatment. Therefore, dogs with PDH could be considered at risk for renal complications. Additionally, exogenous glucocorticoids have been shown to increase glomerular filtration rate (GFR) in humans and in dogs. Still, information about effects of hypercortisolism on renal function is scarce.

The aim of this prospective study was to compare renal function before and after treatment in dogs with PDH.

Twenty dogs with PDH (12 treated with trilostane and eight by transphenoidal hypophysectomy) were included. Serum creatinine (sCr), urinary specific gravity (USG), urinary protein-to-creatinine ratio (UPC) and a urinary tubular marker, i.e. retinol-binding protein-to-creatinine ratio (uRBP/c), were evaluated at baseline (T0) and at one (T1), three (T3), six (T6) and 12 months (T12) after treatment. At T0, T6 and T12, GFR was measured by plasma exogenous creatinine clearance (Cl<sub>creat</sub>).

Based on a general linear model, sCr was significantly lower at T0 compared to T3 ( $p = 0.011$ ) and T6 ( $p = 0.003$ ), but not to T12. USG was significantly higher at T3 ( $p = 0.003$ ), T6 ( $p < 0.001$ ) and T12 ( $p < 0.001$ ) than at T0. At T1 ( $p < 0.001$ ), T3 ( $p < 0.001$ ), T6 ( $p = 0.001$ ) and T12 ( $p = 0.005$ ), UPC was lower than at T0. Despite good control of PDH, UPC remained  $> 0.5$  during treatment in four medically treated dogs. In the surgically treated dogs, UPC remained  $< 0.5$ , except at T6 in two dogs with recurrence of hypercortisolism. Following medical therapy with trilostane, UPC decreased again to  $< 0.5$  in one of the dogs. Urinary RBP/c decreased after treatment in most dogs, although no statistically significant differences were detected compared to T0. Plasma Cl<sub>creat</sub> was significantly higher before treatment than at T6 and T12 ( $p < 0.001$ ).

In conclusion, GFR was increased in dogs with PDH and normalised after treatment. However, UPC and uRBP/c ratios remained high in some treated dogs, the latter suggesting persistent tubular dysfunction. This study underlines that PDH is associated with renal functional changes which do not always reverse after therapy.

#### EN-O-3

**ROUTINE SCREENING OF DIABETIC CATS FOR ACROMEGALY: OVERDUE OR OVERKILL?** S.J.M. Niessen, Y. Forcada, K. Jensen, B. Glanemann, D.B. Church. Royal Veterinary College, NORTH MYMMS, United Kingdom

Feline acromegaly is suspected to be more common than previously assumed. Additionally, the classic phenotypical appearance seems not consistently present. These two findings could jointly foster late diagnosis or complete failure to diagnose and therefore appropriately manage the disease in a significant proportion of diabetic cats. In order to prevent this, routine screening of diabetic cats for acromegaly could be argued for. The current study aimed to prospectively perform such routine screening in a large cohort of diabetic cats in order to verify if its prevalence amongst diabetic cats would justify the effort.

Free fructosamine determination for diabetic cats was offered to veterinary surgeries across the United Kingdom. Submitting clinicians were asked to indicate whether they suspected acromegaly in their patient. Spare serum was used to measure insulin-like growth factor-1 (IGF-1) via radio-immunoassay and if found to be  $> 1000$  ng/ml, free contrast-enhanced brain computed tomography (CT) was offered.

Twelve hundred and twenty-two diabetic cat samples were submitted. IGF-1 measurement suggested possible acromegaly in three hundred and twenty three cats (26.4%). Of those with an

IGF-1<1000ng/ml 588 were male neutered (MN, 65%), 290 female neutered (FN, 32%); mean age $\pm$ -standard deviation (SD) was 11.8 $\pm$ -3.2 (range 0.5-20 years); 744 were domestic short hair cats (DSH, 83%), 77 domestic long hair (DLH (9%), 22 Burmese (2%); clinicians strongly suspected acromegaly in 1% of these cases; of cats with IGF-1>1000ng/ml 226 were MN (70%), 82 FN (25%); mean age $\pm$ -SD was 11.3 $\pm$ -2.7 years (range: 4-19); 281 were DSH (87%), 21 DLH (7%) and 5 Burmese (2%); clinicians strongly suspected acromegaly in 24% of these cases. Body weight and fructosamine were significantly higher in cats with IGF-1>1000ng/ml (5.4 $\pm$ -1.2kg; 559 $\pm$ -142 $\mu$ mol/l versus 5.2 $\pm$ -1.4kg; 476 $\pm$ -151 $\mu$ mol/l; Mann Whitney,  $p$ <0.01), despite significantly higher daily insulin dosages (mean 14.6iu/day versus 5.7iu/day, Mann Whitney,  $p$ <0.01). CT confirmed acromegaly in 34 of 38 (89%) patients with IGF-1>1000ng/ml.

The current study confirms that acromegaly is likely present in a high proportion of diabetic cats (one in four). The data also suggest that historical and phenotypical evidence of the disease are only deemed strongly suggestive of the disease in a minority of cases (also one in four). For these reasons and since acromegaly has significant implications on the understanding, management and prognosis of individual patients, it seems logical to consider routinely screening diabetic cats for acromegaly. If acromegaly is diagnosed and treated earlier, higher diabetic remission rates might be achieved.

#### EN-O-4

**TSH STIMULATION TEST WITH RECOMBINANT HUMAN TSH (RH-TSH) FOR THE DIAGNOSIS OF FELINE HYPERTHYROIDISM.** J.L.A. Müller, R. Neiger, Jstus-Liebig-University Giessen, GIESSEN, Germany

Feline hyperthyroidism is the most common endocrine disease in the older cat. An increased total thyroxin (TT4) serum concentration is found in most patients. However, diagnosis may be more challenging if TT4 value is within the reference range.

This condition is most commonly seen in cats with mild hyperthyroidism as a result of non-specific fluctuation in the thyroid hormone production or in cats with concurrent systemic illness, where TT4 can be suppressed into the reference range. Therefore additional diagnostic tests are needed to diagnose hyperthyroidism in cats suspected of having the disease despite normal TT4 serum concentration. The aim of this study was to show that a TSH stimulation test with rh-TSH is a reliable alternative to thyroid scintigraphy.

In a prospective study over a period of 13 months, TSH-stimulation with rh-TSH test was performed in 40 cats with hyperthyroidism and in 20 clinically healthy cats > 8 years of age with normal TT4 values. Diagnosis of hyperthyroidism was confirmed by thyroid scintigraphy using 40 Mbq of petechnetat. TT4 serum concentrations were measured before and 6 hours after the intravenous administration of 25 $\mu$ g rh-TSH(Thyrogen, Genzyme). Reference basal TT4 concentration was defined as a value between 1.0 and 4.0 $\mu$ g/dl.

Median basal and stimulated TT4 concentration in hyperthyroid cats was 15.6 $\mu$ g/dl (range: 3.8-40.7 $\mu$ g/dl) and 15.3 $\mu$ g/dl (4.1-48.2 $\mu$ g/dl), respectively, while in healthy cats it was 2.05 $\mu$ g/dl (1.1-3.5 $\mu$ g/dl) and 4.8 $\mu$ g/dl (2.8-7.8 $\mu$ g/dl), respectively. Of the 40 cats with hyperthyroidism one cat had a basal TT4 concentration within the reference range (3.8 $\mu$ g/dl) and two cats showed only a marginally increased TT4 serum concentrations (4.2 $\mu$ g/dl and 4.5 $\mu$ g/dl).

Calculation of percentage increase after stimulation showed a significant difference (Mann-Whitney-Test:  $p$ <0.001) in rise in healthy cats (median stimulation: 127%; range: 62-220%) compared to cats with hyperthyroidism (13%; 0-120%). The three hyperthyroid cats based on scintigraphy with high normal or marginally increased basal TT4 had a percentage increase of 0%, 8% and 19%, respectively. Calculation of absolute increase after stimulation showed no significant difference (Mann-Whitney-Test:  $p$ =0.144) between healthy and hyperthyroid cats.

The results of this study suggest that a reliable diagnosis of feline hyperthyroidism in cats with normal to mildly increased TT4 levels can be achieved with a TSH stimulation test if thyroid scintigraphy is not available.

#### EN-O-5

**INDICES OF URINARY CAUXIN AND N-ACETYL- $\beta$ -D-GLUCOSAMINIDASE (NAG) ARE NOT PREDICTORS OF SURVIVAL OR THE DEVELOPMENT OF AZOTAEMIA IN HYPERTHYROID CATS.** T.L. Williams, J. Elliott, H. Syme. ROYAL VETERINARY COLLEGE, LONDON, United Kingdom

Recent studies reported urinary protein:creatinine ratio (UPC) is a predictor of survival but not the development of azotaemia in cats with hyperthyroidism. Hyperthyroid cats are proteinuric, however urinary albumin:creatinine ratio (UAC) does not decrease following treatment of hyperthyroidism, and UPC, but not UAC, is correlated with plasma renin activity. This suggests that the proteinuria associated with hyperthyroidism is predominately of tubular rather than glomerular origin. Cauxin and NAG are proteins of tubular origin, and urinary cauxin:creatinine ratio (CCR) and NAG index (NAGi) have been associated with the development of azotaemia in geriatric cats. This study examined whether CCR and NAGi in hyperthyroid cats were associated with survival time or the development of azotaemia following treatment.

Hyperthyroid cats were recruited into the study at two first opinion practices in London between 1999 and 2009. Hyperthyroidism was treated with anti-thyroid medication alone or in combination with thyroidectomy. Cats were monitored for a six month period after initiating treatment for hyperthyroidism and categorised as azotaemic or non-azotaemic after this time. Hyperthyroid cats with azotaemia pre-treatment were not included in the analysis of predictors of the development of azotaemia, but were included in the survival analysis. Relative urinary cauxin concentration and NAG activity were measured at baseline using previously validated assays, and were indexed to urinary creatinine concentration. CCR was also measured after six months of treatment. Results are reported as median [25th, 75th percentile] values. Baseline CCR and NAGi were compared between pre-azotaemic and non-azotaemic cats using the Mann-Whitney-U test. Pre and post-treatment CCR were compared using the Wilcoxon signed rank test. Cats were divided into groups according to baseline CCR and NAGi (above/below the median value) and survival times compared using the log rank test.

CCR did not decrease following treatment (pre-treatment 0.56 [0.26, 1.84] vs. post-treatment 0.38 [0.13, 1.10],  $n$ =21;  $P$ =0.414). Baseline CCR was not significantly different between pre-azotaemic and non-azotaemic cats (0.54 [0.22, 1.72],  $n$ =24 vs. 0.49 [0.18, 1.67],  $n$ =130;  $P$ =0.69) and was not associated with survival time ( $n$ =154;  $P$ =0.979). Baseline NAGi was also not significantly different between pre-azotaemic and non-azotaemic cats (1.91 [0.67, 6.01],  $n$ =24 vs. 1.26 [0.56, 2.30],  $n$ =28;  $P$ =0.147) and was not associated with survival time ( $n$ =54;  $P$ =0.287).

Cauxin does not appear to contribute significantly to the urinary proteome of hyperthyroid cats. CCR and NAGi were not associated with the presence of underlying azotaemic chronic kidney disease in cats with hyperthyroidism, and are not of prognostic value.

#### EN-O-6

**CHANGES IN PARATHYROID HORMONE (PTH) CONCENTRATIONS AFTER TREATMENT OF HYPERTHYROID CATS WITH VARIABLE RENAL FUNCTION.** T.L. Williams, J. Elliott, H. Syme. ROYAL VETERINARY COLLEGE, LONDON, United Kingdom

Hyperparathyroidism is reported to be common in cats with hyperthyroidism and chronic kidney disease (CKD). Fifteen to forty-nine percent of hyperthyroid cats have CKD, therefore the hyperparathyroidism reported with hyperthyroidism might be caused by renal secondary hyperparathyroidism associated with underlying CKD. The aim of this study was to determine if plasma PTH concentrations were associated with the presence of underlying CKD in hyperthyroid cats, and to investigate the changes in plasma PTH concentrations that occur following treatment of hyperthyroidism.

Hyperthyroid cats were recruited from two London-based first opinion practices between 1999 and 2009. Cats that were

azotaemic at diagnosis were excluded. Hyperthyroidism was treated with anti-thyroid medication alone or in combination with thyroidectomy. Cats were included in the study if they had a plasma total thyroxine concentration <40 nmol/l documented for a six month period following commencement of treatment. Cats were classified as having azotaemic CKD if they developed renal azotaemia within six months of establishment of euthyroidism. Otherwise cats were deemed to have normal renal function. Intact PTH concentrations were measured in stored EDTA plasma samples using a commercially available immunoradiometric assay that was validated as part of the study. The Mann-Whitney U test and the Wilcoxon signed rank test were used to compare between the groups and assess the response to treatment respectively. Results are reported as median [25th, 75th percentile] concentrations.

Intra- and inter-assay variability was <10% and dilutional parallelism was observed. Forty three cats with hyperthyroidism (22 azotaemic and 21 non-azotaemic) were included in the study. Pre-treatment plasma PTH concentrations were significantly higher in cats that developed azotaemia than cats that remained non-azotaemic (21.9 [17.7, 70.0] pg/ml vs. 14.5 [9.8, 22.0] pg/ml;  $P=0.013$ ). Plasma PTH concentrations decreased following treatment in non-azotaemic cats (13.8 [9.5, 20.0] pg/ml vs. 9.7 [7.7, 14.8] pg/ml,  $n=13$ ;  $P=0.023$ ), however plasma PTH concentrations remained unchanged following treatment in azotaemic cats (19.5 [16.3, 58.3] pg/ml vs. 26.6 [11.5, 55.4] pg/ml,  $n=10$ ;  $P=0.646$ ). Post treatment plasma PTH concentrations were also higher in azotaemic than non-azotaemic cats ( $P=0.005$ ).

Plasma PTH concentrations were higher in pre-azotaemic cats than non-azotaemic cats, probably reflecting the presence of secondary renal hyperparathyroidism in hyperthyroid cats with underlying CKD. Plasma PTH concentrations were also higher in hyperthyroid cats without underlying CKD before treatment, suggesting that an additional pathophysiological mechanism is responsible for increased PTH concentrations in some cats with hyperthyroidism.

#### EN-O-7

**A MISSENSE MUTATION IN THE CODING SEQUENCE OF MC4R (MC4R:C.92 C>T) IS PRIMARILY ASSOCIATED WITH SUSCEPTIBILITY TO DIABETES MELLITUS IN OBES DSH CATS.** Y. Forcada Atienza, A. Holder, R. Jepsen, D.B. Church, B. Catchpole. Royal Veterinary College, NORTH MYMMS, United Kingdom

Diabetes mellitus (DM) is one of the most common feline endocrinopathies and is considered to have a similar pathophysiological basis to human type 2 diabetes. Genome-wide association studies in humans have identified several genes that predispose to obesity and/or DM, one of which is melanocortin receptor 4 (MC4R). A previous study identified a missense mutation in the coding sequence of feline MC4R in domestic shorthair (DSH) cats. This mutation was more prevalent in a population of obese diabetic cats compared to a population of lean non-diabetic controls. However, the previous results did not establish whether this mutation had a primary effect on susceptibility to obesity rather than DM. The aim of the current study was to further investigate the role of the mutation on obesity and/or DM in cats.

A total of 120 cats were included in the follow-up study. The first group consisted of 60 lean diabetic cats (30 male, 30 female), mean age 12.7 years (range 5-18y), mean weight 4.15 Kg (range: 2.59-6Kg). The second group consisted of 60 obese non-diabetic cats (30 male, 30 female), mean age 15.2 years (range: 9.1-20y), mean weight 5.7 Kg (range: 3.4-8.3Kg). Primers had been designed that flanked the mutation to allow PCR amplification of this region of MC4R from genomic DNA obtained from EDTA blood. The PCR products were subjected to restriction fragment length polymorphism (RFLP) analysis. BstOI digestion products were analysed by agarose gel electrophoresis. Statistical analysis was performed using Chi squared test. Bonferroni correction was applied when doing multiple group comparisons. Of the 60 lean diabetic cats, 20 (33%) were homozygous for the mutation (CC), compared to 15 (25%) of 60 obese control cats. This compares with 21 of 60 homozygous mutants in the lean non-diabetic population and 33 of 60 homozygous mutants in

the obese diabetic population seen previously. The difference between lean diabetic cats and lean non-diabetic cats was not statistically significant, whereas the difference between the group of obese diabetics and obese non-diabetic was statistically significant ( $p<0.05$ ). This study further confirms that a missense mutation identified in the coding sequence of MC4R in DSH cats is associated with DM in obese DSH cats. Conversely to the information available in human studies, this mutation does not seem to be primarily associated with obesity and secondary type 2 diabetes. In contrast, these results suggest that the mutation might be involved in progression to overt DM in obese cats.

#### EN-O-8

**PANCREATIC ENZYMES ACTIVITY AND ULTRASONOGRAPHIC FINDINGS IN DIABETIC CATS AT DIAGNOSIS AND DURING FOLLOW-UP.** E. Zini<sup>1</sup>, M. Hafner<sup>1</sup>, M. Osto<sup>2</sup>, S. Ohlert<sup>3</sup>, M. Franchini<sup>4</sup>, M. Ackermann<sup>4</sup>, T.A. Lutz<sup>2</sup>, C.E. Reusch<sup>1</sup>. <sup>1</sup>Clinic for Small Animal Internal Medicine, Zurich, Switzerland, <sup>2</sup>Institute of Veterinary Physiology, <sup>3</sup>Division of Diagnostic Imaging, <sup>4</sup>Institute of Virology; Vetsuisse Faculty, University of Zurich, Switzerland

Pancreatitis is observed in up to 50% of cats with diabetes mellitus at necropsy. Whether pancreatitis develops before diabetes, and may be causally involved, or during the disease is unknown. Currently, diagnosis of pancreatitis in vivo in cats relies on a combination of clinical signs, measurement of pancreatic enzymes activity, such as feline pancreatic lipase and trypsin-like immunoreactivity (fPLI and fTLI, respectively), and abdominal ultrasonography. The aim of the study was to use these parameters to investigate the occurrence of pancreatitis in diabetic cats on admission and during follow-up.

Cats with newly diagnosed diabetes not affected by obvious concurrent diseases were enrolled. Activity of fPLI and fTLI was measured, and pancreatic ultrasonography was performed according to a standardized protocol on admission, and after 3-6 months; ultrasonography included assessment of pancreatic contours (regular, irregular), echogenicity (normal, decreased), cross-sectional size of the left and right limb ducts (normal, increased), and nearby presence of free-fluid or hyperechoic peritoneum. Pancreatitis was suspected if fPLI or fTLI activity was increased, or if 2 ultrasonographic abnormalities were observed. Data were analyzed with descriptive statistics.

Twenty-seven diabetic cats were included. At diagnosis fPLI was increased in 8 (29.6%) cats; 2 of them also had increased fTLI or pancreatic lesions seen by ultrasonography. After 3-6 months, fPLI normalized in one of the 8 cats and remained increased in the other 7. TLI and pancreatic ultrasonography normalized in the 2 cats with initial changes. One cat had an abnormal pancreas on ultrasonography at diagnosis which could no longer be demonstrated during follow-up. In 6 cats which showed no abnormalities at diagnosis, increases of fPLI (3), fTLI (2) or pancreatic lesions on ultrasonography (1) were demonstrated after 3-6 months. In 12 cats fPLI, fTLI and pancreatic ultrasound remained normal at diagnosis and during follow-up. Interestingly, none of the 27 cats showed clinical signs of pancreatitis at any time during the study.

The results suggest that pancreatitis is present in approximately 30% of diabetic cats at diagnosis. The increase of fPLI and fTLI, or abnormal ultrasonography during follow-up, i.e. in cats that were initially normal, may suggest that pancreatitis develops as a consequence of diabetes in some cases. Absence of clinical signs in all cats indicates that pancreatitis was probably mild.

#### EN-O-9

**PROGNOSTIC FACTORS IN CATS WITH NEWLY DIAGNOSED DIABETES MELLITUS.** C. Callegari<sup>1</sup>, M. Hafner<sup>2</sup>, M. Osto<sup>3</sup>, M. Franchini<sup>4</sup>, M. Ackermann<sup>4</sup>, T.A. Lutz<sup>3</sup>, C.E. Reusch<sup>2</sup>, E. Zini<sup>1,2</sup>. <sup>1</sup>Istituto Veterinario di Novara, Italy, <sup>2</sup>Clinic for Small Animal Internal Medicine, <sup>3</sup>Institute of Veterinary Physiol-

ogy, <sup>4</sup>Institute of Virology, Vetsuisse Faculty, University of Zurich, Switzerland.

Diabetes mellitus is one of the most frequent endocrinopathies in cats but prognostic factors are largely unknown. The only published study was performed with 55 cats and included cases previously treated with insulin or followed-up by phone call. Old age on admission was negatively correlated with survival, whereas body weight, gender, initial blood glucose concentration and ketonuria were not. Here, the aim was to identify prognostic factors using a large population of diabetic cats, and to include only cases with newly diagnosed diabetes that were followed-up at our institution.

Of 275 diabetic cats examined between 2000 and 2009, 114 fulfilled the inclusion criteria. Clinical records were used to collect data on history, signalement, physical examination, haematological and biochemical profile at the time of diagnosis. The type of insulin, the occurrence and duration of clinical remission, the occurrence of ketoacidosis or concurrent diseases at the time of diagnosis or during follow-up were also retrieved. Survival and outcome predictors were studied with Kaplan-Meier and Cox proportional hazard models.

Median survival of diabetic cats was 516 days (range: 1-3468); 75% lived longer than 54 days and 25% longer than 1420 days. Survival was shorter for cats with concurrent disease (OR: 1.80, CI95%: 1.01-3.23,  $p=0.04$ ) or hyperkalemia (OR: 2.16, CI95%: 1.04-4.48,  $p=0.04$ ) at diagnosis. After exclusion of 19 cats that died before discharge after diagnosis, hyperkalemia remained a predictor of survival (OR: 2.70, CI95%: 1.10-6.76,  $p=0.03$ ). Overall, hyperkalemia was observed in 17 cases (14.9%) and was mild (median  $K^+$ : 5.9 mEq/L, range: 5.5-6.5). Survival was shorter in cats that developed ketoacidosis (OR: 5.99, CI95%: 3.19-11.24,  $p<0.01$ ) or concurrent disease (OR: 2.82, CI95%: 1.48-5.37,  $p<0.01$ ) during follow-up. Age, gender, breed, body weight, hematocrit, leukocyte count, serum glucose, fructosamine, total protein, albumin, creatinine, urea, bilirubin, cholesterol, lipase, ketoacidosis at diagnosis, duration of remission and type of insulin were not associated with survival.

We conclude that hyperkalemia, concurrent diseases, and ketoacidosis at diagnosis or during follow-up predict a poorer outcome in diabetic cats. The causal link between decreased life expectancy and hyperkalemia remains unknown, in particular considering its mild degree. Concurrent diseases may complicate diabetes management or decrease owners' willingness to treat. Of note, ketoacidosis decreased survival during follow-up but not at diagnosis, suggesting that in the latter this complication should not be regarded as unfavourable.

#### EN-O-10

**RELATIVE FIBRINOGEN AND HAPToglobIN DEFICIENCY IN DOGS WITH NATURAL OCCURRING ADDISON DISEASE.** A. Zoia<sup>1</sup>, M. Drigo<sup>2</sup>, M. Caldin<sup>1</sup>. <sup>1</sup>San Marco, Veterinary Clinic, PADUA, Italy, <sup>2</sup>Sanità Pubblica Veterinaria, Padua University, PADUA, Italy

Glucocorticoids are potent regulators of fibrinogen and haptoglobin biosynthesis in both humans and dogs and increase their secretion. Both proteins are synthesised by the liver and increase in humans and dogs with naturally occurring hyperadrenocorticism or following glucocorticoid administration. In addition, fibrinogen and haptoglobin are positive moderate acute phase proteins and their plasma/serum levels increase in inflammatory conditions due to increased hepatic synthesis stimulated by cytokines. There have been no studies into the effect on inflammation-mediated fibrinogen and haptoglobin synthesis in the absence of endogenous glucocorticoid. Naturally occurring Addison's disease in dogs is generally an immune-mediated disease causing impaired glucocorticoid production. The aim of this retrospective study was to assess whether dogs with Addison's disease have appropriate fibrinogen and haptoglobin concentrations for their level of inflammation [assessed by their serum C-reactive protein (CRP) concentration]. To evaluate this hypothesis, only dogs with newly diagnosed, naturally-occurring Addison's disease (diagnosed by an ACTH stimulation test), that had not received any treatment were included. Only dogs with FDPs and D-dimers

within the reference range were included, to exclude fibrinolysis and fibrinogenolysis as the cause of reduced fibrinogen concentrations. Serum samples with macroscopic haemolysis were also excluded as haemolysis may decrease haptoglobin concentrations. Two sick control animals with normal FDPs and D-dimers were included for each Addisonian dog included. The control dogs were matched for age, sex, and breed (or size if the breed was not found) and had serum CRP concentrations within  $\pm 0.5$  mg/dl of those of the Addisonian dog. As CRP concentrations could not be matched exactly, the fibrinogen/CRP and haptoglobin/CRP ratios were calculated to minimize the effects of inequalities in the degree of inflammation between each Addisonian dog and the two control dogs. Normality of data was assessed by the Shapiro-Wilk test. CRP, haptoglobin and fibrinogen concentrations were compared among the two groups using Student's *t*-tests and the ratios were compared using Mann-Whitney tests. Nineteen Addisonian dogs and 38 control dogs were included. All Addisonian dogs had CRP concentrations above the reference range. No difference in CRP concentration was found among the two groups ( $P=0.722$ ) but the fibrinogen and haptoglobin concentrations and the fibrinogen/CRP and haptoglobin/CRP ratios were significantly lower in the Addisonian dogs than the control population ( $P<0.001$ ,  $P<0.001$ ,  $P=0.01$  and  $P=0.002$ , respectively). These results suggest that fibrinogen and haptoglobin plasma/serum concentration is suboptimal when an inflammatory process occurs in dogs with no endogenous glucocorticoids.

#### EN-O-11

**LONG-TERM SURVIVAL OF DOGS WITH ADRENAL-DEPENDENT HYPERADRENOCORTICISM TREATED WITH MITOTANE VERSUS TRILOSTANE.** C. Arenas<sup>1</sup>, C. Melian<sup>2</sup>, A. Vicente<sup>1</sup>, M.D. Pérez Alenza<sup>1</sup>. <sup>1</sup>University Complutense, MADRID, Spain, <sup>2</sup>Clínica Veterinaria Atlántico, LAS PALMAS DE GRAN CANARIA, Spain

The treatment of choice for adrenal dependent hyperadrenocorticism (ADH) is adrenalectomy. When surgery is not possible, medical treatment with mitotane is the recommended alternative. Some studies have indicated that trilostane is a good option for canine ADH. The aim of this study was to compare the long-term survival of dogs diagnosed with ADH treated with mitotane versus trilostane.

This retrospective study included 26 dogs with ADH; 5 dogs were treated with mitotane using the non selective adrenocorticolysis protocol (NSAP), 9 treated with mitotane using the selective adrenocorticolysis protocol (SAP) and 12 treated with trilostane administered twice daily. Diagnosis of ADH was based on clinical and laboratorial data and abdominal ultrasonography, and confirmed by adrenocortical function tests. All dogs had a maximal dorso-ventral thickness of the tumour contra-lateral gland 5 mm. Mitotane (NSAP) was administered at 75-100 mg/kg/day for 25 days, and after the third day, hydrocortisone and fludrocortisone was provided lifelong. The animals on SAP protocol received 50-75 mg/kg/day of mitotane during the induction period; after successful induction, a maintenance dose of mitotane (75-100 mg/kg/week) was given. Trilostane was administered at an initial dose of 3 mg/kg/12 hours. Date and cause of death were recorded. Survival time was analyzed using a Kaplan-Meier survival curve and a Log Rank (Mantel Cox) test was performed to compare survival curves.

One dog on mitotane treatment was alive at the time of writing. Six dogs died due to unrelated diseases, 2 due to reasons attributable to the disease or its treatment and 5 cases for unknown reasons. All dogs on trilostane treatment were dead at censorship; 8 dogs died due to causes not directly attributable to ADH or its treatment, 3 due to the disease, and the remaining dog was lost for follow up. The mean ( $\pm$  SD) survival time in dogs treated with mitotane was  $15.4 \pm 3.2$  months (range 2-37.2; median 15.6 months) and for dogs on trilostane was  $17.7 \pm 4.2$  months (range 3.3-55; median 14 months). Differences between mean survival times were not statistically significant ( $p=0.873$ ).

Survival of dogs on mitotane treatment is similar than previously reported using the SAP protocol (16.4 months). Survival of dogs treated with trilostane administered twice daily is slightly

higher than that described using once daily doses (median 11.6 months). These results support the use of trilostane as a good alternative to mitotane for the treatment of canine ADH.

#### EN-O-12

**INTENSIVE INTRAVENOUS INSULIN THERAPY IN DIABETIC CATS.** M. Hafner<sup>1</sup>, E. Zini<sup>1</sup>, M. Osto<sup>2</sup>, M. Franchini<sup>3</sup>, M. Ackermann<sup>3</sup>, T.A. Lutz<sup>2</sup>, C.E. Reusch<sup>1</sup>. <sup>1</sup>Clinic for Small Animal Internal Medicine, <sup>2</sup>Institute of Veterinary Physiology, <sup>3</sup>Institute of Virology, Vetsuisse Faculty, University of Zurich, Switzerland.

Up to 25–50% of diabetic cats experience clinical remission. Studies in humans showed that remission rate increases after initial short-term intensive insulin treatment due to improved  $\beta$ -cell function. In cats, a study based on questionnaire to owners suggested that remission rate may increase with intensive insulin treatment. However, the protocol used required insulin dose to be adjusted on a daily basis, which is highly demanding. Aim of the study was to assess if intensive intravenous insulin treatment during the first week following diagnosis increases remission rate.

Twenty-eight cats with newly diagnosed diabetes were randomized to one of two treatment protocols. Cats in group 1 (n=14) received intravenous insulin aspart (Novorapid®, starting dose 0.05 IU/kg/h) adjusted to target glucose concentrations at 5–10 mmol/l. Cats in group 2 (n=14) received subcutaneous insulin glargine (Lantus®, cats  $\leq$  4 kg: 1.0 IU, q12h;  $>$  4 kg 1.5 IU, q12h). After hospitalization all cats were discharged with subcutaneous insulin glargine, q12h, and were fed a high-protein, low-carbohydrate diet (Purina Veterinary Diets® DM Diabetes Management). Rechecks and insulin dose adjustments were performed 1, 2–3, 6–8, 12–16 and 24 weeks after hospitalization. Remission was considered if cats were euglycemic without insulin for at least 4 weeks. Nonparametric tests were used for statistical analysis.

In group 1 remission was achieved in 9 (64%) cats, and in 6 (43%) in group 2. Remission rate was not significantly different between groups. In group 1, the median insulin dose given during the 6-month study period was significantly lower ( $p=0.015$ ) than in group 2 (group 1: 0.27 IU/kg/day; group 2: 0.44 IU/kg/day). Among cats that did not achieve remission, 3 (60%) in group 1 and 1 (12.5%) in group 2 were well controlled according to history, clinical and laboratory findings.

The intensive intravenous insulin therapy led to higher remission rate in diabetic cats. However significance was not reached, possibly due to the relatively small size of the groups. Cats receiving the intensive protocol required lower insulin doses after hospitalization, and metabolic control was better than in cats started on subcutaneous insulin. Intensive intravenous insulin therapy may contribute to improve  $\beta$ -cell secretion or peripheral tissue insulin sensitivity in diabetic cats.

#### EN-O-13

**ACCURACY OF CAPILLARY BLOOD 3- $\beta$ -HYDROXYBUTYRATE DETERMINATION FOR THE DETECTION AND TREATMENT OF CANINE DIABETIC KETOACIDOSIS.** F. Bresciani. Alma Mater Studiorum - University of Bologna, OZZANO DELL'EMILIA, BOLOGNA, Italy

Diagnosis and treatment of canine diabetic ketoacidosis (DKA) are usually based on semi-quantitative measurement of urinary acetoacetate (AcAc). In humans, blood 3- $\beta$ -hydroxybutyrate (3-HB) quantitative evaluation is considered more sensitive than urinary ketone measurement to detect and monitor the treatment DKA.

The aim of this study was to evaluate the accuracy of an electrochemical sensor (Optium Xceed Abbott) for the measurement of 3-HB from capillary and venous blood samples; furthermore, the potential utility of the capillary 3-HB measurement for monitoring DKA was assessed.

Twenty-eight patients were enrolled in the study: 19 dogs with diabetic ketosis (hyperglycaemia, ketonemia and/or ketonuria) and 9 dogs with DKA (hyperglycaemia, ketonemia and/or ketonuria and metabolic acidosis).

Eighty-two paired measurement of 3-HB of capillary and venous blood samples, analysed both by electrochemical sensor and enzymatic reference method (D-3-hydroxybutyrate, Randox, Ranbut), were performed. The patients with DKA were treated with fluid replacement and low dose intravenous insulin infusion. In DKA patients blood glucose was measured hourly, capillary 3-HB 4 hourly, venous blood gases analysis and urinary AcAc 8 hourly. Utility of capillary 3-HB measurement during DKA management was evaluated in 53 simultaneous measurements of capillary 3-HB, urinary AcAc, blood glucose and venous blood gases analysis. Data were analyzed by a Bland-Altman plot, Pearson and Spearman's correlation and ROC curve analysis. Significance was set at  $p<0.05$ .

A good agreement was detected between capillary and venous 3-HB measurement by electrochemical sensor and reference method. A significant negative correlation has been found between capillary 3-HB and pH ( $r=-0.40$ ) and bicarbonate ( $r=-0.46$ ) as well as between urinary AcAc and pH ( $r=-0.46$ ) and bicarbonate ( $r=-0.57$ ). A positive correlation was found between capillary 3-HB and anion gap ( $r=0.43$ ) and urinary AcAc and anion gap ( $r=0.42$ ).

Both capillary 3-HB and AcAc showed a good correlation with the degree of acidosis (pH, bicarbonate and anion gap), however 2/9 dogs with DKA (22%) had negative urinary AcAc at the diagnosis and during the treatment. A cut-off value of capillary blood 3-HB  $>$ 4 mmol/l revealed a 44% and 96% sensitivity and specificity (LR+ 12,44 and LR- 0,58) to diagnose DKA, respectively.

The electrochemical sensor accurately measures 3-HB concentration in both capillary and venous blood samples and it is accurate in diagnosing canine DKA. Capillary 3-HB measurement reflects the patient's metabolic status during canine DKA and seems superior to urinary AcAc in monitoring these patients; further studies in a wider population of DKA dogs are warranted.

#### EN-O-14

**ACCURACY OF SERUM FREE THYROXINE CONCENTRATIONS DETERMINED BY A NEW VETERINARY CHEMILUMINESCENT IMMUNOASSAY IN EUTHYROID AND HYPOTHYROID DOGS.** J.C.R. Scott-Moncrieff<sup>1</sup>, R.W. Nelson<sup>2</sup>, K.L. Campbell<sup>3</sup>, J.E. Robertson<sup>4</sup>. <sup>1</sup>Purdue University, WEST LAFAYETTE, United States of America, <sup>2</sup>University of California, DAVIS, United States of America, <sup>3</sup>University of Illinois, URBANA, United States of America, <sup>4</sup>IDEXX Laboratories, Inc., DAVIS, United States of America

Free thyroxine (fT<sub>4</sub>) is the unbound biologically active fraction of total thyroxine (TT<sub>4</sub>). The pituitary-thyroid axis functions to maintain fT<sub>4</sub> within a certain range and fT<sub>4</sub> may reflect thyroid function more accurately than TT<sub>4</sub>. Several methodologies are currently available to measure the concentration of fT<sub>4</sub> in serum.

The purpose of this study was to establish the reference interval in dogs for a new analog veterinary fT<sub>4</sub> immunoassay, IMMULITE® 2000 Veterinary Free T<sub>4</sub> (VfT<sub>4</sub>), Siemens Healthcare Diagnostics Products Ltd., Llanberis, Gwynedd, UK and to compare the accuracy of VfT<sub>4</sub> to the other fT<sub>4</sub> assays currently available for evaluation of dogs with clinical signs of hypothyroidism.

The VfT<sub>4</sub> was compared to 2 fT<sub>4</sub> radioimmunoassays by equilibrium dialysis, which included the Direct Free T<sub>4</sub> by Dialysis (fT<sub>4</sub>EDIVD), IVD Technologies, Santa Ana, CA, USA and the Free T<sub>4</sub> by Equilibrium Dialysis (fT<sub>4</sub>EDAN), Antech Diagnostics, Irvine, CA, USA. The VfT<sub>4</sub> was also compared to a two-step direct fT<sub>4</sub> immunoassay, Gammacoat™ Free T<sub>4</sub> (Two-Step) Radioimmunoassay (DfT<sub>4</sub>), Diasorin, Inc., Stillwater, MN, USA.

The study included 49 clinically healthy dogs, which were confirmed to be euthyroid based upon history, physical examination, CBC, serum biochemistries, and post-TSH stimulation serum T<sub>4</sub>

concentration  $\geq 32.2$  nmol/L. Free T<sub>4</sub> concentrations were determined by each method.

Fifty-six dogs with clinical signs of hypothyroidism were examined. Complete histories, physical examinations, CBCs, serum biochemistries, TT<sub>4</sub>, fT<sub>4</sub> by each method, and TSH stimulation testing were performed. Dogs were classified as either euthyroid (n=31) if post-TSH stimulation serum T<sub>4</sub> concentration  $\geq 32.2$  nmol/L or hypothyroid (n=25) if post-TSH stimulation serum T<sub>4</sub> concentration  $\leq 19.3$  nmol/L.

The reference interval of the Vft<sub>4</sub> was determined to be 7.7 - 47.6 pmol/L. For dogs with clinical signs of hypothyroidism, the sensitivities of the immunoassays were: Vft<sub>4</sub> (80%), fT<sub>4</sub>EDIVD (92%), fT<sub>4</sub>EDAN (71%), DfT<sub>4</sub> (96%). The specificities were: Vft<sub>4</sub> (97%), fT<sub>4</sub>EDIVD (90%), fT<sub>4</sub>EDAN (100%), DfT<sub>4</sub> (90%). Overall accuracies of the fT<sub>4</sub> tests were: Vft<sub>4</sub> (89%), fT<sub>4</sub>EDIVD (91%), fT<sub>4</sub>EDAN (86%), DfT<sub>4</sub> (93%).

Results of this study show that the accuracy of the new Vft<sub>4</sub> immunoassay is comparable to other currently available fT<sub>4</sub> assays for the diagnosis of hypothyroidism in dogs with clinical signs of this disease.

#### EN-O-15

**QUALITATIVE AND SEMI-QUANTITATIVE ASSESSMENT OF THYROXINE BINDING GLOBULIN IN THE GREYHOUND AND OTHER DOG BREEDS.** E. Shiel<sup>1</sup>, M. Nolan<sup>2</sup>, E. Nally<sup>2</sup>, T. Mooney<sup>2</sup>. <sup>1</sup>Murdoch University, PERTH, Australia, <sup>2</sup>University College Dublin, DUBLIN, Ireland

Thyroxine binding globulin (TBG) is the principal thyroxine (T<sub>4</sub>) binding protein in both dogs and humans. In humans, quantitative and qualitative variations in TBG are responsible for serum T<sub>4</sub> concentrations that differ significantly from standard reference intervals despite maintenance of euthyroidism. The aims of the current study were to characterise canine TBG, and to test the hypothesis that variations in TBG structure or serum concentration could contribute to the lower T<sub>4</sub> concentration observed in greyhounds when compared to most other dog breeds.

RT-PCR was used to assess qualitative differences in amino acid sequence of greyhound TBG precursor. The TBG coding sequence (CDS) was amplified from liver cDNA derived post-mortem from a greyhound with decreased total T<sub>4</sub> concentrations and a crossbreed dog. Both TBG sequences were compared to each other and to that predicted from the high quality whole genome boxer sequence. The greyhound TBG CDS was identical to the boxer sequence. The crossbreed TBG CDS contained an A/G non-synonymous SNP at position 526 that results in a conserved substitution (methionine for valine). The lack of cDNA variants unique to the greyhound made breed-restricted structural variations in TBG unlikely.

The predicted amino sequence of canine TBG was of identical length (395 amino acids) to, and had 83% identity with, its human analog, suggesting that anti-human TBG antibodies could cross-react with the canine protein. Canine and human serum proteins were separated by SDS-PAGE and immunoblots probed with polyclonal antibodies to human TBG (and HRP-conjugated secondary antibodies). Single bands of identical apparent molecular weight (approximately 51kDa) representing TBG were observed in both canine and human serum. Immunoblots were then performed on serum samples from 21 greyhounds and 21 non-greyhound dogs and reactive TBG quantified by densitometry, with optical density expressed relative to the mean of two standard samples included in every immunoblot. Values were compared using the unpaired t-test. There was no significant difference between mean TBG values (+/-SD) in greyhound and non-greyhound sera [1.02 (0.286) and 1.07 (0.364), respectively].

In contrast to previous reports, canine TBG is similar in size to its human analog and shares immunological cross-reactivity. Based upon our results, it may be possible to validate human TBG assays to allow accurate measurement of canine TBG concentrations. The absence of qualitative or semi-quantitative differences between greyhound and non-greyhound TBG in the current study makes variations in TBG structure or concentra-

tion unlikely to explain the lower T<sub>4</sub> concentrations observed in greyhounds.

#### HE-O-1

**ENDOVASCULAR TREATMENT AND/OR EVALUATION OF CANINE INTRAHEPATIC PORTOSYSTEMIC SHUNTS: SHORT- AND LONG-TERM EXPERIENCE IN 100 DOGS.** C. Weisse<sup>1</sup>, A. Berent<sup>1</sup>, K. Todd<sup>2</sup>, J.A. Solomon<sup>3</sup>. <sup>1</sup>Animal Medical Center, NEW YORK, United States of America, <sup>2</sup>Univ of Penn School of Vet Med, PHILADELPHIA, United States of America, <sup>3</sup>Hospital of Univ of Penn, PHILADELPHIA, United States of America

The purpose of this study was to retrospectively evaluate the short- and long-term results following endovascular management of canine intrahepatic portosystemic shunts.

100 dogs with congenital IHPSS received 112 procedures (80% had one treatment, 15% had >1 treatment, and 5% had 0 treatments due to excessive portal-central venous pressure gradients). Percutaneous vascular access and angiography identified 41 right divisional, 35 left divisional, and 19 central divisional shunts (5 not reported) of which 9% were complex/multiple shunts. Partial shunt attenuation was performed in 92 cases using caval stent placement and thrombogenic coils within the shunt while monitoring portal blood pressure. Complete acute shunt occlusion was possible in 3 cases. Major intra-operative complications (2/112; 2%) included temporary severe portal hypertension in one dog and GI hemorrhage in one dog. Major peri-operative (<1 week post-op) complications (12/110; 11%) included seizures/HE (6%), cardiac arrest (2%), jugular site bleeding (2%), pneumonia (1%), and acute death (1%). Median follow time for treated cases was 828 days (range 0-3411). Median survival time for treated dogs was 2204 days (range 0-3411) with 93% 60 day, 83% 1yr, 74% 2yr, and 63% 3yr survival rates. Outcome was considered excellent (48/90; 53%) or good (19/90; 21%) in 74% of treated dogs.

Endovascular treatment for canine intrahepatic shunts may result in lower peri-operative morbidity and mortality rates with similar success rates when compared with previously reported open surgical procedures. Gastrointestinal ulceration was a common finding among this population of dogs and life-long gastroprotectant medications are now recommended by the authors.

#### HE-O-2

**EVALUATION OF HEPATIC DISEASES IN IN VIVO DOGS WITH FIBROSCAN® DEVICE: A FEASIBILITY STUDY.** J. Oudry<sup>1</sup>, V. Miette<sup>1</sup>, P. Lecoindre<sup>2</sup>. <sup>1</sup>Echosens, PARIS, France, <sup>2</sup>Clinique vétérinaire des Cerisiez, SAINT PRIEST, France

One non-invasive quantitative medical device (Fibroscan®, Paris, France), based on ultrasound transient elastography (TE) technique, has emerged into clinical practice for diagnosing hepatic fibrosis in humans by means of liver stiffness as a marker for disease state. Motivated by the usefulness of this technique, the goal of this study was to investigate the feasibility of using the Fibroscan® device in dogs, certain pedigree are prone to chronic liver hepatitis and clinical tests are rarely evident until an advanced stage.

44 dogs (Scottish Terrier, Labrador, etc) were enrolled in the study after the owner consent. Dogs underwent a liver biopsy (METAVIR score), blood tests, ultrasound scan and Liver Stiffness Measurements (LSM). LSM was performed by TE in the left lobe of the liver under the ribs at the xiphoid with the dog standing. Fibroscan® device was used without any modification (software or hardware). It is based on a special probe: a shear wave is induced by a mechanical vibrator, its speed measured by an ultrasound transducer then the tissue stiffness is directly calculated. Operators were blinded to the results from the other tests during the measurements.

Results show the possibility to measure the liver stiffness of dogs with the TE device. A pilot study (8 dogs) was made with

encouraging results. Measurements were correct and the shear wave propagated well through the liver. Nevertheless, for the complete cohort, no significant correlation was demonstrated between liver stiffness and biological parameters (Spearman coefficient: 0.058, 0.223, 0.209 and 0.296 for ALT, AST, ALP and GGT respectively). Furthermore, no discrimination of liver elasticity/fibrosis stage can be performed with statistical analysis (box plots).

This work was the first feasible study of measuring in vivo dog liver stiffness with TE technique. Classification of tissue fibrosis stage or blood parameters compared to liver stiffness was not possible and remains challenging in dogs. Some stiffness data were widely dispersive. Indeed, the liver stiffness may be overestimated due to the dog liver anatomy compared to the human one and an inappropriate measurement procedure (dog movement, size, stress and jerky breath, no ultrasound imaging), the device being adapted to humans. However, TE may provide veterinary doctors with new important options for improving dog care regarding liver chronic diseases in terms of diagnosis and monitoring fibrosis progression. It could be of great interest and needs to be adapted to dog morphology and evaluated further.

### HE-O-3 OPTIMIZING DIAGNOSIS AND TREATMENT OF COPPER ASSOCIATED HEPATITIS IN THE LABRADOR. H. Fieten<sup>1</sup>, T.S.G.A.Mvanden Ingh<sup>2</sup>, T.S. Waalwijk<sup>1</sup>, K. Dirksen<sup>1</sup>, J. Rothuizen<sup>1</sup>. <sup>1</sup>Utrecht University, UTRECHT, The Netherlands, <sup>2</sup>TCCI Consultancy BV, UTRECHT, The Netherlands

**Introduction :** The best studied example of copper toxicosis in dogs is the Bedlington terrier. In this dog breed, a mutation in the COMMD1 gene causes severe liver copper accumulation.<sup>1</sup> In several other breeds, among which the Labrador, familial copper associated hepatitis has been recognized.<sup>2</sup> In the Labrador, pedigree analyses show a complex, rather than a monogenetic background. Labradors affected with hepatitis tend to have lower liver copper levels than affected Bedlington terriers. Conventional treatment in Bedlington terriers consists of lifelong chelation therapy with penicillamine, whereas in affected Labradors, lifelong treatment may be unnecessary.

Application of an accurate treatment protocol relies on objective quantification of liver copper. Histochemical scoring methods are accurate to detect increased liver copper levels, but often lack precision. Also interindividual variation in scoring between pathologists is a common problem. Other analytical methods like Instrumental Neutron Activation Analysis (INAA) and Atomic Absorption Spectroscopy (AAS) are often not available in clinical practice.

**Aims:** In the current study we aim to verify the use of a morphometric method for quantitative scoring of rubeanic acid stained liver histology slides. To determine optimal duration of penicillamine, patient files of Labradors treated with penicillamine were studied retrospectively.

**Methods:** Sections of liver biopsies from 25 Labradors and 15 Bedlington were stained with rubeanic acid (RA) and analyzed morphometrically by determining the ratio of RA stained surface to total liver surface. Correlation between quantitative copper determination and morphometry was calculated. Quantitative liver copper values of 40 Labradors before and after treatment with penicillamine, were used in a generalized mixed model to predict liver copper after a certain period of treatment.

**Results and conclusion:** Treatment duration until normalization of liver copper was strongly dependent on liver copper values at start of therapy. In order to get an optimal recheck interval and avoid unnecessary control liver biopsies, it is important to quantify the liver copper level at the start of treatment accurately. A good correlation was observed between morphometric quantitation and quantitation by INAA. Therefore, morphometry can be a practical solution in obtaining a reliable estimate for the amount of liver copper.

**References:** 1. Forman, O. P. et al. Characterization of the COMMD1 (MURR1) mutation causing copper toxicosis in Bedlington terriers. *Anim. Genet.* 36, 497–501 (2005).

2. Hoffmann, G. Copper-associated liver diseases. *Vet. Clin. North Am. Small Anim. Pract.* 39, 489–511 (2009).

### HE-O-4 COMPARATIVE BACTERIOLOGIC AND MOLECULAR EXAMINATIONS OF GALLBLADDER BILE OF HEALTHY DOGS. P.H. Kook<sup>1</sup>, S. Lutz<sup>1</sup>, I.M. Reichler<sup>2</sup>, M. Markel<sup>3</sup>, J.M. Steiner<sup>3</sup>, C.E. Reusch<sup>1</sup>, J.S. Suchodolski<sup>3</sup>. <sup>1</sup>Clinic for Small Animal Internal Medicine, ZURICH, Switzerland, <sup>2</sup>Section of Small Animal Reproduction, Clinic of Reproductive Medicine, ZURICH, Switzerland, <sup>3</sup>Gastrointestinal Laboratory, Texas A&M University, COLLEGE STATION, United States of America

In humans, gallbladder bile is generally surmised to be sterile. Comparable data are scarce in veterinary medicine. Although various potential pathogenic bacterial organisms have been cultured from liver biopsies in a large percentage of healthy dogs in one study, we could recently show that gallbladder bile of healthy dogs may only periodically harbour bacteria as detected by conventional culture techniques. However, it was suggested that a higher percentage of bactibilia might have been identified with molecular techniques. Therefore the aim of this prospective study was to compare culture and quantitative polymerase chain reaction (PCR) techniques on bile from healthy dogs in order to assess if PCR may be more sensitive for the detection of bactibilia compared to culture.

Gallbladder bile from 20 healthy dogs was collected by percutaneous ultrasound-guided cholecystocentesis (10) or during exploratory surgery (10). The dog's age ranged from 1-12 (median 5) years. Dogs were judged to be healthy based on history, physical examination, and results of CBC, serum biochemistry, and urinalysis. Prior antibiotic therapy to sampling was considered an exclusion criterion. Informed owner consent was obtained from all owners.

Bile samples were cultured aerobically and anaerobically within 2 hours of collection and aliquots were frozen for subsequent molecular testing. Three different DNA extraction methods were evaluated to identify the method with the highest sensitivity. *E. coli* was spiked into sterile saline (positive control) and canine bile and serial dilutions (1x10<sup>8</sup> CFUs down to 1x10<sup>4</sup> CFUs) were prepared. Bile- and water-spiked samples were extracted with Gene Releaser (BioVentures, Inc.), FastDNA kit (MP Bio), and a combined phenol chloroform and isopropanol precipitation protocol. Quantitative PCR assays were run on each sample in duplicates using universal bacterial primers. All 3 DNA extraction methods yielded a similar efficiency and analytical sensitivity, with a detection limit of 1x10<sup>5</sup> CFU per ml. Bile samples from healthy dogs were then extracted with the FastDNA kit (1 ml bile used) and analyzed by PCR. Bacterial culture results and PCR examinations were negative for all bile samples.

Our results indicate that canine bile appears to contain inhibitors leading to a PCR detection limit of 1x10<sup>5</sup> CFU in this study. At this point PCR does not appear to have any advantage over conventional bacterial culture techniques and future studies will need to employ more sensitive DNA extraction protocols to eliminate potential PCR inhibitors.

### HE-O-5 EMBOLIZATION OF EXTRAHEPATIC PORTOSYSTEMIC SHUNTS IN DOGS WITH A SINGLE COIL. A.S. Stosic<sup>1</sup>, S.B. Bayer<sup>1</sup>, M.S. Schneider<sup>1</sup>. <sup>1</sup>Small animal clinic (Internal Medicine and Surgery), GIESSEN, Germany

Coin embolization of extrahepatic shunts in dogs is rarely described. Major problems are coil migration and portal hypertension due to rapid thrombosis. The study hypothesis was that single proto-type coil implantation combined with heparin therapy is an effective treatment for extrahepatic shunts in dogs.

25 dogs with a single extrahepatic shunt were treated. A transvenous retrograde portography and portal vein pressure measurement was done. The shunt-diameter was sized by using

a 5F wedge catheter. Prototype coil diameter was selected larger than the blocked shunt diameter and was implanted through a 6F catheter. Before coil implantation, 100 IU/kg unfractionated heparin were injected into the portal vein, followed by continuous rate infusion (25 IU/kg/h) and subcutaneous injections (200 IU/kg q6h). Initially, the range of the activated coagulation time was adjusted at 150 to 200 seconds. The heparin application was modified based on development/resolution of ascites.

Most common breeds were Yorkshire-Terrier, West-Highland-White-Terrier and Jack Russell Terrier with a median age of 25.0 months (range 6.0 - 65.3) and a median body weight of 5.0 kg (range 2.1 - 9.0). 21/25 dogs showed neurologic signs in the history. The following shunt morphologies were found: porto-caval (n=17), porto-azygos (n=7), and porto-phrenico (n=1). During temporary shunt closure the portal vein pressure was > 20 cmH<sub>2</sub>O in 8 dogs, additionally 12 dogs showed indirect signs of portal hypertension. Coil implantation was successful in 23/25 dogs. Shunt perforation occurred in two small dogs with a shunt from the right gastric vein due to manipulation with the implantation catheter. One of these dogs died, the other one was treated with another technique. Three dogs died after coil implantation of different reasons (splenic rupture, pneumonia, intestinal ulceration). Two dogs showed seizures after the intervention. 3 dogs developed subcutaneous bleedings. Twenty dogs were discharged and presented with good clinical improvement for a 3 months follow-up. Fifteen dogs showed a normal ammonia-tolerance-test and a complete (n=12) or nearly (n=3) complete shunt closure in the portography. The remaining 5 dogs showed abnormal resting-ammonia (n=1) or abnormal ammonia-tolerance-test (n=4), no angiography was performed. At 12 months reexamination one dog was lost to follow-up, 3 dogs had a normal ammonia-tolerance-test despite small residual-shunting; the remaining dog had an abnormal ammonia-tolerance-test with moderate residual-shunting.

The size of the implantation catheter makes this coil inapplicable for smaller dogs with right gastric vein shunts. A normal ammonia tolerance test in 18/19 dogs indicates a promising long time success.

#### NU-O-1

**CHRONIC OBESITY DOES NOT ALTER INFLAMMATORY CYTOKINES BUT EVOKES AN IMPAIRED LYMPHOCYTE FUNCTION IN ADULT HEALTHY DOGS.** H.van deVelde<sup>1</sup>, G.P.J. Janssens<sup>2</sup>, E. Cox<sup>3</sup>, L. Tedin<sup>4</sup>, J. Zentek<sup>4</sup>, P. Nguyen<sup>5</sup>, V. Biourge<sup>6</sup>, M. Hesta<sup>2</sup>. <sup>1</sup>Ghent University, MERELBEKE, Belgium, <sup>2</sup>Laboratory of Animal Nutrition, Ghent University, MERELBEKE, Belgium, <sup>3</sup>Laboratory of Immunology, Ghent University, MERELBEKE, Belgium, <sup>4</sup>Institut für Tierernährung, Freie Universität Berlin, BERLIN, Germany, <sup>5</sup>Unité de Nutrition et Endocrinologie, Ecole Vétérinaire de Nantes, NANTES, France, <sup>6</sup>Royal Canin SAS, AIMARGUES, France

Obesity is a highly important nutritional disorder in all species, including companion animals. Overweight in humans and rodents induces immunological alterations, resulting in a decreased resistance towards infections and concomitant disorders. This trial investigated whether diet-induced chronic obesity alters immunological parameters in adult healthy dogs.

Sixteen adult beagles with a body condition score (BCS) of 3/5 (using a 5-point score system) were randomly assigned to a control (CG) and weight gain (WGG) group. Weight gain was induced by an energy dense diet (1759 kJ/100 gram dry matter), which was administered at 1.3 x maintenance energy requirement during 47 weeks (Weight Gain Period:WGP), where-after obese body weight was maintained during 26 weeks (Stable Period:SP). The CG was maintained with the same diet at stable ideal body weight during the entire trial (WGP + SP = 73 weeks).

Body weight and BCS were followed up weekly, while immunological parameters and leptin were measured at week 0-4-9-16-24-31-40-47-59-73. Body composition was determined using the deuterium-technique at week 0-20-35-47-73. Data were analyzed

using Repeated Measures for which significance was set at  $P < 0.05$ .

After the WGP, a significant higher body weight was reached in the WGG. During the SP, body weight was maintained. The CG remained at initial body weight during both WGP and SP.

A significant increase in absolute fat mass was noted in the WGG. Leptin (ng/ml) increased in the WGG from  $1.88 \pm 0.29$  to  $14.95 \pm 1.90$  during the WGP but dropped back to  $6.16 \pm 1.30$  in the SP. This adipokine remained at baseline level in the CG during both WGP (from  $2.34 \pm 1.05$  to  $2.04 \pm 0.19$ ) and SP ( $1.1 \pm 0.12$ ).

Proliferation of peripheral blood mononuclear cells (PBMC), stimulated by ConA, PHA and PWM was not altered during the WGP in both groups but was significantly lower in the WGG during the SP. Certain subtypes of PBMC (CD3, CD4, CD5 and MHC) were increased in the WGG at week 24, where-after it slowly decreased to be significantly lower at week 47, compared to the CG. Tumor necrosis factor  $\alpha$  (TNF- $\alpha$ ) was undetectable in CG and WGG.

Chronic obesity was successfully reached in this trial. Leptin and absolute fat mass run parallel with weight gain. From this study, it can be stated that lymphocyte function and phenotyping are altered during chronic obesity in adult healthy dogs, which might influence their body defence against infections and diseases.

#### NU-O-2

**CAMPYLOBACTER SPECIES AND MULTILOCUS SEQUENCE TYPES FROM COMMERCIAL RAW MEAT DIETS FOR PETS.** E. Acke, A. Midwinter, J. Collins-Emerson, N. French. Massey University, PALMERSTON NORTH, New Zealand

*Campylobacter* spp. are an important cause of zoonotic infections in humans, resulting in gastro-enteritis and a range of other disorders. The most common cause of infection is ingestion of undercooked or contaminated foods. *Campylobacter* spp. are commensal enteric pathogens in dogs and cats, but may also act as primary or secondary pathogens especially in young or immunosuppressed animals. Contact with pets is a risk factor for campylobacteriosis in humans. Feeding raw meat diets to dogs and cats has become the increasingly popular, and high isolation rates of *E. coli*, *Salmonella* and *Clostridium* spp. from raw meat diets, increased faecal shedding of *Salmonella* spp. by pets fed these diets, and raw meat feeding associated disease in pets has been reported recently. *Campylobacter* spp. from raw meat pet diets may be a previously underestimated source of infection for pets and pet owners who handle the meat and have close contact with pets.

Fifty samples of commercially packed raw meat diets for cats and dogs were purchased from supermarkets and pet stores in Palmerston North, New Zealand. Samples were purchased fresh cooled with or without preservatives, or frozen, stored as recommended at the laboratory, and analysed prior to the use by date. The origin of the meat was mainly beef, pork or chicken meat, or a mixture of meats. Selective enrichment media were used for isolation of *Campylobacter* spp. Positive isolates were confirmed and speciated by PCR.

Twenty one out of fifty raw meat diet samples (42%) were cultured positive for *Campylobacter* spp. Eleven of these were confirmed *C. jejuni* (52.4%), three were *C. coli* (14.3%). Multilocus sequence typing (MLST) was performed on the *C. jejuni* and *C. coli* isolates for cluster analysis with isolates from human, human retail food and animal origin to assess epidemiological significance.

*Campylobacter* spp., including *C. jejuni* and *C. coli* which are the species most commonly associated with clinical signs in humans, were detected in a large proportion of the raw meat diet samples in this study. Raw meat diets for pets represent a major public health risk and increased awareness is needed to prevent zoonotic infections through direct handling of the pet diets, and reduction of environmental contamination in households where these diets are fed.

**NU-O-3**

**EFFECTS OF WEIGHT LOSS IN OBESE CATS ON BIO-CHEMICAL ANALYTES RELATED TO INFLAMMATION AND GLUCOSE HOMEOSTASIS.** A.T. Tvarijonaviciute<sup>1</sup>, J.J. Ceron<sup>1</sup>, S.L. Holden<sup>2</sup>, P.J. Morris<sup>3</sup>, V. Biourge<sup>4</sup>, A.J. German<sup>2</sup>. <sup>1</sup>University of Murcia, MURCIA, Spain, <sup>2</sup>University of Liverpool, LIVERPOOL, United Kingdom, <sup>3</sup>WALTHAM Centre for Pet Nutrition, WALTHAM-ON-THE-WOLDS, United Kingdom, <sup>4</sup>Royal Canin Research Center, AIMARGUES, France

Obesity is common disorder in domestic cats and can predispose to a number of diseases. In humans, derangements in metabolic and inflammation-related substances occur, and these have been linked to the associated disease processes. However, it is unclear as to whether similar changes occur in cats, whether they can be improved with successful weight loss, and whether such analytes might influence outcome of a weight programme.

Thirty-seven overweight cats were studied, representing a range of ages and both genders. An individualised weight loss program was devised for each cat, and monitored until completion. Body fat mass was measured by dual-energy X-ray absorptiometry, whilst plasma concentrations of acute phase proteins (e.g. Hp, SAA), endocrine and metabolic substances (e.g. insulin, IGF-1, glucose, and adiponectin), and enzymes (e.g. BChE and PON1) were measured.

Weight loss in obese cats lead to significant increases in plasma adiponectin and IGF-1 concentrations, whilst insulin and homeostasis model assessment (HOMA) decreased significantly. Pre-weight-loss plasma PON-1, adiponectin, and IGF-1 were lower in cats that failed to complete weight loss than those who were successful, whilst glucose and fructosamine were significantly higher. Finally, multivariable linear regression revealed that loss of lean tissue during weight management was associated with pre-weight-loss plasma adiponectin concentration (lower adiponectin, more lean tissue loss).

In conclusion, a range of metabolic derangements occurs in feline obesity, and these may be linked to outcomes of weight loss programs. Weight loss can lead to changes in the profile of biochemical analytes, consistent with improving metabolic status. Of the biochemical analytes studied, low concentrations PON-1, adiponectin, and IGF-1 could be used as biomarkers to predict cases that may fail weight loss, whilst low adiponectin concentrations may also predict cats that may lose excessive lean tissue.

**NU-O-4**

**IMPACT OF TREATMENT WITH RECOMBINANT FELINE INTERFERON OMEGA ON RESOLUTION OF ANOREXIA OF UNKNOWN ORIGIN IN CATS.** T.A. Lutz<sup>1</sup>, D. Megahie<sup>2</sup>, M. Albouy<sup>2</sup>. <sup>1</sup>Institute of Veterinary Physiology, ZURICH, Switzerland, <sup>2</sup>Medical Department, Virbac, CARROS, France

Several anecdotal reports have recorded an increase in eating in anorexic cats after the use of recombinant feline interferon omega (reFeIFN-w; Virbagen Omega®, Virbac), and it has been postulated that this may be due to a direct interferon effect. To investigate this in more detail we performed a double-blinded, randomised, controlled trial.

30 client-owned cats with acute anorexia of unexplained origin and no significant alterations on routine biochemistry or haematology were included. Cats displaying hyperthermia and apathy were included, but cats with digestive disturbances, oral lesions, or having received any immunomodulating treatment in the previous 4 weeks were not eligible. Informed client consent was obtained. The cats were hospitalised and fresh food (Hills a/d) was offered every 12 hours. Any cat not eating within 24 hours of hospitalization or demonstrating deterioration was given intravenous fluids and antibiotics if required. The treatment group (n=15) received 1MU/kg reFeIFN-w, the placebo group (n=15) received equivalent injections of excipient reconstituted with diluent. This was repeated every 12 hours for up to three injections, but discontinued if the cat started to eat normally. Qualitative scores were recorded for parameters such as activity and behaviour. Quantitative recordings were made for body temperature, cumulative food intake and time from first injection to commencement of eating.

The primary outcome measure of time from first injection to first meal was significantly shorter in the treated group (p=0.0014, Pooled T-Test). Charting cumulative food intake revealed a significant difference between groups over the first 12 hours (p=0.025) but not thereafter. It was interesting to note that in the treated group, 6 cats received only 1 injection and 2 cats received 2 injections compared to 2 and 3 cats, respectively, in the placebo group. All other cats received 3 injections. Over the first 12 hours there was also a significantly greater improvement in the behaviour scores in the treated group (p=0.0343, Fisher's exact test). Body temperature decreased in both groups; commencement of eating was not linked to the reduction in body temperature but treated cats with an initial hyperthermia appeared to respond more frequently.

These results suggest that a more rapid return to eating may be an additional benefit in hyperthermic, anorexic cats receiving reFeIFN-w for viral infections. Any such benefit should be evident within 12 hours of the first injection because no additional effect from subsequent injections was noted.

**NU-O-5**

**WEIGHT LOSS IMPROVES OXYGENATION IN RELATION TO FAT LOSS DURING SEDATION IN OBESE PET DOGS.** I. Iff<sup>1</sup>, A.J. German<sup>2</sup>, S.L. Holden<sup>2</sup>, P. MacFarlane<sup>2</sup>, P.J. Morris<sup>4</sup>, V. Biourge<sup>5</sup>, M. Mosing<sup>6</sup>. <sup>1</sup>Veterinary Anaesthesia Services-International, WINTERTHUR, Switzerland, <sup>2</sup>University of Liverpool, NESTON, United Kingdom, <sup>4</sup>WALTHAM Centre for Pet Nutrition, WALTHAM-ON-THE-WOLDS, United Kingdom, <sup>5</sup>Royal Canin Research Center, AIMARGUES, France, <sup>6</sup>University of Zurich, ZURICH, Switzerland

Obesity has a negative impact on respiratory function in dogs made experimentally obese through overfeeding, and may increase the risk of complications during anaesthesia and sedation. This prospective clinical study examined the effect of obesity and subsequent weight loss on level of sedation, recovery from sedation, oxygenation and ventilation in pet dogs.

Nine overweight dogs were enrolled in a formalised weight loss programme involving dietary caloric restriction and exercise. Owners gave informed written consent, and both the WALTHAM Ethical Review Committee and University of Liverpool Research Ethics Committee (RETH000285) approved the study. Dual-energy X-ray absorptiometry was used to quantify body fat mass prior to and after weight loss. Dogs were sedated for this procedure with intravenous medetomidine (0.007mg.kg<sup>-1</sup>), butorphanol (0.1mg.kg<sup>-1</sup>) and midazolam (0.1mg.kg<sup>-1</sup>), and positioned in dorsal recumbency. The degree of sedation was scored using a published system (Kuusela AJVR 2001;62:1073) and time to standing after administration of atipamezole (0.035 mg.kg<sup>-1</sup>, IM) recorded. Monitoring consisted of pulse oximetry, starting oxygen supplementation where indicated (SpO<sub>2</sub><90%) via facemask, inspired oxygen was measured with a nasal catheter advanced to the level of the medial canthus of the eye. After 10 minutes, PaO<sub>2</sub> and PaCO<sub>2</sub> were measured from an arterial blood gas sample. The ratio of arterial to alveolar oxygen partial pressures (Pa/AO<sub>2</sub>) was calculated for each of the samples.

Median weight loss was 26% (range 13-37%) starting body weight (SBW), at a rate of 0.6% (0.4-1.7%) SBW/week. Median body fat mass was 43±4.56% and 28±4.2% pre- and post-weight loss, respectively. Median sedation score (pre: 17, range 10-26; post: 19, range 12-21; P=0.820) and time to recovery from sedation (head lift on clap: pre 37±10.1min vs. post 35±6.0min, P=0.937; time to stand: pre 42±12.6min vs. post 39±5.3min, P=0.875) were not different before and after weight loss. Oxygenation improved significantly after weight loss (Pa/AO<sub>2</sub> pre: 0.46±0.174 vs. post: 0.64±0.183, P=0.0223) but there was no change in ventilation (PaCO<sub>2</sub>pre: 6.6±1.52 kPa vs. post: 6.4±1.15 kPa; P=0.707). Multivariable linear regression identified Pa/AO<sub>2</sub> ratio to be positively associated with sedation score (high sedation score higher Pa/AO<sub>2</sub>; r=0.64, r<sup>2</sup>=0.41, P=0.006) and negatively associated with body fat mass (higher body fat, lower Pa/AO<sub>2</sub>; r=-0.58, r<sup>2</sup>=0.34, P=0.015).

Sedation resulted in impaired oxygenation, but not ventilation, in obese dogs lying in dorsal recumbency. The degree of impairment correlated weakly with fat mass, but oxygenation

improved after weight loss. Supplemental oxygen administration is recommended when obese dogs are sedated for clinical procedures.

#### NU-O-6

**EFFECT OF THE ADDITION OF SOLUBLE FIBERS TO DRY EXPANDED DIETS ON FECAL AND URINE WATER CONTENT IN HEALTHY CATS.** Y. Queau, I.vanHoek, L. LeVerger, Y. Soulard, V. Biourge. Royal Canin, AIMARGUES, France

It has been hypothesized that diets promoting fecal volume and moisture may decrease diuresis by diverting part of ingested water into feces. The goal of this study was to investigate the effect of increasing fecal moisture by the addition of soluble fiber on urine and fecal water excretion. Healthy adult colony cats were sequentially fed 2 identical dry expanded diets except for the sources of dietary fiber. The levels of total dietary fiber (TDF) were similar between the 2 diets (7.3% on a dry matter basis), but the main sources of fibers included cellulose, fructooligosaccharides and chicorei pulp for diet A (high in soluble fiber) and cellulose for diet B (high in insoluble fiber). Each diet was fed for a period of 14 days; 9 days of adaptation followed by 5 days of collection, during which cats were individually housed with daily recording of food and water intakes and collection of all feces and urine. Food amounts were adjusted to maintain the cats' weights. Water was offered *ad libitum*. Water content of the feces was determined by weight difference after desiccation in a drying oven. Water content of the urine was derived from the total urine weight. Data are presented as median [range]. Wilcoxon signed ranks test was used to compare the amount of water intake and water production in urine and feces between the two diets. Statistical significance was set at  $P < 0.05$ . Seven cats, aged 7.4 years [4.3-7.5] and weighing 4.2 kg [3.6-7.3], were included. Food intakes did not differ between diets A and B (81 [52-90] and 77 [52-100] kcal/kgBW/day respectively). Voluntary water intake was 17% greater in average for diet A (36.9 [18.4-41.0] g/kg/d) than for diet B (29.0 [16.7-36.5] g/kg/d) ( $P < 0.05$ ). Urinary and fecal water excretion with diet A (15.3 [12.0-23.6] and 4.4 [2.3-4.9] g/kg/d respectively) were also greater than with diet B (11.6 [9.4-18.9] and 2.0 [1.6-3.8] g/kg/d) ( $P < 0.05$ ). Urine specific gravity was lower with diet A (1.054 [1.040-1.059]) than with diet B (1.060 [1.045-1.067]) ( $P < 0.05$ ). Overall, including diet moisture, water intake and excretion were greater for diet A than diet B ( $P < 0.05$ ). This study shows that in cats, the addition of soluble fibers to a dry expanded diet results in higher water content in the feces; but also in the urine contrary to the initial hypothesis. Cats appear to be able to adjust their water intake to compensate for increased fecal water losses.

#### NU-O-7

**WEIGHT LOSS AND POST WEIGHT LOSS MAINTENANCE ENERGY REQUIREMENT OF OBESE COLONY CATS.** S. Serisier, S. Vialle, E. Martinez, S. Michel, V. Biourge. Royal Canin, AIMARGUES, France

The prevalence of overweight or obesity in cats is between 12.5% and 52% in Western countries. Overweight is associated with many health problems and dietary energy restriction remains its treatment of choice. Weight rebound after weight loss is a well-known phenomenon in humans and companion animals. Little is known about the post-weight loss maintenance energy requirement in cats, however. The aims of this study were to induce weight loss in obese colony cats and to estimate their post weight loss maintenance energy requirement to avoid weight rebound.

Ten obese colony cats (BCS = 5 on a 5-point scale, 9 neutered males and 1 entire female) undertook a weight loss program (Royal Canin Slimfit program) by using a moderate protein high fiber commercial dry diet designed to promote satiety (3090 kcal metabolisable energy/kg-1, 34% proteins, 9% fat, 23% dietary fibers, as fed). Initial energy allocation was calculated as 35 kcal

per kg of calculated target body weight (BW). Energy allocation was then adjusted to achieve weight loss rate between 1% and 3% per week (wk). Following reaching target weight, maintenance energy requirements were adjusted to maintain optimal weight within  $\pm 5\%$  and weekly followed during 23 weeks while cats were fed different diets. The study was approved by the Royal Canin Ethics Committee.

Mean ( $\pm$ SEM) weight loss was  $23 \pm 1\%$  (16-30%), mean rate of weight loss was  $1.13 \pm 0.07\%$ /wk (0.79-1.48%/wk), and the mean energy allowance to induce between 1%-3% weight loss was  $32 \pm 1$  kcal/kg of target BW (29-36 kcal/kg). Except for increase in food consumption speed, no behavior changes were observed during weight loss. The difference between the target BW (calculated) and the ideal BW (BCS = 3, estimated by observation) was  $6 \pm 2\%$  (-6 - 15%). The post-weight loss maintenance energy requirement was  $46 \pm 2$  kcal/kg of ideal BW (37-61 kcal/kg). Mean time necessary to establish the maintenance energy requirement after weight loss was  $6 \pm 1$  weeks. The mean variation of BW after 23 weeks of maintenance with different diets was  $-1.6 \pm 1.0\%$  (-5.3 - 5.5%).

This colony-based study showed that a moderate protein high fiber diet associated with the Royal Canin Slimfit program is suitable to induce weight loss in obese cats. As expected, the maintenance energy requirements following weight loss are low, most likely due to increased metabolic efficiency and could explain the commonly observed weight rebound.

#### NU-O-8

**ADJUNCTIVE DIRLOTAPIDE THERAPY FOR DURING DIET-BASED WEIGHT MANAGEMENT - A PILOT STUDY.** A.J. German<sup>1</sup>, S.L. Holden<sup>1</sup>, P.J. Morris<sup>2</sup>, V. Biourge<sup>3</sup>. <sup>1</sup>University of Liverpool, NESTON, United Kingdom, <sup>2</sup>WALTHAM Centre for Pet Nutrition, WALTHAM-ON-THE-WOLDS, United Kingdom, <sup>3</sup>Royal Canin Research Center, AIMARGUES, France

Although many obese dogs will successfully lose weight on a conventional weight management regime, involving dietary caloric restriction and physical activity, a proportion do not succeed. Dirlotapide is licensed for weight loss in obese dogs, but is not designed for use in conjunction with a conventional diet-based plan. However, it is not known whether using it in conjunction with a conventional weight loss regime will have an additive effect.

Five obese dogs (various breeds, ages and genders) undergoing conventional weight management at the Royal Canin Weight Management Clinic, University of Liverpool were included in the study. All dogs were severely overweight (median [range] start weight 47.1kg [14.2-51.2kg] body fat mass 49.3% [46.8-55.0%], body condition score 9/9 [9/9-9/9]), and had already commenced a weight loss regime. However, progress had been excessively slow (weight loss  $< 0.5\%$ /week) in all dogs, and all owners had cited excessive appetite as a key hurdle to progress. Dirlotapide was administered according to the manufacturer's recommendations, except that the current high-protein high-fibre weight loss diet was continued and the level of energy restriction unaltered. Dogs were reweighed every 2-4 weeks, and the dose of dirlotapide increased if weight loss was  $< 0.3\%$ /week.

Prior to dirlotapide, median (range) weight loss was 14.3% (4.3-25.2%) starting body weight (SBW), at a rate of 0.4% (0.3-0.5%) SBW/wk. However, no significant progress had been made over 67d (22-196d) when rate of weight loss was only 0.05% (0.00-0.34%) SBW/wk. After dirlotapide therapy, a further 8.7% (0.7-23.9%) SBW was lost, at a rate of 0.27% (0.02-0.47%) SBW/wk. Starting dose of dirlotapide was 0.05mg/kg/d in all dogs, increasing to 0.20 (0.10-0.45) mg/kg/d during therapy.

One of the five dogs reached its target weight, whilst a second was euthanased after 213d for an unrelated reason. In one dog, dirlotapide therapy was started on three occasions but the owner discontinued because of excessive side effects (vomiting and lethargy). The remaining two dogs lost some weight, but therapy was subsequently stopped at the request of the owner. One of these dogs was lost to follow up, whilst the second rebounded to its pre-dirlotapide weight. A further 239d of dirlotapide in this dog failed to produce any additional weight loss.

Adjunctive therapy with dirlotapide in a conventional canine weight loss strategy can assist weight loss, although outcome is variable and weight loss is slow. Further prospective randomised controlled studies are recommended to confirm the findings of this pilot study.

#### FE-O-1

**SCREENING OF APPARENTLY HEALTHY MIDDLE-AGED AND OLDER CATS.** G.F.A. Verjans<sup>1</sup>, D. Paepc<sup>2</sup>, L. Duchateau<sup>2</sup>, K. Piron<sup>2</sup>, L. Ghys<sup>2</sup>, S. Daminet<sup>2</sup>. <sup>1</sup>Ghent University, Belgium, GHENT, France, <sup>2</sup>Ghent University, GHENT, Belgium

The interpretation of health screening in aged cats is limited because information regarding clinical and laboratory abnormalities in older cats is lacking. This study was undertaken to describe physical examination (PE) abnormalities, routine laboratory test results and blood pressure measurements in a middle-aged and older cat population apparently healthy for the owner.

One hundred cats, 6 years or older, clinically healthy for the owner, free of medication for at least 2 months were included and categorised in 2 groups: 6 to 10 years and 11 years or older. First the systolic blood pressure (SBP) was measured according to ACVIM guidelines, followed by a complete PE. A fundoscopic exam and Schirmer tear test were performed. Complete blood count, serum biochemistry (including total thyroxine concentration), screening for FIV and FeLV, and urinalysis (including urinary sediment, urinary protein to creatinine ratio (UPC) and bacteriological culture), were assessed. The comparison between both age groups was based on the Fisher exact test (discrete variables) and on the linear fixed effects model (continuous variables) at a 0.05 significance level.

SBP exceeded 160 mmHg in 8 out of 100 cats. None showed signs consistent with systemic hypertension on fundoscopic exam. PE revealed a heart murmur in 11, one or two enlarged thyroid glands in 20, submandibular lymphadenopathy in 32 and gingivitis in 72 cats. The median tear production was 14 mm/minute (range 8.3-19.7) for the right eye and 13.5 mm/minute (8-19) for the left. Increased serum creatinine concentration was observed in 29 cases, hyperglycemia in 23 (3/23 having glucosuria) and an increased total thyroxine in 3 cats. Fourteen cats were FIV positive and none FeLV positive. Crystalluria was commonly detected (41/100), mostly amorph and less than 1 crystal per low power field (few). Struvite (in 12.2% of the cats with crystalluria) and calcium oxalate (in 7.3%) crystals were also observed. Cats of 11 years or older showed a significantly higher systolic blood pressure, heart rate, thrombocyte count, serum urea concentration, total bilirubin concentration and UPC; and a significantly lower PCV, albumin and calcium concentration and urinary pH, than cats between 6 and 10 years.

These results clearly indicate that abnormalities on PE and routine laboratory tests are common in apparently healthy older cats. This study underlines the need for regular health check and for refinement and determination of reliable age specific reference intervals.

#### FE-O-2

**PREVALENCE OF INFECTIOUS AGENTS IN CATS IN SOUTH AFRICA.** R. Lobetti<sup>1</sup>, M. Lappin<sup>2</sup>. <sup>1</sup>Bryanston Veterinary Hospital, BRYANSTON, South Africa, <sup>2</sup>Department of Clinical Sciences, Colorado State University, FORT COLLINS, United States of America

Although vector-borne agents and *Toxoplasma gondii* are common in cats, little is known about the prevalence of select agents in client-owned cats in South Africa.

The purpose of this study was to determine the prevalence of infectious agents in convenience collected blood samples from cats in the Johannesburg area. Whole blood and sera were obtained from 102 cats with a variety of disease conditions. Total DNA was extracted from the blood and assayed using PCR techniques for *Mycoplasma haemofelis*, '*Candidatus M. haemominu-*

*tum*', '*Candidatus M. turicensis*, Bartonella spp., *Ehrlichia* spp., *Anaplasma* spp. and using genetic sequencing to confirm the results. ELISAs were used to detect IgG and IgM serum antibodies to *T. gondii* and IgG serum antibodies to *Bartonella* spp.. In addition, associations between test results, patient characteristics, and haematological values were evaluated.

Of the 102 cats, 56 (55%) were positive in one or more of the assays. While haemoplasma DNA was amplified from 26 cats (*M. haemofelis*: 4 cats [3.9%]; '*Candidatus M. haemominutum*': 22 cats [21.6%]) and *Bartonella* spp. DNA was amplified from 8 cats (*B. henselae*: 5 cats [4.9%]; *B. clarridgeiae*: 3 cats [2.9%]), DNA of *Ehrlichia* spp. or *Anaplasma* spp. were not amplified. Of the cats, 24 (23.5%) were seropositive for *Bartonella* IgG and 18 (17.6%) were positive for *T. gondii* IgM (12 cats), IgG (8 cats), or both (2 cats).

The primary difference between test results, patient characteristics, and haematological values was seen in the *M. haemofelis* positive cats where all affected animals were young males and showed severe anaemia and thrombocytopenia. When the haemoplasma test results were combined, there were more positive males (18 cats) than females (8 cats).

The study concluded that *Bartonella* spp, haemoplasmas, and *T. gondii* are common in client-owned cats in the region. The results also emphasize that the diagnosis of feline vector-borne agents and *T. gondii* is difficult without the use of specific diagnostic tests as there are minimal patient characteristics or haematological changes that indicate infection. Further study of associations between these infections and clinical syndromes is indicated in the area.

#### FE-O-3

**RETROSPECTIVE ANALYSIS OF THE EFFECT OF LONG-TERM USE OF MELOXICAM ON LONGEVITY OF CATS WITH CHRONIC PAIN AND CHRONIC KIDNEY DISEASE.** R. Gowan<sup>1</sup>, R.M. Baral<sup>2</sup>, M. Gunew<sup>3</sup>, L. Johnston<sup>4</sup>, R.Malik<sup>5</sup>. <sup>1</sup>The Cat Clinic, MELBOURNE, Australia, <sup>2</sup>Paddington Cat Hospital, SYDNEY, Australia, <sup>3</sup>Creek Road Cat Clinic, BRISBANE, Australia, <sup>4</sup>Boehringer Ingelheim Animal Health, INSELHEIM, Germany, <sup>5</sup>Centre for Veterinary Education, SYDNEY, Australia

Chronic painful conditions, such as musculoskeletal disorders, affect the quality of life of cats and often require medical treatment. Chronic kidney disease (CKD) and chronic pain are common in the cat, and often coexist. However, impaired kidney function is currently listed as a contraindication on NSAID data sheets. The objective of this study was to investigate the impact of long-term use of meloxicam on longevity, when used to treat chronic pain in cats with pre-existing CKD.

The medical records of two feline-only practices were searched for cats that had been prescribed meloxicam long-term to treat chronic pain. Pre-existing CKD was considered to be stable by ruling out pre- and post-renal azotaemia, and by demonstrating minimal changes in creatinine value and/or bodyweight in the 4-8 weeks prior to instituting meloxicam therapy. Cats were included into the study if the meloxicam treatment duration was greater than 6 months and complete medical records were available for review. Biochemistry, urinalysis, body condition score and body weight were monitored regularly. Concurrent medical conditions and treatments were recorded.

48 cats with CKD had been treated long-term with meloxicam (Metacam® oral suspension). 15 cats had IRIS Stage 1, 28 cats Stage 2 and 5 cats Stage 3 chronic kidney disease, prior to treatment. After titration to the lowest effective dose, the median "maintenance dose" was 0.02 mg/kg daily [range: 0.01-0.06]. The median age of cats at start of treatment was 15.3 years old [range: 3.5-19]. The median treatment duration was 617 days [range: 179-1418]. Using the Kaplan-Meier method the median survival time of treated cats was estimated as 1159 days (95% confidence interval [853, 1540]). This compares favourably to published longevity of cats with CKD1,2. Of the 28 cats which had died at the time of analysis, only 3 cats had CKD listed as the cause of euthanasia or death.

These results suggest that long-term oral meloxicam treatment at 0.02 mg/kg does not reduce longevity of cats with pre-existent, stable IRIS stage 1-3 CKD. Therefore meloxicam can be

considered as a treatment for cats with chronic painful conditions and concurrent CKD. Careful monitoring of cats treated with long-term NSAID therapy is important.

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**IM-O-1**  
**IDENTIFICATION OF ANTIGENS OF LEPTOSPIRA REACTIVE WITH SERA FROM DOGS WITH ACUTE LEPTOSPIROSIS WITH AND WITHOUT PULMONARY HAEMORRHAGE.** S. Schuller<sup>1</sup>, M. Mckenna<sup>1</sup>, A. Schweighauser<sup>2</sup>, T. Francey<sup>2</sup>, J. Nally<sup>1</sup>. <sup>1</sup>University College Dublin, DUBLIN, Ireland, <sup>2</sup>Vetsuisse Faculty University of Berne, BERNE, Switzerland

Leptospiral Pulmonary Haemorrhage Syndrome (LPHS) is a severe form of leptospirosis which is increasingly recognised in both humans and dogs. LPHS is associated with high mortality rates. The pathogenic mechanisms are poorly understood. Studies in human patients and experimentally infected guinea pigs with LPHS have demonstrated antibody deposits in infected lung tissue in the presence of only small numbers of leptospires. We hypothesise that infection with *Leptospira* results in generation of autoimmune antibodies which mediate in part the pathogenesis of LPHS. This project aimed to characterize the antigens of *in vitro* cultivated *Leptospira* (IVCL) reactive with serum IgG from dogs with acute leptospirosis with and without LPHS and dogs with acute kidney injury (AKI) due to other causes.

Serum samples from 12 dogs with acute leptospirosis (n=8 LPHS, n=4 no LPHS) and 9 control dogs (n=9 AKI) were included. Diagnosis of leptospirosis was based on the presence of consistent clinical and clinicopathologic signs and either of the following: single microscopic agglutination test (MAT) titre of >1:800, a 4-fold increase in MAT titre or positive tissue PCR. A dyspnoea score (0 = no dyspnoea, 1 = mild, 2 = moderate, 3 = oxygen-dependent) and a radiographic score (0=no lesions; 1=mild; 2=moderate; 3=severe lesions consistent with LPHS) were given to each patient. Proteins from IVCLs were separated via 1-D gel electrophoresis and transferred to a PVDF-membrane for immunoblotting with dog sera. The degree and pattern of reactivity were compared between groups.

L. interrogans serogroup Australis was the most common positive serogroup (n=8). Three dogs showed equal MAT titres to serogroup Australis and Grippothyphosa. One dog was positive for serogroup Icterohaemorrhagiae. There was no difference in MAT titres or infecting serogroups between dogs with LPHS and dogs without LPHS. Serum IgG from dogs with acute leptospirosis showed a high degree of reactivity with IVCL antigens. Antigens with an apparent molecular mass of 32 and 41kDa were the most consistently reactive with serum IgG in infected animals. There was no difference in reactivity between dogs with and without LPHS. Serum reactivity was not correlated with dyspnoea or radiographic scores. In contrast, control sera showed very little reactivity with IVCL antigens.

Immunoblotting allowed for differentiation between dogs with acute leptospirosis and control dogs with AKI from other aetiologies. Serum IgG from infected dogs reacted most consistently with IVCL antigens at 32 and 41kDa. Reactive antigens will now be characterised using 2-D electrophoresis and mass spectrometry.

**IM-O-2**  
**NEW POTENTIAL BIOMARKERS FOR CANINE IDIOPATHIC PULMONARY FIBROSIS.** L. Lilja-Maula, M. Palviainen, H.P. Heikkilä, M.R. Raekallio, M.M. Rajamäki. Helsinki University / Faculty of Veterinary Medicine, HELSINKI UNIVERSITY, Finland

Canine idiopathic pulmonary fibrosis (IPF) is a chronic interstitial lung disease occurring particularly in West Highland White

terriers (WHWTs). Etiology is unknown. Main differential diagnosis is chronic bronchitis (CB). The objective of the present study was to analyze bronchoalveolar lavage fluid (BALF) protein expression to find potential biomarkers for IPF.

Differences in protein expression in BALF samples from six WHWTs with histopathologically confirmed IPF, from five dogs with CB and from four healthy laboratory beagles were studied by using two dimensional difference gel electrophoresis (2D-DIGE). Proteins that were up-regulated minimum 1.5 fold when compared to healthy controls were chosen to protein identification with liquid chromatography-tandem mass spectrometry (LC-MS/MS).

Statistically significant up-regulation was identified in eight proteins in dogs with IPF or CB. Five of them could be identified with LC-MS/MS. One of these proteins,  $\beta$ -actin, was unique to IPF group.  $\beta$ -actin is involved in cell structure, motility and integrity. Rise in  $\beta$ -actin may be due to increased cellular turnover in lung epithelium. Four other identified proteins, complement C3 (part of complement system), haptoglobin (an acute phase protein), apolipoprotein A-1 (anti-inflammatory role) and transketolase (enzyme involved in glucose metabolism) were up-regulated in both IPF and CB groups.

These findings can give new insights to pathophysiological mechanisms involved in IPF.  $\beta$ -actin may also work as a potential biomarker for IPF and help to differentiate IPF from CB. Future research is required to determinate the role of these proteins in disease pathogenesis and to confirm the results in a larger population.

**IM-O-3**  
**COMPUTED TOMOGRAPHIC IMAGING OF DOGS WITH LARYNGEAL OR TRACHEAL AIRWAY OBSTRUCTION.** T. O'Brien, L. Stadler, K. Hartman, S. Matheson. University of Illinois, URBANA, United States of America

The value of CT in dogs with upper airway obstruction has been limited by the presence of the endotracheal tube (ET) used to support general anesthesia. A visual laryngeal examination or endoscopy may not be suitable because of the requirement for sedation or general anesthesia and possible need for corrective surgery or tracheostomy before recovery from anesthesia. The purposes of the study were twofold; 1) describe the CT and three-dimensional internal rendered endoscopy findings in awake or sedated dogs with suspected upper airway obstruction and 2) evaluate the correlation between the CT imaging findings and the definitive diagnosis.

Seventeen dogs with clinical signs attributable to non-neoplastic obstruction of the larynx, trachea or large bronchi underwent computed tomography (CT) imaging. All dogs underwent head, neck and thoracic CT examination using a 16 slice helical CT scanner. CT was performed without general anesthesia using a positioning device (VetMouseTrapTM). Fifteen of the dogs were imaged without sedation or general anesthesia. Three-dimensional internal rendering was performed on each image set using appropriate CT software to create virtual endoscopic images. The images began rostral to the pharynx and continued through the lobar bronchi as indicated based on lesion localization on routine image planes. The final diagnosis was obtained by either visual laryngeal examination, endoscopy, video fluoroscopy or necropsy.

The CT and three-dimensional internal rendering accurately indicated the presence and cause of upper airway obstruction in all dogs. CT diagnosis of laryngeal collapse and laryngeal paralysis required three-dimensional internal rendering. CT findings indicative of laryngeal paralysis included failure to abduct the arytenoid cartilages, narrowed rima glottis and air filled laryngeal ventricles. Laryngeal collapse findings depended on the grade of collapse and included everted laryngeal sacculles, collapse of the cuneiform processes and corniculate processes, and narrowed rima glottis. Trachea abnormalities included hypoplasia, stenosis or collapse syndrome. The CT findings in tracheal hypoplasia consisted of a severely narrowed lumen throughout the entire length. Tracheal stenosis was represented by a circumferential decrease in tracheal lumen size limited to one region. Tracheal collapse syndrome was diagnosed by severe asymmetric

narrowing. Lobar bronchi collapse appeared in CT images as a narrowed asymmetric lumen diameter. CT imaging of unanesthetized dogs with upper airway obstruction compares favorably with traditional non-imaging diagnostic methods.

#### IM-O-4

**HIGH SERUM TRANSFORMING GROWTH FACTOR-BETA 1 CONCENTRATION IN WEST HIGHLAND WHITE TERRIERS: A KEY TO THE BREED PREDISPOSITION IN CANINE IDIOPATHIC PULMONARY FIBROSIS?** E. Krafft<sup>1</sup>, H. Heikkilä<sup>2</sup>, J. Jespers<sup>3</sup>, K. Mc Entee<sup>3</sup>, A.S. Lequarré<sup>4</sup>, D. Peeters<sup>1</sup>, M. Rajamäki<sup>2</sup>, C. Clercx<sup>1</sup>. <sup>1</sup>Faculty of veterinary medicine, LIÈGE, Belgium, <sup>2</sup>Department of Equine and Small Animal Medicine Faculty of Veterinary Medicine, HELSINKI, Finland, <sup>3</sup>Faculté de Médecine Université Libre de Bruxelles, BRUSSELS, Belgium, <sup>4</sup>European Commission (DG-RTD), BRUSSELS, Belgium

Canine idiopathic pulmonary fibrosis (IPF) is an interstitial fibrotic pulmonary disease of unknown etiology and pathogenesis, mainly described in middle-aged to old West Highland white terriers (WHWT). In human IPF, pathways involving transforming growth factor beta 1 (TGFB1), a cytokine with profibrotic properties, seem to be central in the pathogenesis and are considered as potential therapeutic targets. In veterinary medicine, serum TGFB1 concentration is elevated in both healthy WHWT and WHWT with IPF, as compared to healthy dogs of various breeds. This suggests the occurrence of a breed specificity that might be linked to the breed predisposition observed in WHWT for IPF. The aim of the present study was to determine serum TGFB1 concentration in middle-aged to old healthy dogs from terrier breeds other than WHWT and from non-terrier breeds, and to compare these results with those obtained in healthy WHWT.

Ten WHWT (mean age 9 years, range 3-14), 8 Jack Russell terriers (JR) (9, 5-12), 10 whippets (W) (8, 6-13), 8 old English sheepdogs (OES) (6, 4-10), 10 Malinois sheepdogs (MS) (6, 5-6) entered the study. Clinical exam and haematology were performed on all dogs, as well as serum biochemistry on dogs over 7 years and bronchoscopy and high-resolution CT scan on all WHWT. Serum TGFB1 concentrations were determined by ELISA (Mouse/Rat/Porcine/Canine TGFB1 Quantikine ELISA kit, R&D systems). Age and breed influences were analysed by covariance analysis (ANOVA 2). Results in the different groups were then compared using non-parametric test (Mann-Whitney).

No effect of age but a highly significant breed effect ( $p < 0.0001$ ) was shown. Serum TGFB1 concentration was significantly higher in WHWT (median  $\pm$  IQ 66.5ng/mL, 58.7-74.4) as compared to JR (33.8, 28.6-43.6;  $p = 0.009$ ), W (24.0, 15.3-31.6;  $p < 0.001$ ) and to MS (21.8, 20.1-27.9;  $p < 0.001$ ). The difference between WHWT and OES (49.5  $\pm$  5.8) was not significant but the power of the test was only 0.28.

Results of the present study show that breed variation occur in serum TGFB1 concentration, with high levels found in WHWT. Although serum TGFB1 measurements should be performed in larger groups of dogs and in dogs from other terrier breeds, these results suggest that TGFB1 might be one of the proteins involved in canine IPF pathogenesis, and related with the specific breed predisposition of the WHWT for the disease.

#### IM-O-5

**MATRIX METALLOPROTEINASE-2 AND -9 IN BRONCHOALVEOLAR LAVAGE FLUID OF DOGS WITH IDIOPATHIC PULMONARY FIBROSIS AND CHRONIC BRONCHITIS.** H.P. Heikkilä<sup>1</sup>, E. Krafft<sup>2</sup>, D.deLorenzi<sup>3</sup>, C. Clercx<sup>2</sup>, M.M. Rajamäki<sup>1</sup>. <sup>1</sup>University of Helsinki, HELSINKI, Finland, <sup>2</sup>University of Liège, LIÈGE, Belgium, <sup>3</sup>San Marco Private Veterinary Clinic, PADOVA, Italy

Idiopathic pulmonary fibrosis (IPF) is a chronic, progressive interstitial lung disease of unknown etiology affecting mainly the

West Highland white terrier (WHWT) and occasionally other breeds. IPF is characterized by an excessive deposition of extracellular matrix (ECM) components into lung parenchyma. The main differential diagnosis is chronic bronchitis (CB). Matrix metalloproteinases (MMPs) are proteases known to participate in the breakdown and remodeling of ECM. Our objective was to investigate gelatinolytic MMP-2 and -9 activities in dogs with IPF versus dogs with CB and healthy dogs.

In total 38 dogs were included in the study: 17 WHWTs, 1 Scottish terrier, and 1 lhasa apso with IPF, 8 dogs with CB (various breeds), and 11 healthy control WHWTs. Dogs underwent thorough examinations including bronchoscopy with bronchoalveolar lavage (all dogs), thoracic radiography ( $n = 36$ ), and high resolution computed tomography (HRCT) ( $n = 26$ ). IPF diagnosis was confirmed either by histopathology post mortem ( $n = 12$ ) or by HRCT ( $n = 7$ ). MMP-2 and MMP-9 activities were analyzed from bronchoalveolar lavage fluid (BALF) by gelatine SDS-page zymography, and the zymograms were evaluated for MMP activity with AlphaEase software.

BALF proMMP-9 activity was significantly elevated in dogs with IPF (median 0.44, interquartile range, IQ, 0.10-1.41) compared with dogs with CB (0.09, IQ 0-0.13),  $p = 0.046$ , and healthy dogs (0, IQ 0-0.02),  $p < 0.001$ . Dogs with CB had higher proMMP-9 activity than healthy dogs,  $p = 0.043$ . BALF proMMP-2 activity was significantly elevated in dogs with IPF (0.12, IQ 0-0.53) compared with dogs with CB (0, IQ 0-0),  $p = 0.011$ . Although dogs with IPF also had higher BALF proMMP-2 activity than healthy dogs (0, IQ 0-0.03), this difference was not statistically significant,  $p = 0.061$ . There was no difference in BALF proMMP-2 activities between dogs with CB and healthy dogs. Active MMP-9 was detected in 4 dogs with IPF, 1 dog with CB but in none of the healthy dogs. Active MMP-2 was only detected in 1 dog with IPF.

Our results indicate that there is an enhancement in BALF gelatinolytic activity characterized by the overexpression of MMP-9 in IPF, and, to a lesser extent, in CB. This finding suggests that the upregulation of MMP-9 participates especially in the pathogenesis of IPF possibly contributing to the pulmonary structural remodeling characteristic of the disease.

#### IM-O-6

**EVALUATION OF POLYMERASE CHAIN REACTION IN THE DIAGNOSIS OF CANINE LEPTOSPIROSIS: COMPARISON WITH SEROLOGIC TESTING IN 33 DOGS.** MHugonnard, Z. Djelouadji, C. Pouzot-Nevoret, A. Barthélémy, A. Kodjo, I. Goy-Thollot. VetagroSup, veterinary campus Lyon, MARCY L'ETOILE, France

Biological diagnosis of canine leptospirosis is currently based on Microscopic Agglutination Test (MAT). Polymerase Chain Reaction (PCR) may help the diagnosis. The objective of this study was to compare MAT and PCR assays from dogs with clinically suspected leptospirosis.

Thirty-three dogs with an initial differential diagnosis that included leptospirosis were prospectively enrolled in the study between 2008 and 2010. Each dog underwent a MAT assay (acute +/- convalescent titers) and a PCR assay on blood and urine (+/- organs). Two internists blinded to the results of biological assays reviewed history, vaccinal status, epidemiology, clinical signs at presentation, laboratory, imaging findings and clinical course for each dog. They defined 3 groups according to the level of clinical suspicion (0c-2c): 0c-probably not leptospirosis, 1c-possibly leptospirosis, 2c-probably leptospirosis. The cut-off value for a negative MAT result was 1/80. Titers less than 1/640 for vaccinal serogroups in vaccinated dogs were considered postvaccinal. Three groups were defined according to the level of biological suspicion (0b-2b): 0b-weak biological suspicion (negative MAT results or postvaccinal titers, all PCRs negatives), 1b-moderate biological suspicion (negative MAT results or postvaccinal titers, at least one PCR positive), 2b-high biological suspicion (seroconversion or single positive MAT results irrespective of PCRs results).

The median age of dogs was 7 years. Seventeen of 33 (52%) were males and 27/33 (82%) were vaccinated. At presentation, 21/33 (64%) dogs were azotemic. Azotemia was

associated with glucosuria in 13/21 cases and considered acute in 16/21 cases. Acute hepatitis was documented in 6 dogs and hemorrhagic syndrome in 11 dogs. Nineteen lethal outcomes occurred. The 33 cases enrolled were categorized as follows (group, n): 0c (7), 1c (16), 2c (10). In group 2c, 9/10 dogs had a high biological suspicion (group 2b; all PCRs positive for 3 dogs, one PCR positive for either blood, urine or organ for 5 dogs and all PCRs negative for 2 dogs). In group 0c, 3/7 had a weak biological suspicion (group 0b) and 4/7 had a moderate biological suspicion (group 1b). In group 1c, 4/16 dogs were categorized in the group 2b, 10/16 in the group 1b and 2/16 in the group 0b. With high clinical suspicion of leptospirosis, excellent agreement was observed with biological results as a whole.

With weak or moderate clinical suspicion, strong variations were observed in PCR results. In this situation, a positive PCR could indicate a carrier state or an atypical form of leptospirosis.

#### IM-O-7

**CYTOKINES IN STORED ERYTHROCYTE CONCENTRATES.** A. Smith, R. Corsi, M.A. McMichael, J.M. Herring, T.R. Ngwenyama, A. Galligan, M.A. O'Brien, A.N. Beloshapka, P. Deng, K.S. Swanson. University of Illinois, URBANA, IL, United States of America

Transfusion of erythrocyte concentrates may cause adverse effects in the recipient, particularly with packed red blood cells (pRBCs) stored >2 weeks. Pre-storage removal of leukocytes and platelets (leukoreduction, LR), improves clinical outcome in the human recipient. As blood ages during storage, alterations occur in the cells that may lead to the production of proinflammatory cytokines. We hypothesized that 1) Cytokine levels in supernatants from canine pRBCs increase with storage time and 2) Pre-storage LR attenuates formation of cytokines.

pRBC units (n=10) were collected from normal dogs and randomized to either be stored without LR (n=5), or to be subject to pre-storage LR (n=5). Supernatants were aseptically collected from the pRBC units weekly for 5 weeks, centrifuged at 1850g X 20 min to remove cells, aliquotted, and frozen. Supernatants were thawed and centrifuged at 19000g X 10 min. Cytokine concentrations were then batch assayed for canine cytokines by ELISA.

IL-10 levels were very low in all samples. TNF $\alpha$  levels were significantly higher in non-LR vs LR units, but levels did not increase with storage. IL-8 increased during the 35 day storage (range: 3-16 fold) in non-LR units, but did not change in LR units. IL-8 levels were similar between non-LR and LR units on storage days 0 thru 21, but were significantly higher in non-LR units on days 28 and 35.

There is marked formation of IL-8 during storage of canine pRBCs. Pre-storage LR attenuates the generation of IL-8, and decreases the amount of TNF $\alpha$  present.

#### IM-O-8

**EFFECTS OF NEBULISED N-ACETYLCYSTEINE ON PLETHYSMOGRAPHIC FINDINGS IN CATS.** S. Shibly, A. Galler, R.A. Hirt. University of Veterinary Medicine Vienna, VIENNA, Austria

N-Acetylcysteine (NAC) is a pharmaceutical with a multifaceted application spectrum utilised in human and veterinary medicine inter alia for its mucolytic and radical scavenging effects. However, numerous side effects have been reported in humans as well as asthma models in rats and guinea pigs, including bronchorrhoea, nausea, pruritus, urticaria, dizziness and bronchospasm. The latter is held responsible for fatal casualties in human patients and is attributed to an anaphylactoid reaction caused by non-immunological mechanisms via local histamine-release independent of tryptase suggesting a non-mast cell source, namely neutrophils and basophils. Hence,

warning notices concerning the use of NAC in veterinary literature are numerous, although only few specific studies for small animals have been conducted so far. Moreover, the role of histamine in feline bronchoconstriction is discussed controversially.

The goal of this study was to evaluate the safety of nebulised NAC in client-owned cats concerning possible bronchospasm. 21 cats without respiratory disease aged 2 months to 24 years (mean 6 years) and 8 cats with respiratory disease aged 8 months to 13 years (median 3 years) were placed in the chamber of a barometric whole-body plethysmograph (volume, 38 L), and box pressure changes associated with respiration were measured at baseline and after aerosol administration of 150 mg NAC. Airway reactivity was assessed by monitoring increases in enhanced pause (PENH), a unitless variable that measures bronchoconstriction as derived from dose-response curves, and by visual evaluation of these curves. Data of cats with respiratory disease were compared to data of healthy cats via student's t-test. A student's t-test for paired samples was used to compare basal PENH to PENH post NAC nebulisation among all cats. A p-value <0.05 was considered significant.

Basal PENH in healthy cats was  $0.44 \pm 0.22$  and did not differ (p=0.59) from values post NAC application ( $0.40 \pm 0.25$ ). In cats with respiratory disease, basal PENH was  $0.47 \pm 0.22$  and did not differ significantly from healthy cats (p=0.67) or to values post NAC nebulisation (p=0.15; PENH:  $0.67 \pm 0.22$ ). However, values of post-NAC-application-PENH differed significantly (p=0.012) between healthy cats and cats with respiratory disease. Furthermore, 1 cat with feline asthma showed signs of bronchoconstriction when nebulised with NAC. We conclude that short-term nebulisation of therapeutic doses of NAC does not cause airflow limitation in healthy cats, but should be used with caution in cats with respiratory disease.

#### IM-O-9

**PREVALENCE OF MYCOPLASMA SPP. IN BRONCHOALVEOLAR AND NASAL LAVAGE IN CATS WITH CHRONIC BRONCHIAL DISEASE.** KWeber, B. Schulz, P. Richter, R.S. Mueller, K. Hartmann. LMU München / Medizinische Kleintierklinik, MÜNCHEN, Germany

The pathogenesis of chronic feline bronchial disease (FBD) remains unclear. Persistent infections with *Mycoplasma* spp. are considered to be a contributing factor to chronic airway diseases in humans. The objective of this study was to determine the prevalence of *Mycoplasma* spp. in the upper and lower respiratory tract of cats suffering from chronic bronchial disease compared to cats without respiratory disease. Seventeen cats with clinical signs of FBD and 14 cats without respiratory disease were included in the study. Bronchoalveolar lavage (BAL) and nasal lavage were performed under sterile conditions on all cats. Cytological and routine bacteriological analysis was done on all samples. Furthermore, all samples were cultured for growth of *Mycoplasma* spp. and DNA was extracted from the lavage fluids and analyzed by PCR to detect feline-specific mycoplasmas. Statistical analysis was done using Fisher's exact test. *Mycoplasma* spp. could only be cultivated from two BAL samples from FBD cats and from two BAL and two nasal lavage samples from cats without respiratory disease. Six cats out of 17 with FBD and seven out of 14 cats without respiratory disease were positive for mycoplasmas by PCR in BAL. In nasal lavage samples, mycoplasmas could not be detected by PCR in cats with FBD, but in five cats without respiratory disease, which were also all positive in the BAL fluid. Sequencing of PCR products confirmed the detected species to be *Mycoplasma felis* in all PCR-positive samples. The prevalence of mycoplasmas was not significantly different between the two groups. Limitations of the study are the small group size and that the group without respiratory disease consisted of cats that were euthanized due to other diseases. *Mycoplasma felis* can be detected in cats with and without respiratory signs in the upper and lower airways and does not appear to be linked to FBD.

**IM-O-10**

**EFFICACY OF DOMPERIDONE FOR THE TREATMENT OF MILD AND MODERATE CASES OF CANINE LEISHMANIOSIS: CLINICAL AND IMMUNOLOGICAL SHORT-TERM FOLLOW-UP.** P. Gomez Ochoa<sup>1</sup>, D. Sabaté<sup>2</sup>, J. Homedes<sup>2</sup>, L. Ferrer<sup>3</sup>. <sup>1</sup>Veterinary Faculty of Zaragoza, ZARAGOZA, Spain, <sup>2</sup>Esteve veterinaria R&D. Laboratorios Dr. Esteve S.A., BARCELONA, Spain, <sup>3</sup>Dept. de Medicina i Cirurgia Animals, Universitat Autònoma de Barcelona, BARCELONA, Spain

Domperidone, a dopamine D2 receptor antagonist, has recently been included in the list of available anti-Leishmania drugs in the current consensus guidelines for treatment of canine Leishmaniosis1. Its mechanism of action is based on the activation of the cell-mediated immune response. The Nitro-blue tetrazolium reduction test (NBT) has been proposed as a valuable tool for the monitoring of cellular immunity in canine leishmaniosis2. The aims of this study were to evaluate the short-term efficacy of Domperidone in mild and moderate canine leishmaniosis by clinical and immunological follow-up, and to assess the eventual effects of treatment on the phagocytic function of polymorphonuclear cells and monocytes using the NBT.

Twenty dogs of different age, breed and sex, with mild and moderate clinical leishmaniosis (antibody titer =1/400-1/1600 using the Direct Agglutination Test and lymphadenopathy as the only clinical sign) were included. All dogs were orally treated with Domperidone at 0.5mg/kg/24h for 30 consecutive days. On days D0 (before treatment), D15, D30, D60 and D90 each dog underwent a clinical examination and blood sampling for serology and NBT.

Throughout the study, antibody titers decreased in 17 dogs (below the cut-off value =1/400 in two) and remained unchanged in two dogs. Lymphadenopathy decreased in 9 dogs (completely disappearing in four) and remained unchanged in eleven dogs. Baseline mean±SE percentages of activated (NBT-positive) neutrophils and monocytes were 10.2 ± 1.66 and 6.5 ± 1.34, respectively. These percentages rapidly increased after treatment initiation and they remained high until the end of the study, reaching their highest value on day D30 (43.3 ± 4.65 and 21.9 ± 2.11). Differences with baseline values were statistically significant (p<0.05) from day D15 onwards in both cell populations.

The results on clinical and immunological improvement are consistent with those reported in a previous study demonstrating that Domperidone is effective in reducing and controlling clinical signs and antibody titers of diseased dogs3. In addition, changes observed in NBT test are consistent with the mechanism of action described for Domperidone. The results of this study evidenced a close relationship between a favourable clinical evolution of dogs when treated with Domperidone and the increase of the NBT rate in blood neutrophil and monocyte populations, being this test a reliable partner in the clinical follow-up of leishmaniosis.

1 Oliva et al. (2010) J Am Vet Med Assoc. Jun 1;236(11):1192-8.

2 Gómez-Ochoa et al. (2010) Vet Parasitol. Aug 27;172(1-2):135-8.

3 Gómez-Ochoa et al. (2009) Vet J. Feb;179(2):259-63.

**IM-O-11**

**COMPARISON OF THE EFFECT OF HUMAN INTRAVENOUS IMMUNOGLOBULIN VERSUS VINCRIStINE ON PLATELET RECOVERY TIME IN DOGS WITH SEVERE IDIOPATHIC IMMUNE-MEDIATED THROMBOCYTOPENIA.** K. Balog<sup>1</sup>, A. Huang<sup>1</sup>, G.E. Moore<sup>1</sup>, S. Sum<sup>2</sup>, J.C. Scott-Moncrieff<sup>1</sup>. <sup>1</sup>Purdue University, WEST LAFAYETTE, United States of America, <sup>2</sup>University of Georgia, ATLANTA, United States of America

Dogs with severe immune-mediated thrombocytopenia (ITP) are at risk of spontaneous hemorrhage when the platelet count drops below 30,000/μL. While corticosteroids are the mainstay of treatment for ITP, adjunctive treatment may decrease the duration of thrombocytopenia. In separate prospective studies, adjunctive treatment with both vincristine and human intravenous immunoglobulin (hIVIg) has been shown to decrease

platelet recovery time compared to corticosteroid treatment alone. This prospective study was designed to compare the adjunctive effect of hIVIg versus vincristine on platelet recovery in dogs with severe ITP.

Twenty dogs with severe, idiopathic ITP (platelet count < 16,000/μL) were enrolled in the study. All dogs received standard care for ITP, including treatment with corticosteroids. Each dog was randomly assigned to receive a single, intravenous dose of either hIVIg (0.5 gm/kg) or vincristine (0.2 mg/kg) within 12 hours of enrollment. The age, sex, weight, breed and initial platelet count for each group were compared to ensure that the groups were equivalent in respect to signalment and disease severity. Outcome measures were platelet recovery time, defined as days required for the platelet count to reach 40,000/μL, and duration of hospitalization for each group.

There was no significant difference in age, sex, weight, or initial platelet count between the dogs treated with hIVIg and dogs treated with vincristine. The median length of hospitalization for all dogs was 4 days and did not differ between groups (p = 0.313). There was no significant difference between groups for platelet recovery time (p = 0.424) as the median time for both groups was 2.5 days.

This study failed to identify a significant difference in platelet recovery time for dogs with severe ITP receiving either hIVIg or vincristine, in conjunction with standard doses of corticosteroids.

**IM-O-12**

**INVESTIGATION OF RELATIONSHIP BETWEEN VITAMIN D STATUS AND MYCOBACTERIAL INFECTIONS IN CATS.** M. Lalor, R.J. Mellanby, D. Gunn-Moore. University of Edinburgh, EDINBURGH, United Kingdom

Mycobacterial disease in the domestic cat can result in several different syndromes including tuberculosis (*M. bovis*, *M. microti*), feline leprosy (*M. lepraemurium*) and opportunistic mycobacteriosis (e.g. *M. fortuitum*, *M. avium-intracellulare* complex). The diagnosis of mycobacteriosis typically depends on the detection of acid-fast bacilli in tissue biopsy and culture of mycobacteria which often takes several weeks. An interferon gamma test has been developed as a research tool to allow for more rapid detection of mycobacteriosis in cats. Management of all types of mycobacteria infection in cats tends to be complex often requiring long term therapy with frequent treatment failures.

In human medicine, vitamin D has been shown to play an important role in enhancing immunity to mycobacteria infection and there is growing evidence that vitamin D supplementation may enhance immunity to mycobacteria. The aim of this study was to measure serum 25 hydroxyvitamin D (25 (OH)D) concentrations in cats with mycobacteriosis, in ill hospitalised control cats and in healthy control cats.

This study comprised 89 cats divided into 3 groups. Group 1 consisted of 24 healthy cats. Group 2 consisted of 41 hospitalised ill cats with no history of gastrointestinal disease or recent corticosteroid use. Group 3 consisted of 24 cats with mycobacteriosis which were confirmed to be tuberculosis on culture or Ziehl Nielsen positive aspirates/biopsies with concurrent positive result with interferon gamma test. Blood samples were collected into plain tubes which were then centrifuged and the serum was separated and frozen at -20°C until 25(OH)D concentrations were measured by HPLC.

The median serum 25(OH)D concentration of healthy cats was 44.7 ng/ml (range 27.6-61.0, n=24), for hospitalised ill cats it was 33.8 ng/ml (range 10.6-53.5, n=41) and for cats with mycobacteriosis it was 22.15 ng/ml (range 9.7-54.8, n=24). There was a significant difference in the 25(OH)D concentrations between the three groups (p<0.0001). Post-test analysis revealed a significant difference between the 25(OH)D concentrations of healthy cats and cats with mycobacteriosis (p<0.001) and between healthy cats and hospitalised ill cats (p<0.001).

This study detected significantly lower concentrations of serum 25(OH)D in ill cats, with the lowest concentrations in cats with mycobacteriosis. Further studies are required to evaluate the effect of hypovitaminosis in clinical diseases such as mycobacteriosis.

**IM-O-13**

**EFFECT OF PROPENTOFYLLINE ON THE SURVIVAL TIME AND QUALITY OF LIFE OF CATS WITH FELINE INFECTIOUS PERITONITIS.** Y. Fischer<sup>1</sup>, S. Ritz<sup>1</sup>, K. Weber<sup>1</sup>, C. Sauter-Louis<sup>2</sup>, K. Hartmann<sup>1</sup>. <sup>1</sup>Clinic of Small Animal Medicine, MUNICH, Germany, <sup>2</sup>Clinic for Ruminants, MUNICH, Germany

Up to now, there is no drug proven to effectively treat cats with feline infectious peritonitis (FIP). The methylxanthine derivative pentoxifylline (PTX) has been shown to decrease TNF- $\alpha$  concentrations and improve vasculitis. In some case reports, PTX prolonged survival time in cats with FIP, and increased their quality of life. Propentofylline (PPF) is licensed for cats and is similar to PTX in its chemical structure and medical effects. Therefore, the aim of this study was to evaluate the efficacy of PPF in cats with a confirmed diagnosis of FIP.

The study was performed as a placebo-controlled double-blind trial. Twenty-three privately owned cats were included in the study. FIP was confirmed by histology or immunostaining of feline coronavirus (FCoV) antigen in effusion or tissue macrophages. Cats were randomly assigned to treatment with PPF or placebo. All cats received additional treatment with glucocorticoids. Parameters to evaluate efficacy were survival time, general condition characterized by the Karnofsky's score, physical and ultrasound examination, complete blood count and biochemistry profile, and TNF- concentrations measured with an enzyme linked immunosorbent assay (ELISA).

There was no statistically significant difference in the survival time of cats treated with PPF versus cats treated with placebo. The cats survived between 4 and 36 days (median, 8 days). There was neither a difference in quality of life, in the amount of effusion, the TNF- $\alpha$  concentration, nor in any other relevant parameter investigated in this study.

This study showed no effect of PPF on survival time, quality of life, or any clinical or laboratory parameter in cats with FIP. Therefore, PPF does not seem to be an effective treatment option, at least not in cats that are presented at a late stage of FIP when a definitive diagnosis can be achieved.

**IM-O-14**

**EFFICACY AND SYSTEMIC EFFECTS OF INHALED GLUCOCORTICOIDS IN NATURALLY OCCURRING FELINE ASTHMA AND CHRONIC BRONCHITIS.** A. Galler, S. Shibly, A. Bilek, R.A. Hirt. University of Veterinary Medicine Vienna, VIENNA, Austria

Despite recent studies investigating new therapy modalities, corticosteroids are still the mainstay therapy in feline chronic non infectious bronchial disease (feline asthma and chronic bronchitis). Inhaled glucocorticoids (iGC) allow for maximized local efficacy, while minimizing systemic effects. However, systemic absorption of iGC is not completely eliminated and is possible through the gastrointestinal tract, lungs, and nasal mucosa. The aim of this study was to evaluate owner adherence to therapeutic recommendations, acceptance by patients, efficacy, side effects and impact on the hypothalamus-pituitary gland-adrenocortical axis (HPAA) of long-term iGC therapy in naturally occurring feline asthma and chronic bronchitis. The study was performed with the approval of the ethics committee of the University of Veterinary Medicine Vienna as well as the Federal Ministry of Research and Science (bmwf: GZ 68205/118-II/10b/2010). Owners of 40 cats (34 with feline asthma, 6 with chronic bronchitis) treated with 400 $\mu$ g of inhaled budesonide twice daily were contacted, and information was retrieved by a questionnaire. Patients still on iGC were re-evaluated clinically and underwent barometric whole body plethysmography (BWBP) and ACTH-stimulation testing. Data of BWBP, namely baseline enhanced pause (Penh) and the carbachol concentration provoking a 300% increase of baseline

Penh (PC-Penh300) before treatment were compared to data under treatment using a paired student's T-test. In one half of the cats therapy had been withdrawn because of poor owner compliance (n=6), poor acceptance by patients (n=4), resolution of clinical signs with no relapse after drug discontinuation (n=4), lack of efficacy (n=4) or death due to unrelated reasons (n=2). Therapy duration in the 20 patients still on iGC ranged from 2 to 76 months (median 10 months). In the owners' opinion cats had improved (n=9) or clinical signs had resolved (n=11). Cats on iGC had significantly lower basal Penh (p=0.048) and higher PC-Penh300 (p=0.049) values than before treatment. In none of the cats corticosteroid-induced side effects were observed. HPAA suppression was detected in 3 of 14 patients (stimulated cortisol levels: 226,25 +/- 107.2 nmol/l). Treatment with iGC improved clinical signs and BWBP-results in cats with naturally occurring chronic non infectious bronchial disease. Although iGC have the potential to suppress the HPAA, treated cats had no clinical signs or biochemical alterations attributable to systemic corticosteroid effects.

**IM-O-15**

**A COMPARISON OF THE PACKED CELL VOLUME (PCV) AND TOTAL PLASMA PROTEIN (TPP) VALUES GAINED FROM CANINE LITHIUM HEPARIN AND EDTA BLOOD SAMPLES USING MICROHAEMATOCRIT AND REFRACTOMETRY METHODS.** A. Tamborini, S. Brennan, E.J. O'Neill. University College Dublin, DUBLIN, Ireland

In emergency situations, the measurement of the packed cell volume (PCV) and total plasma protein (TPP) are commonly performed as part of the initial evaluation of canine patients.

The aim of this study was to compare PCV and TPP in lithium heparin (LiH) and K3ethylenediaminetetraacetic (K3EDTA) canine blood samples obtained using a microhaematocrit reader and a hand-held refractometer with those gained using the ADVIA 2120 haematology analyzer and RANDOX Imola chemistry analyzer.

LiH and K3EDTA (n=43 each) canine blood samples were taken for routine haematological and biochemical analyses during the course of investigation of clinical cases presenting at the Veterinary Hospital of the University College of Dublin. 70  $\mu$ l LiH and EDTA blood were collected into two separate microtubes. Each tube was then centrifuged and a PCV obtained using a microhaematocrit reader and TPP determined using a hand-held refractometer.

D'Agostino and Pearson's omnibus normality test demonstrated that data for both PCV and TPP were normally distributed ( $\alpha < 0.05$ ). Linear regression analysis of PCV data indicated strong correlation (p<0.001) between the ADVIA 2120 and LiH PCV (r<sup>2</sup>=0.93), ADVIA 2120 and K3EDTA PCV (r<sup>2</sup>=0.92) and LiH and K3EDTA PCV (r<sup>2</sup>=0.92). Bland-Altman test analysis showed that bloods from LiH tubes consistently underestimated PCV with a mean bias of -0.69 (SD=2.14) and bloods from K3EDTA tubes underestimated PCV with a mean bias of -1.99 (SD=2.15). Paired t-test showed statistically significant differences between the means for ADVIA 2120 compared to LiH (p<0.05) and K3EDTA (p<0.001). Results for TPP demonstrated moderate correlation (p<0.001) between RANDOX Imola and refractometry results from LiH tubes (r<sup>2</sup>=0.62) and K3EDTA tubes (r<sup>2</sup>=0.61) but strong correlation between LiH and K3EDTA tubes (r<sup>2</sup>=0.94). Refractometry readings from LiH tubes and K3EDTA tubes consistently overestimated TPP with a mean biases of +2.51 (SD=0.77) and +2.59 (SD=0.77) respectively. Means for TPP from LiH and K3EDTA tubes differed significantly (p<0.001) compared to the RANDOX Imola. This study showed that a strong correlation exists between the PCV value obtained from LiH and K3EDTA tubes using the microhaematocrit method and ADVIA 2120 generated haematocrit but only moderate correlation was found for TPP obtained by refractometry compared with RANDOX Imola.

**IM-O-16**

**A NEW APPROACH TO PLEURAL EFFUSION IN DOGS: MARKERS TO DISCRIMINATE BETWEEN TRANSUDATES AND EXUDATES.** A. Zoia<sup>1</sup>, M. Drigo<sup>2</sup>, M. Caldin<sup>1</sup>. <sup>1</sup>San Marco, Veterinary Clinic, PADUA, Italy, <sup>2</sup>Sanità Pubblica Veterinaria, Padua University, PADUA, Italy

Traditionally in veterinary medicine, pleural effusions (PE) are classified as transudates, modified transudates and exudates based on the total protein (TPp) and total nucleated cell count of the fluid (TNCCp). The large and variable number of disorders associated with modified transudates and the fact that this category has overlapping TPp and TNCCp with transudates and exudates, limit this classification scheme. In human medicine, only two categories are used: transudates and exudates. The biochemical parameters used in human medicine to discriminate transudates from exudates have been shown to be reliable in cats for classifying PE and superior to those traditionally used. The aim of this retrospective study was to evaluate the ability of 9 biochemical parameters, used in human medicine, to distinguish transudates from exudates in dogs.

Parameters investigated were: Light's criteria [pleural fluid lactate dehydrogenase concentration (LDHp), pleural fluid/serum LDH ratio (LDHr), TPp, pleural fluid/serum TP ratio (TPr), pleural fluid cholesterol concentration (CHOLp), pleural fluid/serum cholesterol ratio (CHOLr), cholesterol gradient [serum cholesterol minus CHOLp (CHOLg)], albumin gradient [serum albumin minus pleural fluid albumin concentration (ALBg)], and TNCCp. Based on the final diagnoses, PEs were classified according their pathophysiology of formation into either transudates [effusions caused by congestive heart failure (CHF), protein losing enteropathy (PLE) and hepatic insufficiency (HI)] or exudates (effusions caused by neoplasia, pyothorax and miscellaneous causes of exudate formation). Chylous effusions were classified, in accordance with the human literature, as exudates. Normality of data was assessed by the Shapiro-Wilk test. Mann-Whitney tests were used to compare the TPp, CHOLp, CHOLr and CHOLg between transudates and exudates and Student's t-tests were used to compare LDHp, TNCCp, LDHr, TPr and ALBg. ROC curve analysis was then used for each parameter studied to establish the optimal cut-off point to maximise both sensitivity and specificity in differentiating a transudate from an exudate.

Forty six PEs were included; 16 transudates (12 secondary to CHF, 3 to PLE, and 1 to HI) and 30 exudates (18 secondary to neoplasia, 4 to pyothorax, 2 to chylothorax and 6 to miscellaneous causes). LDHp, LDHr, TPr, TPp, CHOLp, CHOLr, ALBg, and TNCCp were significantly different between the two groups (P<0.0001, P<0.0001, P=0.0024, P<0.0001, P<0.0001, P=0.0005, P=0.0068 and P<0.0001, respectively). LDHp, TNCCp and LDHr were the most accurate parameters to discriminate between transudates and exudates, with sensitivities of 96.7%, 96.6% and 100% and specificities of 100%, 100%, and 93.7%, respectively.

**IM-O-17**

**ACCURACY OF DIAGNOSTIC TESTS FOR FELINE INFECTIOUS PERITONITIS (FIP) IN CATS WITH BODYCAVITY EFFUSION.** S. Held<sup>1</sup>, M. König<sup>2</sup>, H.P. Hamann<sup>3</sup>, R. Senge<sup>4</sup>, E. Hüllermeier<sup>4</sup>, R. Neiger<sup>5</sup>. <sup>1</sup>Justus-Liebig-University Giessen, GIESSEN, Germany, <sup>2</sup>Diagnostic Laboratory, Institute of Virology, J-L-University, GIESSEN, Germany, <sup>3</sup>The Hessen State Laboratory (LHL), GIESSEN, Germany, <sup>4</sup>Department of Mathematics and Computer Science, Phillips-University, MARBURG, Germany, <sup>5</sup>Small Animal Clinic, J-L-University, GIESSEN, Germany

Even though there are several different tests available to diagnose FIP, a reliable diagnosis *in vivo* is still challenging. The aim of this prospective study was to evaluate the accuracy of eight tests for FIP in cats with body cavity effusion.

Between June 2009 and February 2011 70 cats with effusion (thoracic n=32, abdominal n=37, pericardial n=1) were examined. In each cat the following eight tests were performed:

immunofluorescence staining (IFA) of feline coronavirus (FCoV) antigen in macrophages in the effusion, polymerase chain reaction (PCR) to detect FCoV-RNA in the effusion, in EDTA-blood and in feces, albumin to globulin ratio (A/G) in the effusion and in serum, presence of antibodies against FCoV (titer) and the Rivalta test. If possible, immunohistochemistry of tissue sections was used as gold standard to diagnose FIP. The remaining unlabeled cats, for which no immunohistochemistry was available, were classified as either FIP positive or negative by means of a statistical method, namely through majority voting among an ensemble of 100 linear support vector machines (SVM) trained on the set of labeled cats using bagging as a resampling technique. In this way, all but three cats, for which the voting was not sufficiently close to unanimity, could be classified with high confidence.

17 cats were diagnosed with FIP, 50 without FIP. Sensitivity and specificity of the different tests were the following: IFA: 0.71 and 0.96, respectively; PCR in effusion: 1.00 and 0.98, respectively; PCR in EDTA-blood: 0.82 and 1.00, respectively; PCR in feces: 0.82 and 0.76, respectively; A/G in effusion (threshold: 0.6): 0.82 and 0.80, respectively; A/G in serum (threshold: 0.6): 0.88 and 0.66 respectively; anti-FCoV antibody titer (any detectable titer = positive): 1.00 and 0.68, respectively; and Rivalta's test: 0.53 and 0.92, respectively.

PCR in the effusion had excellent accuracy and might be used as single test to diagnose FIP in cats with effusion. Sensitivity and specificity of PCR in blood was higher than previously described, possibly due to using EDTA-blood instead of serum. If this is also the case in cats with non-effusive FIP needs to be further investigated. Interestingly, two FIP-negative cats were IFA positive, showing that this test might not be as helpful as previously thought. The Rivalta test, previously published to have excellent sensitivity, was much weaker in the present study indicating that this test should be used with caution in cats with effusion.

**RE-O-1**

**URETERAL STENTING FOR FELINE URETERAL OBSTRUCTIONS: TECHNICAL AND CLINICAL OUTCOMES: 74 URETERS (2006-2011).** A.C. Berent<sup>1</sup>, C. Weisse<sup>1</sup>, C. Letezia<sup>1</sup>, H. Bade<sup>1</sup>, D. Bagley<sup>2</sup>. <sup>1</sup>The Animal Medical Center, NEW YORK, United States of America, <sup>2</sup>Thomas Jefferson University, PHILADELPHIA, United States of America

The most common ureteral dilemma in feline patients is ureterolith-induced obstruction. Traditional therapy is associated with excessive morbidity/mortality. Our objective is to evaluate the technical and clinical outcomes in feline patients after double-pig-tail ureteral stent placement.

66 cats (74 ureters) had stent placement attempted for ureteral stone(s) (72%) or stricture (28%). 94% were successful surgically and 15.7% endoscopically. The overall success was in 70/74 (95%: 9 bilateral and 56 unilateral). The median number of stones per ureter was 6 (range, 1->40), and 86% had concurrent nephroliths. 85% were azotemic pre-operatively (median creatinine of 4.7 mg/dL [1.1-16.7]) and 65% were azotemic post-operatively (median creatinine of 2.5 mg/dL [1.0-11.8]). Procedure-related, peri-operative (<7days), short-term (7-30days) and long-term (>30days) complications occurred in 7%, 10%, 3%, and 28% of cats, respectively. Dysuria was detected in 38.5%, only persisting in 7%. The peri-operative mortality rate was 5/62 (8%), none of which due to ureteral disease or CKD. The median survival time was >321 days (53% still alive; range 2-1606 days). 20.9% died of suspected CKD (range, 40-590 days). Stent exchange (n=15) or placement of a SUB (n=4) was necessary in 19/70 ureters due to stricture in-growth (n=5), development of a surgical induced-ureteral stricture (n=6), stent migration (n=3), proliferative ureteritis (n=2), severe pollakiuria (n=2), or ureteral reflux (n=1).

Ureteral stenting is effective for treatment of feline ureteral obstructions regardless of stone number or location. Short-term complications are rare but long-term complications necessitating stent exchange should be monitored for, particularly with strictures. The prognosis after ureteral stenting could be considered good.

**RE-O-2**

**THE USE OF A SUBCUTANEOUS URETERAL BYPASS DEVICE FOR URETERAL OBSTRUCTIONS IN CATS.** A.C. Berent<sup>1</sup>, C. Weisse<sup>1</sup>, H. Bade<sup>1</sup>, C. Letezia<sup>1</sup>, D. Bagley<sup>2</sup>. <sup>1</sup>The Animal Medical Center, NEW YORK, United States of America, <sup>2</sup>Thomas Jefferson University, PHILADELPHIA, United States of America

Ureteral obstructions are difficult to treat and traditional therapy is associated with substantial morbidity/mortality. Nephrostomy tubes are effective but externalized drainage is the major risk. The development of an indwelling ureteral bypass using a combination locking-loop nephrostomy/cystostomy tube was created. The objective is to describe the technical and clinical outcome using of a novel device called a subcutaneous ureteral bypass (SUB) in cats and dogs with ureteral obstructions when traditional therapies have either failed or were contraindicated.

Eighteen cats (19 kidneys) and 1 dog (1 kidney) had a SUB placed for: ureterolithiasis (4), ureteral stricture (13), and ureterolithiasis with stent rejection (3). The median pre- and post-procedure creatinine was 6 (range: 1.7-13) and 2.7 mg/dL (range: 1.6-7), respectively. The median pelvis diameter measured pre and post-procedure were 15 (range: 7-28) and 5mm (range 1.9-10), respectively. Six french tubes were placed in 16, and 5 french in 3, and 8 french in 1. The bypass remained in place for a median of >237 days (range 6->627) and all were patent at last follow up. There were 3 major complications (15%) resulting in nephrostomy tube leakage (n=2), port leakage (n=1), and tube occlusion with blood clot (n=1). There were no short-term (7-30 days) or long-term (>1 month) complications.

Overall, the use of a SUB for cats with ureteral obstructions can be considered a functional option when other therapies have failed or are contraindicated, but should be considered a salvage procedure at this time.

**RE-O-3**

**RETROSPECTIVE STUDY OF URETERAL ECTOPIA IN MALE DOGS: TEN CASES (2002-2011).** M. Canonne-Guibert, H. Combrisson, M. Manassero, E. Gomes, C. Maurey. National Veterinary School of Alfort, MAISONS-ALFORT, France

Very few reports of ureteral ectopia exist in male dogs; this refers to a congenital abnormality characterized by termination of 1 or both ureters at a site distal to the bladder trigone. It represents the most common cause of urinary incontinence in young dogs and is reported to be > 20 times as common in female. This study purposes to review epidemiological features in the male dog by means of a retrospective study recruiting 10 dogs between January, 2002 and February, 2011.

The breeds include Golden and Labrador Retriever (4), Boxer (1), French Bulldog (1), English Bulldog (1), and American Staffordshire Terrier (1). Affected dogs range in age at evaluation from 3 to 108 months (mean, 22.7; median, 9.1). The predominant clinical feature is an intermittent or continuous dribbling of urine (9), since acquisition for 8 dogs and from 3 years of age for the remaining case. Incontinence seems worsened by excitement (3) or when asleep (2). For one male, urine dribbling occurs only on sleeping place. All dogs display normal voiding patterns, even with bilateral involvement. Definitive diagnosis is established by abdominal ultrasonography in all dogs (with or without furosemide).

Unilateral disease is the most represented; 11/13 ureters run intramurally. For all dogs, ectopic opening is in prostatic urethra. Ipsilateral hydronephrosis and hydronephrosis are displayed in 5 cases and an ureterocoele identified for 3 dogs. Renal function is always normal. Concomitant urinary tract infection is detected in 3 dogs. Urethral sphincter incompetence is confirmed in 2 males by urodynamic studies. Surgical techniques consisted of ureteroneocystostomy or nephro-ureterectomy in case of ipsilateral hydronephrosis and hydronephrosis. Treatment with oxybutinine (1) or phenylpropranolamine (2) is prescribed successfully for a post-operative incomplete improvement.

It is the first recent study gathering so many male dogs with ureteral ectopia. The breed predispositions, the older age at evaluation than females and the diagnostic accuracy of diuretic-enhanced ultrasonography are confirmed. The anatomic features

of ectopic ureters are similar to those of females. This condition remains infrequent in the male; the true prevalence is certainly underestimated because of the longer urethra providing a stronger muscular sphincter caudal to the ectopic orifice, so preventing urinary incontinence. Nevertheless, to our knowledge, this is the first report characterizing urethral sphincter incompetence in male dogs with ureteral ectopia by urodynamic data. This condition hampers a complete post-surgical resolution of incontinence and urethral pressure profiles could be used prognostic indicators.

**RE-O-4**

**REGIONAL CITRATE ANTICOAGULATION FOR EXTRACORPOREAL BLOOD PURIFICATION TECHNIQUES IN DOGS.** T. Francey, ASchweighauser. Vetsuisse Faculty University of Berne, BERNE, Switzerland

Extracorporeal blood purification techniques such as hemodialysis (HD) require anticoagulation of the circulating blood. The most common protocol uses systemic heparinisation that can however hinder urgent surgical procedures or worsen existing hemostatic disorders such as disseminated intravascular coagulation or leptospirosis-associated pulmonary hemorrhages. Regional anticoagulation techniques have thus been developed in humans for critical patients treated with continuous renal replacement therapy. One technique aims at reducing the ionized calcium concentration in the extracorporeal circuit (< 0.4 mmol/l) by infusing trisodium citrate in the arterial line and restoring calcemia (> 0.8 mmol/l) with calcium chloride administration in the venous line prior to returning the blood to the patient. The aim of this retrospective study was therefore to evaluate the adequacy of a canine protocol of regional citrate anticoagulation (RCA) in intermittent HD for acute kidney injury (AKI).

The RCA protocol was based on established human protocols and on in vitro pilot experiments. 211 HD sessions have been performed with Gambro AK200 UltraS system in 55 dogs treated for acute leptospirosis (n=33), toxic nephrosis (n=14), or other causes of AKI (n=8) following individually adjusted standard HD protocols. The initial flow ratio of blood : citrate (102 mmol/l) : calcium (340 mmol/l) was 10 ml/min : 15 ml/h : 1.5 ml/h and it was adjusted based on the ionized calcium concentrations in the circuit and in the animal. Satisfactory anticoagulation was assessed based on successful completion of the procedure, change in the dialyzer pressure gradient, urea and creatinine reduction ratios (URR, CrRR), and visual scoring of the extracorporeal circuit after blood rinseback.

The initial citrate and calcium infusion rates required adjustments in 27% and 44% of the treatments, respectively. Anticoagulation was judged overall satisfactory in 93% of the treatments. Four HD treatments (2%) hindered by severe catheter malfunction had to be stopped early due to severe clotting. The dialyzer pressure gradient increased >25% from baseline in 14% of the treatments. The extracorporeal circuits were considered moderately and severely clotted in 3% and 1% of the treatments, respectively. URR and CrRR were >25% below the expected ratios in 10% and 17% of the treatments, respectively. No clinical or laboratory side effect were observed.

With the described protocol of RCA, extracorporeal circulation could be safely and efficiently performed in dogs without the need for systemic heparinisation, representing a major advance in the treatment of animals at risk of bleeding.

**RE-O-5**

**INTOXICATION WITH GRAPES OR RAISINS CAUSING SEVERE ACUTE KIDNEY INJURY AND NEUROLOGICAL SIGNS IN DOGS.** A. Schweighauser<sup>1</sup>, D. Henke<sup>1</sup>, A. Oevermann<sup>1</sup>, B. Gerber<sup>2</sup>, T. Francey<sup>1</sup>. <sup>1</sup>Vetsuisse Faculty, University of Berne, BERN, Switzerland, <sup>2</sup>Vetsuisse Faculty, University of ZURICH, ZURICH, Switzerland

Ingestion of grapes or raisins has been reported to cause acute kidney injury (AKI) in dogs. Clinical signs typically include

vomiting, diarrhea, anorexia, lethargy, rapidly developing uremia, and possibly oligo-anuria. Ataxia was reported in a retrospective review and in a few case reports, however precise clinical description, localisation and evolution of neurological signs are missing in the literature. The aim of this study was to evaluate clinical, clinicopathological, and pathological data of dogs diagnosed with grape or raisin toxicity with particular emphasis on renal and neurological manifestations.

Fifteen dogs were diagnosed with grape or raisin toxicity between November 2009 and March 2011. All dogs were presented with severe AKI with a mean creatinine at presentation of 1069  $\mu\text{mol/l}$  (range 599-1743), urea 56.4  $\text{mmol/l}$  (35.7-77.8), total calcium 3.2  $\text{mmol/l}$  (2.25-3.97), ionized calcium 1.1  $\text{mmol/l}$  (0.88-1.29), and phosphorus 4.5  $\text{mmol/l}$  (2.51-7.93). Twelve dogs were treated with hemodialysis as part of their therapy (range 2-9 treatments).

Eleven dogs (73 %) showed marked neurological signs, including moderately to severely reduced consciousness (n=9), seizures (n=2), generalized ataxia (n=11), hypermetric gait (n=2), and tremor (n=2). Based on these findings, the lesions were considered multifocal intracranial (cerebrum, cerebellum) in all dogs. These signs were observed at initial presentation in 10 dogs. Systemic hypertension (systolic blood pressure > 160 mmHg) was present in 4/15 (26%) dogs with a range of 163-174 mmHg, but neurological signs in all 4 dogs persisted despite successful medical control of blood pressure. Similarly, neurological manifestation did not improve immediately with correction of azotemia. Laboratory parameters were not statistically different between dogs with or without neurological signs.

Overall 8/15 (53%) dogs survived. All had at least partial renal recovery (creatinine below 270  $\mu\text{mol/l}$ ). Of these, 4 had neurological signs during their acute phase of illness and showed complete neurological recovery. Non-survivors died or were euthanized for renal related reasons (n=4) or other non-neurological causes (n=3). Necropsy of 4 dogs with neurological signs revealed toxic nephrosis consistent with grape toxicity. Histopathology of the brains were unremarkable.

Severe neurological signs seem to be an important feature of intoxication with grapes or raisins and may even predominate the early clinical picture, mimicking ethylene glycol intoxication. These signs are reversible and likely unrelated to the degree of uremia. The pathophysiology of the neurological dysfunction could include direct neurotoxicity, electrolyte or mineral disturbances or increased intracranial pressure.

#### RE-O-6

**EXCITEMENT DURING EXAMINATION AT A CLINIC AFFECTS RENAL FUNCTION IN HEALTHY DOGS.** K. Hoglund<sup>1</sup>, S. Hanås<sup>2</sup>, C. Carnabuci<sup>1</sup>, I. Ljungvall<sup>1</sup>, A. Tidholm<sup>3</sup>, J. Häggström<sup>1</sup>. <sup>1</sup>Swedish University of Agricultural Sciences, UPPSÅLA, Sweden, <sup>2</sup>Strömsholm Animal Hospital, STRÖMSHOLM, Sweden, <sup>3</sup>Albano Animal Hospital, DANDERYD, Sweden

Renal function may be altered by excitement during examination at a clinic, which may affect the outcome of diagnostic tests. The aim of the study was to investigate if glomerular filtration and renal concentration mechanisms are affected during clinical examinations in healthy dogs. Privately-owned male dogs aged 2-5 years; Labrador retrievers (32), Cavalier King Charles spaniels (21), Dachshunds (14) and Boxers (13) underwent physical examination, ECG, echocardiography, routine urine analysis and blood sampling by venipuncture for haematology and biochemistry for verification of health status, as part of the EU-funded Lupa-project.

Prior to examination, owners practiced urine sampling at home to train their dogs. On the examination day, dogs were fasted and water was removed at 7 am. Voided morning urine samples were collected by owners at home. The examination began at 10 am and lasted 2 hours. A second voided urine sample was thereafter collected by owners.

Urine density was measured by a digital refractometer. Urine samples were then frozen and kept at -70°C. Creatinine and cystatin-C were analysed by commercially validated ELISA kits,

whereas ELISA kits for analysis of cortisol, aquaporin-2 and vasopressin were validated for dog urine in our laboratory.

Cortisol, aquaporin-2 and vasopressin were assessed as ratios to creatinine concentrations (cort/crea, AQP-2/crea and VP/crea, respectively). For statistical analysis, all variables were logarithmically transformed, and a mixed model for repeated measurements with level of significance set at  $p < 0.05$  and Bonferroni adjustment was used.

After the clinical examination, significant increases in urine concentrations of creatinine, cystatin-C, and in cort/crea were detected. AQP-2/crea and VP/crea did not change between samples. Urine density showed that dogs had concentrated urine within normal reference ranges both before and after examination, but density was significantly lower after the examination. This indicated a higher urine flow although dogs had not had access to drinking water since at least 7 am.

In conclusion, the increase in creatinine and cystatin-C, concomitant with lowered density indicates higher glomerular filtration rate, probably due to excitement during examination as shown by the elevated cort/crea. AQP-2/crea and VP/crea did not change, which was expected with regard to the high density. Hence, a stress-induced increase in glomerular filtration rate is the most likely cause of the decrease in urinary density. These findings should be taken into consideration when evaluating results of urinary tests in a clinical environment.

#### RE-O-7

**ASSESSMENT OF RENAL DYSFUNCTION USING URINARY MARKERS IN SOUTH AFRICAN CANINE BABESIOSIS.** P. Defauw<sup>1</sup>, J.P.S. Schoeman<sup>2</sup>, P. Smets<sup>1</sup>, A.G. Goddard<sup>2</sup>, E. Meyer<sup>1</sup>, C. Liebenberg<sup>2</sup>, S. Daminet<sup>1</sup>. <sup>1</sup>University of Ghent, MERELBEKE, Belgium, <sup>2</sup>University of Pretoria, ONDERSTEEPOORT, South Africa

Renal damage is a common, yet not well documented, complication in canine babesiosis. Serum urea and creatinine concentrations are insensitive and aspecific markers for early detection of renal dysfunction. Furthermore, measurement of both serum urea and urine specific gravity are influenced by the haemolysis caused by babesiosis. Urinary markers may allow a more sensitive assessment of renal dysfunction and additionally may enhance the specificity as they are able to localize renal dysfunction.

The aim of this study was to assess the degree of renal dysfunction and its localization in dogs with South African babesiosis, using urinary immunoglobulin G (uIgG) and urinary C-reactive protein (uCRP) as markers for glomerular, and urinary retinol-binding protein (uRBP) as marker for tubular dysfunction.

Eighteen dogs diagnosed with uncomplicated babesiosis, without other concurrent diseases, were included. Diagnosis of *Babesia canis rossi* was confirmed by polymerase chain reaction and reverse line blot. Eight dogs of comparable age and body weight were included as healthy controls, based on history, physical examination, complete blood count, biochemistry profile and urinalysis, including bacterial culture. Previously validated commercially available anti-canine or anti-human ELISA kits were used for measurement of uIgG, uCRP respectively uRBP. Results were related to urinary creatinine concentrations (c) and expressed as ratios.

Based on a Mann-Whitney U-test with a significance level of 5%, dogs with babesiosis had a significantly higher urinary protein/c ratio (UPC) (1.6; 0.2-5.5) (median; range) compared to controls (0.1; 0.05-0.4) ( $P < 0.001$ ). Urine specific gravity and serum creatinine concentrations were not significantly different between dogs with babesiosis (1.036; 1.012-1.065 and 68  $\mu\text{mol/l}$ ; 39-215) and controls (1.031; 1.014-1.048 and 92  $\mu\text{mol/l}$ ; 48-103) ( $P = 0.4$  and  $P = 0.2$ , respectively). In marked contrast, dogs with babesiosis had significantly higher uIgG/c (226.71  $\text{mg/g}$ ; 11.32-2296.35) and uCRP/c (0.02  $\text{mg/g}$ ; 0-0.81) ratios compared to controls (1.27  $\text{mg/g}$ ; 0.52-3.23 and below detection limit) ( $P < 0.001$  and  $P = 0.011$ , respectively). Finally, uRBP/c ratios were significantly higher in dogs with babesiosis (10.84  $\text{mg/g}$ ; 0.91-58.23) than in the healthy controls (0.05  $\text{mg/g}$ ; 0-0.16) ( $P < 0.001$ ). All measured urinary markers (uIgG/c, uCRP/c, uRBP/c and UPC) were positively correlated with each other ( $P < 0.001$ ).

These data show the presence of both glomerular and tubular dysfunction in naturally occurring uncomplicated canine babesiosis, caused by *Babesia canis rossi*. Further studies are needed to assess the reversibility of renal dysfunction after treatment and the ability of these more sensitive and specific urinary markers to predict the risk for renal azotemia.

**RE-O-8**  
**DOGS WITH VISCERAL LEISHMANIASIS AND SOME CORRELATIONS BETWEEN RENAL DAMAGE INDICATORS.** P.P. Martinez Padua. São paulo state university, JABOTICABAL/SP, Brazil

Canine visceral leishmaniasis (VL) is a multisystemic disease that can affect the kidneys. We evaluated changes in indices of renal function in dogs with VL. We measured serum creatinine (Cr), urea (Ur), urinary creatinine protein ratio (Up/c), systemic blood pressure (BP), fractional excretion of calcium (FECa), phosphorus (FEP), sodium (FENa) and magnesium (FEMg) in 41 dogs of various breeds with VL. We used the IRIS 2006 classification scheme to classify dogs into 2 groups (G1=IRIS 1-2; G2=IRIS 3-4), and compared them to dogs without renal impairment (control). Groups were compared by a Kruskal-Wallis test followed by Dunnett's test for post-hoc comparisons; significance was set at  $P < 0.05$ . Selected variables were compared by linear correlation. G2 dogs showed increased serum concentrations of Mg and P compared with control dogs ( $P < 0.01$ ). Control dogs had higher FEMg, FEP and FENa than G2 dogs ( $P < 0.05$ ). Control dogs had lower BP, Up/c, Cr and Ur than G2 dogs ( $P < 0.01$ ). We observed a weak positive correlation between Cr and FEMg ( $r=0.37, P=0.01$ ), Ur and FEMg ( $r=0.37, P=0.01$ ), and BP and FEMg ( $r=0.34, P=0.02$ ).

Our results show that dogs with VL and IRIS Stage 3-4 kidney disease (but not IRIS Stage 1-2 disease) display alterations in various indices of renal function including Up/c, Cr, Ur, and FE of various electrolytes. Additionally, FEMg was related to BP and azotemia in these dogs.

**RE-O-9**  
**DISSEMINATION OF THE HUMAN PANDEMIC ST131-O25B CLONE AMONG UROPATHOGENIC ESCHERICHIA COLI ISOLATES FROM COMPANION ANIMALS IN PORTUGAL.** C. Pomba, N. Couto. Faculty of Veterinary Medicine, Technical University of Lisbon, LISBOA, Portugal

A multiresistant CTX-M-15-producing clone of *E. coli* isolates ST131-O25b, has recently disseminated in three continents, including Portugal. The isolates belonging to this clone present significant virulence, and despite being described as multidrug-resistant worldwide, susceptible non-ESBL-producing isolates have been also detected. Studies to assess the real prevalence of this lineage remain scarce. The aim of this study was to evaluate the frequency of the *E. coli* sequence type 131 (ST131) serotype O25-variant b worldwide pandemic clone among companion animal urinary tract infection (UTI) strains in Portugal and its association with fluoroquinolone-resistance and ESBL and AmpC cephalosporinases production. *E. coli* isolates ( $n=119$ , 87 from dogs and 32 from cats) were collected from 2004 until 2009 at the Veterinary Teaching Hospital of the Faculty of Veterinary Medicine and at veterinary private practices mainly in the Lisbon area. The clone was screened by PCR with specific primers for the O25 serogroup and allele 3 of the pabB gene. Susceptibility testing and interpretation was performed using the disc diffusion method according to CLSI guidelines. ESBL and AmpC production was screened by double-disk synergy and antagonism test, respectively, and characterized by PCR using specific primers and full-gene PCR products sequencing. From a total of 119 animal *E. coli* isolates, 44 were O25b-pabB3 positive (37%). Five (11%) CTX-M producer *E. coli* O25b-pabB3 positive strains were isolated from 3 dogs (2 CTX-M-15 and 1 CTX-M-32) and 2 cats respectively (1 CTX-M-15 and 1 CTX-M-32). One *E. coli* O25b-

pabB3 positive strain (2%) isolated from a dog produced the CMY-2 plasmid-encoded cephalosporinase. The prevalence of ESBL and AmpC plasmid-mediated cephalosporinases among all *E. coli* isolates was 10% ( $n=12$ , one strain produced simultaneously a SHV-11  $\beta$ -lactamase and a CMY-2 enzyme). All animal ESBL- and plasmid-encoded cephalosporinase producer strains were also ciprofloxacin-resistant. This study demonstrates that ST131-O25b is a prevalent clone in our area among companion animals with urinary tract infection and the majority of these isolates lack ESBL or plasmidic AmpC genes. Our findings are of critical relevance, as they show companion animals as ST131-O25b pandemic *Escherichia coli* clone reservoirs for human infections. Furthermore, transfer of resistance markers and resistance strains between animals and owners/caretakers is a strong possibility either by infection or direct contact. Our results also demonstrated that multidrug-resistant *E. coli* cephalosporinases and ESBL producing strains prevalence is increasing in uropathogenic *E. coli* and this fact may compromise effective therapeutic options.

**RE-O-10**  
**IDENTIFICATION OF BACTERIAL DNA IN THE URINE OF CATS WITH IDIOPATHIC CYSTITIS.** R. Dorsch, I. Blanke, K. Hartmann, K. Weber. Clinic for Small Animal Internal Medicine, MÜNCHEN, Germany

The aetiology of Feline idiopathic cystitis (FIC) is still unknown. The disease is currently considered multifactorial involving local bladder abnormalities, the nervous and endocrine system as well as environmental factors. Bacterial uropathogens have not been identified using conventional methods but recently a protein of potentially bacterial origin was identified in the urine of cats with FIC.

Therefore, the aim of this study was to apply a highly sensitive PCR to urine samples of cats with FIC to detect a broad spectrum of different bacterial species that are difficult to grow in culture. Urine samples of 21 cats with FIC and 14 healthy controls were included. Urine sediment was used for DNA extraction. Amplification of a highly conserved region of bacterial intergenic-transcribed-spacer sequence was performed. For further analyses, PCR products were cloned and sequenced. Nucleotide sequences were compared with each other and with a nucleotide data bank (NCBI/BLAST).

PCR was positive in 11/21 samples of cats with FIC and in 2/14 samples of control cats ( $p < 0.05$ ). Sequencing of clones was possible in 11/14 samples (11 FIC, 3 controls). Ten of 11 clones showed 83 - 93 % homology to *Rhodococcus* species. Homology between clones was 83 - 99.8 %.

Results of the study suggest a significantly higher incidence of bacterial organisms identical or closely related to *Rhodococcus* species in the urine of cats with FIC compared to controls. However, the clinical significance of the presence of these bacteria is still uncertain.

**RE-O-11**  
**INTRAVESICAL LIDOCAINE TREATMENT OF CATS WITH OBSTRUCTIVE LOWER URINARY TRACT DISEASE (LUTD).** L. Zezza, C.E. Reusch, B. Gerber. Vetsuisse Faculty, University of Zurich, Switzerland, ZUERICH, Switzerland

Re-obstruction of the urethra in cats after obstructive lower urinary tract disease (LUTD) is common and often occurs shortly after the first episode. Bladder pain might lead to urethral spasms and play a role in the re-obstruction. In women with interstitial cystitis intravesical treatment with lidocaine lead to a sustained short and long time amelioration of symptoms. We hypothesized that cats might also benefit from lidocaine treatment. The aim of this study was to assess the rate of re-obstruction in male cats with idiopathic obstructive LUTD treated with intravesical lidocaine and compare this with placebo treated cats and cats with no intravesical treatment.

Idiopathic LUTD was diagnosed by exclusion of other causes of LUTD. The owners of the cats were asked to rate their cat's clinical signs on a visual analogue scale. Standard treatment consisted of unblocking the cats and leaving an indwelling urinary catheter for 3 days. Furthermore the cats received intravenous fluid and pain medication. Antibiotics were usually given after removal of the catheter. In addition to that in the treatment and the placebo group lidocaine (2mg/kg or 4mg/kg) or an equivalent amount of 0.9% saline respectively, together with a sodium bicarbonate solution were instilled through the catheter into the previously emptied bladder. This treatment was repeated once a day for up to 3 days. In the standard group standard treatment was performed without intravesical medication. The number of cats which re-obstructed within one month was assessed.

Seventeen male cats were included in the study. Nine cats were in the treatment group, 4 cats in the placebo group and 4 cats in the standard group. There was no significant difference in age or body weight between groups. The clinical signs score prior to treatment was not significantly different between groups. In 3 cats of the treatment group and in one cat of the placebo group the duration of the treatment was less than 3 days because the cats were aggressive or removed the catheter early.

In the treatment group 4/9 cats (44%) re-obstructed, while in each the placebo and the standard group 3/4 (75%) re-obstructed. Re-obstruction occurred within 1 day in all but one cat which obstructed 2 days after removal of the urinary catheter.

The results in this small number of cats indicate that intravesical lidocaine treatment might reduce the rate of re-obstruction in cats with obstructive LUTD. The intravesical treatment did not negatively influence the outcome.

**RE-O-12**  
**LARGER DAILY URINARY VOLUMES IN HEALTHY CATS INDUCE MORE FREQUENT MICTURITIONS OF SMALLER VOLUMES.** I.vanHoek, A. Feugier, L.LeVerger, C. Venet, V. Biourge. Royal Canin, AIMARGUES, France

In humans, increased micturition volume and frequency parallel a decreased urinary specific gravity (USG). In dogs, micturition volumes and frequencies are reduced in small breed dogs, which might explain in part the higher prevalence of uroliths in these dogs. To our knowledge, these relationships have not been investigated in cats. The objective of this study was to determine the relationships between daily urinary volume (V, mL/d), urinary volume per kg bodyweight (Vbw, mL/kg/d), micturition frequency (F), micturition volume (Vmi, mL) and USG in healthy cats.

The database consisted of 3580 separate 5 day-urinary volumes from 30 healthy cats (age 1-8 years) fed different dry expanded and wet diets over 2 weeks during a period of 2 years. Data were grouped according to F (1, 2 or >3 micturitions/day), and analyzed using non-parametric Kruskal-Wallis test for global effects, Mann-Whitney test for between-group effects and Kendall-tau test for concordance between USG and Vmi.  $P < 0.05$  was considered significant. Results are expressed as mean $\pm$ SD.

F was significantly higher ( $P < 0.001$ ) with both larger V (72.9 $\pm$ 40.6 mL/day) and Vbw (14.6 $\pm$ 7.7 mL/kg/d). Vmi (55.5 $\pm$ 31.4 mL) and USG (1.060 $\pm$ 0.02) decreased significantly with higher F ( $P < 0.001$ ). USG and Vmi were significantly ( $P < 0.001$ ) but weakly correlated ( $r^2 < 0.15$ ).

This study showed that in cats larger daily urinary volumes increase micturition frequency. This is of interest for the management of urolithiasis as crystals are less likely to aggregate with a shorter bladder residence time.