



Research Communications of the 24th ECVIM-CA Congress

Research Communications of the 24th ECVIM-CA Congress
Rheingoldhalle in Mainz, Germany, 4th to 6th September 2014

LIST OF ORAL RESEARCH COMMUNICATIONS

ESVIM – European Society of Veterinary Internal Medicine

Thursday 4 September

10.00-10.15	ESVIM-O-1	Canonne-Guibert	DETECTION OF ANGIOSTRONGYLUS VASORUM IN THE BRONCHOALVEOLAR LAVAGE FLUID OR FAECES OF COUGHING AND HEALTHY DOGS IN BELGIUM
12.20-12.35	ESVIM-O-2	Mason	LIGNEOUS MEMBRANITIS IN A FAMILY OF SCOTTISH TERRIERS
12.35-12.50	ESVIM-O-3	Willems	SCREENING OF APPARENTLY HEALTHY ELDERLY DOGS
14.25-14.40	ESVIM-O-4	Hansson-Hamlin	ANTINUCLEAR ANTIBODY SPECIFICITY IN DOGS WITH IMMUNE-MEDIATED RHEUMATIC DISEASE

ISCAID - International Society for Companion Animal Infectious Diseases

Thursday 4 September

09.00-09.15	ISCAID-O-1	Spiri	CLINICAL AND GENETIC CHARACTERIZATION AND VIRUS NEUTRALIZATION PATTERNS OF FELINE CALICIVIRUS ISOLATES FROM FOUR VIRULENT-SYSTEMIC DISEASE OUTBREAKS IN CATS IN SWITZERLAND AND LIECHTENSTEIN
09.15-09.30	ISCAID-O-2	Willi	CASE CONTROL STUDY ON FELINE CALICIVIRUS TO INVESTIGATE RISK AND PROTECTIVE FACTORS FOR INFECTION
09.30-09.45	ISCAID-O-3	Friedl	EFFICACY OF PASSIVELY TRANSFERRED ANTIBODIES IN CATS WITH ACUTE VIRAL UPPER RESPIRATORY TRACT INFECTION
09.45-10.00	ISCAID-O-4	Schulz	SAMPLING SITES FOR DETECTION OF FELINE HERPESVIRUS-1, FELINE CALICIVIRUS, AND CHLAMYDOPHILA FELIS IN CATS WITH FELINE UPPER RESPIRATORY TRACT DISEASE
10.15-10.30	ISCAID-O-5	Schuller	IMMUNOHISTOCHEMICAL DETECTION OF IGG AND IGM IN LUNG TISSUE OF DOGS WITH LEPTOSPIRAL PULMONARY HAEMORRHAGE SYNDROME (LPHS)
11.20-11.35	ISCAID-O-6	Knoepfler	CLINICAL, LABORATORY, RADIOLOGICAL FEATURES AND OUTCOME OF 99 DOGS WITH LEPTOSPIROSIS (2006-2013)
11.35-11.50	ISCAID-O-7	Buono	CARDIAC AND EXOCRINE PANCREATIC INVOLVEMENT IN DOGS WITH LEPTOSPIROSIS

11.50-12.05	ISCAID-O-8	Mylonakis	MOLECULAR IDENTIFICATION OF BARTONELLA IN DOGS WITH LEISHMANIOSIS (LEISHMANIA INFANTUM) WITH OR WITHOUT ARTHRITIS
12.05-12.20	ISCAID-O-9	Grellet	RISK FACTORS OF GIARDIA INFECTION AND PATHOGENICITY IN WEANING PUPPIES

ESVCP – European Society of Veterinary Clinical Pathology

Thursday 4 September

14.40-14.55	ESVCP-O-1	Giger	FELINE ACUTE INTERMITTENT PORPHYRIA IN NEW BRUNSWICK, CANADA: CLINICAL TO MOLECULAR GENETIC CHARACTERIZATION
15.10-15.25	ESVCP-O-2	Zoia	HAEMOSTATIC FINDINGS OF PLEURAL FLUID: A CROSS-SECTIONAL STUDY IN 33 DOGS
15.25-15.40	ESVCP-O-3	Zoia	ASSOCIATION BETWEEN PLEURAL EFFUSIONS AND PRIMARY HYPERFIBRINOGENO-LYSIS: A CASE CONTROL STUDY IN 99 DOGS
15.10-15.55	ESVCP-O-4	Gommeren	INFLAMMATORY CYTOKINES AND C-REACTIVE PROTEIN IN CANINE SYSTEMIC INFLAMMATORY RESPONSE SYNDROME
15.55-16.10	ESVCP-O-5	Heilmann	BIOLOGICAL VARIATION OF CANINE CALPROTECTIN CONCENTRATIONS IN SERUM
16.10-16.25	ESVCP-O-6	Koutinas	CORRELATION OF ACUTE PHASE PROTEINS WITH CLINICAL AND LABORATORY PARAMETERS, AND CLINICAL STAGING IN 80 DOGS WITH LEISHMANIASIS CAUSED BY L. INFANTUM/CHAGASI
16.25-16.40	ESVCP-O-7	Dunning	DEVELOPMENT OF A REMOTE PLATELET P-SELECTIN TEST FOR DELAYED MEASUREMENT OF PLATELET FUNCTION IN DOGS AND CATS

ESVCN – European Society of Veterinary Clinical Nutrition

Thursday 4 September

14.25-14.40	ESVCN-O-1	German	THE KINETICS OF WEIGHT LOSS IN OBESE CLIENT-OWNED DOGS
14.40-14.55	ESVCN-O-2	Söder	METABOLIC AND HORMONAL RESPONSE TO A FEED-CHALLENGE TEST IN LEAN AND OVERWEIGHT DOGS
14.55-15.10	ESVCN-O-3	Christmann	EFFECTIVENESS OF A NEW DIETETIC WEIGHT MANAGEMENT FOOD TO ACHIEVE WEIGHT LOSS IN CLIENT-OWNED OBESE CATS

SCH – Society of Comparative Hepatology

Thursday 4 September

15.10-15.25	SCH-O-1	Dolera	MAGNETIC RESONANCE SPECTROSCOPY (MRS) IN CANINE HEPATIC ENCEPHALOPATHY DIAGNOSIS AND MONITORING
15.25-15.40	SCH-O-2	Lecoindre	DIAGNOSTIC COMPARISON OF NEEDLE AND WEDGE LAPAROSCOPIC BIOPSIES IN DOGS: 22 CASES

15.40-15.55 SCH-O-3 Strommer
A PILOT STUDY TO ASSESS THE FEASIBILITY OF
TRANSCUTANEOUS INDOCYANINE GREEN CLEARANCE AS
LIVER FUNCTION TEST IN CATS AND DOGS

15.55-16.10 SCH-O-4 Rahmani
EVALUATION OF PREPRANDIAL AND POSTPRANDIAL
GALLBLADDER VOLUME USING THREE-DIMENSIONAL
ULTRASONOGRAPHY IN HEALTHY DOGS

VBPS – Veterinary Blood Pressure Society

Thursday 4 September

11.20-11.35 VBPS-O-1 Manczur
COMPARISON OF DIRECT AND INDIRECT BLOOD PRESSURE
MEASUREMENTS IN CONSCIOUS BEAGLES

11.35-11.50 VBPS-O-2 Crosara
SYSTOLIC ARTERIAL PRESSURE MEASURED SIMULTANEOUSLY
BY DOPPLER TECHNIQUE, USING FORELIMB AND HINDLIMB,
IN DOGS

11.50-12.05 VBPS-O-3 Elliott
EFFICACY AND CLINICAL SAFETY OF A NEW PALATABLE
FORMULATION OF AMLODIPINE IN THE TREATMENT OF
HYPERTENSIVE CATS

12.05-12.20 VBPS-O-4 Elliott
PHARMACODYNAMIC AND PHARMACOKINETIC MODELLING
OF AMLODIPINE IN FELINE HYPERTENSIVE PATIENTS

ESVONC – European Society of Veterinary Oncology

Thursday 4 September

14.25-14.40 ESVONC-O-1 Hergt
APPLICABILITY OF THE 'KIUPEL' 2-TIER GRADING SYSTEM ON
CYTOLOGY SPECIMENS IN CANINE CUTANEOUS MAST
CELL TUMOURS

14.40-14.55 ESVONC-O-2 Keller
EVALUATION OF THE JAK2-STAT5 PATHWAY AS A THERAPEUTIC
TARGET IN CANINE MASTOCYTOMAS

14.55-15.10 ESVONC-O-3 Krupa
RETROSPECTIVE ANALYSIS OF MASITINIB-BASED TREATMENT
OF SUBCUTANEOUS MAST CELL TUMOURS IN 25 CHEMO NAÏVE
DOGS

15.10-15.25 ESVONC-O-4 Sayag
OUTCOME OF CANINE DIFFUSE LARGE B-CELL LYMPHOMA
DURING CHEMOTHERAPY WITH L-ASPARAGINASE,
CYCLOPHOSPHAMIDE, VINCRISTINE AND PREDNISOLONE
(LCOP): 30 CASES (2003 – 2013)

15.25-15.40 ESVONC-O-5 Mason
EPIRUBICIN RESCUE CHEMOTHERAPY IN CANINE HISTIOCYTIC
SARCOMA

15.40-15.55 ESVONC-O-6 Queiroga
STUDY OF THE MECHANISMS BEHIND COX-2 EXPRESSION IN
CANINE MELANOCYTIC TUMOURS

15.55-16.10 ESVONC-O-7 Larsen
NEUTER STATUS AND RISK OF CANCER IN A DANISH DOG
POPULATION

16.10-16.25 ESVONC-O-8 Vilhena
SERUM ACUTE PHASE PROTEINS IN FELINE MALIGNANT
MAMMARY TUMOURS

16.25-16.40 ESVONC-O-9 Martin
ESTABLISHMENT AND CHARACTERISATION OF A NOVEL
CANINE HISTIOCYTIC SARCOMA CELL LINE

ESVC – European Society of Veterinary Cardiology**Friday 5 September**

14.25-14.40	ESVC-O-1	Rishniw	LEFT ATRIAL FUNCTION DIFFERS BETWEEN DOGS WITH DIFFERENT SEVERITIES OF MYXOMATOUS MITRAL VALVE DISEASE
14.40-14.55	ESVC-O-2	Menciotti	QUANTITATIVE EVALUATION OF CANINE MITRAL VALVE IN DOGS USING THREE-DIMENSIONAL ECHOCARDIOGRAPHY
14.55-15.10	ESVC-O-3	Tidholm	REAL-TIME 3- AND 2-DIMENSIONAL ECHOCARDIOGRAPHIC ASSESSMENT OF EFFECTIVE REGURGITANT ORIFICE AREA IN DOGS WITH MYXOMATOUS MITRAL VALVE DISEASE
15.10-15.25	ESVC-O-4	Lu	MICRORNAS EXPRESSION IN CANINE MYXOMATOUS MITRAL VALVE DISEASE
15.25-15.40	ESVC-O-5	Eriksson-Palojarvi	INCREASED LEFT HEART SIZE PREDICTS RISK OF CONGESTIVE HEART FAILURE IN CAVALIER KING CHARLES SPANIELS WITH MITRAL REGURGITATION CAUSED BY MYXOMATOUS VALVE DISEASE
15.40-15.55	ESVC-O-6	Koutinas	LONGITUDINAL STUDY OF SYSTOLIC ARTERIAL BLOOD PRESSURE IN DOGS WITH VARIOUS STAGES OF MITRAL VALVE DISEASE.
16.30-16.45	ESVC-O-7	Lee	CAN THE CALCIUM UPTAKE GENES EXPRESSED IN BLOOD REFLECT THE MYOCARDIAL DYSFUNCTION IN CHRONIC HEART FAILURE?
16.45-17.00	ESVC-O-8	Modler	INCREASED NORMALIZED PULMONARY TRANSIT TIMES AND PULMONARY BLOOD VOLUMES IN CARDIOMYOPATHIC CATS WITH OR WITHOUT CONGESTIVE HEART FAILURE
17.00-17.15	ESVC-O-9	Aroch	DIFFERENCE IN PERIPHERAL AND CENTRAL VENOUS BLOOD GLUCOSE CONCENTRATIONS IN ACUTE ARTERIAL THROMBOEMBOLISM IN CATS AND DOGS

Saturday 6 September

09.00-09.15	ESVC-O-10	Dolera	PERIPHERAL GLYCAEMIA IN DOGS WITH LIMB THROMBOSIS: A PROSPECTIVE STUDY
09.15-09.30	ESVC-O-11	Roels	RIGHT PULMONARY VEIN TO PULMONARY ARTERY RATIO: A NEW ECHOCARDIOGRAPHIC INDEX OF PULMONARY HYPERTENSION IN WEST HIGHLAND WHITE TERRIERS WITH IDIOPATHIC PULMONARY FIBROSIS
09.30-09.45	ESVC-O-12	Abrantes	CHARACTERIZATION AND LONG-TERM OUTCOME OF VENTRICULAR SEPTAL DEFECTS IN DOGS AND CATS
09.45-10.00	ESVC-O-13	Gouni	TISSUE DOPPLER AND STRAIN IMAGING ALTERATIONS IN AGING PTPLA-DEFICIENT LABRADOR RETRIEVERS WITHIN A FRENCH PEDIGREE SEGREGATING CENTRONUCLEAR MYOPATHY
10.00-10.15	ESVC-O-14	Mizuno	MACROSCOPIC EVALUATION OF THE MITRAL VALVE CHORDAE TENDINEAE DURING MITRAL VALVE REPAIR
10.15-10.30	ESVC-O-15	Dickson	TWO-DIMENSIONAL AORTIC VALVE MEASUREMENTS DIFFER THROUGHOUT DIASTOLE

11.20-11.35	ESVC-O-16	Christiansen	CARDIAC MITOCHONDRIAL AND CYTOSKELETAL CHANGES IN FELINE HYPERTROPHIC CARDIOMYOPATHY: AN ULTRASTRUCTURAL STUDY
11.35-11.50	ESVC-O-17	Spalla	CARDIAC MECHANICS IN DOGS WITH PATENT DUCTUS ARTERIOSUS: A SHORT TERM SPECKLE-TRACKING ECHOCARDIOGRAPHY STUDY
11.50-12.05	ESVC-O-18	Yu	PREVALENCE AND PROGNOSIS OF CARDIAC CACHEXIA IN DOGS AFFECTED WITH CHRONIC DEGENERATIVE MITRAL VALVE DISEASE
12.05-12.20	ESVC-O-19	Lu	EPIDEMIOLOGY OF MITRAL VALVE DISEASE IN MALTESE DOGS IN TAIWAN
12.20-12.35	ESVC-O-20	Martinelli	CARDIORENAL SYNDROME IN DOGS WITH CHRONIC VALVULAR HEART DISEASE: A RETROSPECTIVE STUDY
12.35-12.50	ESVC-O-21	Höglund	BREED DIFFERENCES IN CONCENTRATIONS OF NEUROENDOCRINES AND CORTISOL IN HEALTHY DOGS

ESVE – European Society of Veterinary Endocrinology

Friday 5 September

14.25-14.40	ESVE-O-1	Hrovat Vernik	BEHAVIOUR IN DOGS BEFORE AND AFTER TREATMENT OF SPONTANEOUS HYPOTHYROIDISM WITH L-THYROXINE
14.40-14.55	ESVE-O-2	Peterson	OVERT OR SUBCLINICAL IATROGENIC HYPOTHYROIDISM IN CATS: CLINICAL, LABORATORY, AND THYROID SCINTIGRAPHIC FINDINGS IN 35 CASES
14.55-15.10	ESVE-O-3	Peterson	IATROGENIC FELINE HYPOTHYROIDISM: CHALLENGES AND COMPLEXITIES OF THYROID HORMONE REPLACEMENT IN CATS
15.10-15.25	ESVE-O-4	Giger	CONGENITAL HYPOTHYROIDISM WITH GOITER IN CATS DUE TO A TPO MUTATION
15.25-15.40	ESVE-O-5	Riederer	ASSESSMENT OF A GLUCAGON-LIKE PEPTIDE-1 ANALOGUE (EXENATIDE EXTENDED-RELEASE) IN CATS WITH NEWLY DIAGNOSED DIABETES MELLITUS
15.40-15.55	ESVE-O-6	Öhlund	LASER MICRODISSECTION OF PANCREATIC ISLETS ALLOWS FOR QUANTITATIVE REAL-TIME PCR DETECTION OF ISLET-SPECIFIC GENES IN HEALTHY AND DIABETIC CATS
16.30-16.45	ESVE-O-7	Scudder	THE FELINE AIP GENE: THE KEY TO HYPERSOMATOTROPISM TUMORIGENESIS?
16.45-17.00	ESVE-O-8	Kenny	EXPERIENCES OF A NEWLY ESTABLISHED HYPOPHYSECTOMY CLINIC FOR TREATMENT OF FELINE HYPERSOMATOTROPHISM
17.00-17.15	ESVE-O-9	Fracassi	URINARY EXCRETION OF CALCIUM AND PHOSPHATE IN DOGS WITH PITUITARY DEPENDENT HYPERCORTISOLISM: CASE CONTROL STUDY IN 167 DOGS
17.15-17.30	ESVE-O-10	Cosgrove	COMPARISON OF FOUR MONITORING METHODS FOR TRILOSTANE TREATMENT OF CANINE HYPERADRENOCORTICISM
17.30-17.45	ESVE-O-11	Hanson	SURVIVAL ANALYSIS OF DOGS WITH ADRENOCORTICAL INSUFFICIENCY
17.45-18.00	ESVE-O-12	Beatrice	MULTIPLE ENDOCRINE NEOPLASIA IN DOGS AND CATS: A RETROSPECTIVE STUDY OVER 10 YEARS (2004-2014)

ESCG – European Society of Comparative Gastroenterology**Friday 5 September**

14.25-14.40	ESCG-O-1	Bresciani	EFFICACY OF SACCHAROMYCES BOULARDII IN THE TREATMENT OF DOGS WITH CHRONIC ENTEROPATHIES. RANDOMIZED DOUBLE BLIND PLACEBO-CONTROL STUDY
14.40-14.55	ESCG-O-2	Jergens	EFFECT OF VSL#3 PROBIOTIC STRAINS ON THE INTESTINAL MICROBIOTA IN CANINE INFLAMMATORY BOWEL DISEASE
14.55-15.10	ESCG-O-3	Schmitz	TREATMENT WITH THE PROBIOTIC ENTEROCOCCUS FAECIUM IN DOGS WITH INFLAMMATORY BOWEL DISEASE: EFFECT ON MICROBIOME COMPOSITION
15.10-15.25	ESCG-O-4	Pomba	RISK FACTORS FOR FAECAL COLONIZATION WITH ESCHERICHIA COLI PRODUCING EXTENDED-SPECTRUM AND PLASMID-MEDIATED AMPC BETA-LACTAMASES IN DOGS WITHOUT ANTIMICROBIAL PRESSURE
15.25-15.40	ESCG-O-5	Allenspach	LONG-TERM RELAPSE RATE, COMPLIANCE AND CLINICAL SEVERITY IN DOGS DIAGNOSED WITH CHRONIC ENTEROPATHIES (29 CASES)
15.40-15.55	ESCG-O-6	Tappin	THE USE OF MULTIFACTORIAL ANALYSIS FOR THE DETECTION OF PANCREATITIS IN DOGS
16.30-16.45	ESCG-O-7	Dunning	THE USE OF FAECAL LACTOFERRIN AS A MARKER OF INTESTINAL INFLAMMATION IN DOGS WITH CHRONIC ENTEROPATHY
16.45-17.00	ESCG-O-8	Grellet	COMPARISON OF FECAL S100A12 AND FECAL CALPROTECTIN CONCENTRATIONS AS INDICATORS OF DISEASE ACTIVITY IN DOGS WITH CHRONIC DIARRHEA
17.00-17.15	ESCG-O-9	Williams	ORAL COBALAMIN SUPPLEMENTATION EFFECTIVELY RAISES SERUM COBALAMIN CONCENTRATION IN GERIATRIC CATS WITH IDIOPATHIC MALABSORPTION, BUT CONCENTRATIONS DECREASE RAPIDLY FOLLOWING CESSATION OF SUPPLEMENTATION

ESVNU – European Society of Veterinary Nephrology and Urology**Saturday 6 September**

09.00-09.15	ESVNU-O-1	Ghys	BIOLOGICAL VALIDATION OF FELINE CYSTATIN C: THE EFFECT OF BREED, AGE AND GENDER AND ESTABLISHMENT OF A REFERENCE INTERVAL
09.15-09.30	ESVNU-O-2	Steinbach	A PILOT STUDY TO ASSESS THE FEASIBILITY OF TRANSCUTANEOUS GLOMERULAR FILTRATION RATE MEASUREMENT USING FLUORESCENCE-MARKED SINISTRIN IN DOGS AND CATS
09.45-10.00	ESVNU-O-4	Williams	INITIAL EVALUATION OF AUTOMATED URINARY ALBUMIN: CREATININE RATIO (UAC) AND URINARY CYSTATIN C: CREATININE RATIO (UCYSC) AS SCREENING TESTS FOR THE DETECTION OF AZOTAEMIC CHRONIC KIDNEY DISEASE IN CATS
10.00-10.15	ESVNU-O-5	Sewell	CYSTINURIA IN KROMFOHRLÄNDER DOGS
10.15-10.30	ESVNU-O-6	Baril	URETERAL CALCULI IN CATS: RETROSPECTIVE ANALYSIS OF SIGNALMENT, CLINICAL DATA, MEDICAL MANAGEMENT AND SHORT-TERM OUTCOME IN 83 CASES (2005 – 2013)

11.20-11.35	ESVNU-O-7	Taffin	SYSTOLIC BLOOD PRESSURE AND RENAL DISEASE IN CLINICALLY ILL CATS NATURALLY INFECTED WITH FELINE IMMUNODEFICIENCY VIRUS
11.35-11.50	ESVNU-O-8	Dorsch	EVALUATION OF FIRST-LINE ANTIMICROBIAL AGENTS FOR URINARY TRACT INFECTIONS IN CATS WITH AND WITHOUT DOCUMENTED COMORBIDITIES
11.50-12.05	ESVNU-O-9	Marques	EMERGENCE OF MULTIDRUG-RESISTANT BACTERIA CAUSING URINARY TRACT INFECTIONS IN COMPANION ANIMALS: A 14-YEAR RETROSPECTIVE STUDY IN PORTUGAL
12.05-12.20	ESVNU-O-10	Pelander	INCIDENCE- AND MORTALITY RATES OF KIDNEY DISEASE IN A LARGE POPULATION OF INSURED SWEDISH DOGS

LIST OF POSTER RESEARCH COMMUNICATIONS

ESVIM – European Society of Veterinary Internal Medicine

ESVIM-P-1	Lilja-Maula	ACTIVIN B, BUT NOT ACTIVIN A, IS UPREGULATED IN LUNGS OF WEST HIGHLAND WHITE TERRIERS WITH CANINE IDIOPATHIC PULMONARY FIBROSIS
ESVIM-P-2	Bremer	MORBIDITY IN THE NOVA SCOTIA DUCK TOLLING RETRIEVER WITH A FOCUS ON IMMUNE-MEDIATED DISEASE
ESVIM-P-3	Viitanen	VIRAL CO-INFECTIONS IN DOGS WITH BACTERIAL PNEUMONIA
ESVIM-P-4	Lin	CAUSES OF CANINE ANEMIA IN TAIWAN: A FIVE-YEAR RETROSPECTIVE SURVEY
ESVIM-P-5	Canonne-Guibert	DETECTION OF MYCOPLASMA CANIS AND MYCOPLASMA CYNOS BY SPECIFIC QUANTITATIVE POLYMERASE CHAIN REACTION ASSAYS IN THE BRONCHOALVEOLAR LAVAGE FLUID IN DOGS INFECTED WITH BORDETELLA BRONCHISEPTICA
ESVIM-P-6	Canonne-Guibert	DETECTION OF BORDETELLA BRONCHISEPTICA, MYCOPLASMA CANIS AND MYCOPLASMA CYNOS BY SPECIFIC QUANTITATIVE POLYMERASE CHAIN REACTION ASSAYS IN THE BRONCHOALVEOLAR LAVAGE FLUID OF DOGS WITH EOSINOPHILIC BRONCHOPNEUMOPATHY

ISCAID - International Society for Companion Animal Infectious Diseases

ISCAID-P-1	Monteiro Carvalho Mori Cunha	MORPHOLOGICAL RENAL LESIONS IN DOGS SEROPOSITIVE FOR CANINE VISCERAL LEISHMANIASIS
ISCAID-P-2	Chirek	PREVALENCE OF ANAPLASMA PHAGOCYTOPHILUM IN HEALTHY CANINE BLOOD DONORS
ISCAID-P-3	Fernandez Aragonés	STUDY OF RISK FACTORS IN CATS WITH URTD, CONJUNCTIVITIS AND CHRONIC GINGIVOSTOMATITIS IN SPAIN
ISCAID-P-4	Pomba	HIGH FREQUENCY OF COLONIZATION BY METHICILLIN-RESISTANT STAPHYLOCOCCI IN HEALTHY HUMANS IN DAILY CONTACT WITH ANIMALS IN PORTUGAL
ISCAID-P-5	Weingart	INFECTION WITH HEMOPLASMA SPECIES IN 17 CATS WITH ANEMIA
ISCAID-P-6	Liu	CLINICAL APPLICATION OF PORCINE ANTI CDV ANTIBODY SUBUNIT F(AB') ₂ IN CANINE DISTEMPER DOGS

ESVCP – European Society of Veterinary Clinical Pathology

ESVCP-P-1	Stiller	VALIDATION OF A NEW SANDWICH-ELISA TO MEASURE FELINE HAPTOGLOBIN
ESVCP-P-2	Roels	VASCULAR ENDOTHELIAL GROWTH FACTOR: A BLOOD BIOMARKER IN CANINE IDIOPATHIC PULMONARY FIBROSIS?
ESVCP-P-3	Rose	COMPARISON OF TOTAL PROTEIN CONCENTRATIONS IN PLEURAL AND ABDOMINAL FLUID IN DOGS ANALYSED WITH A REFRACTOMETER AND USING PENTRA 400 [®]
ESVCP-P-4	Giger	WELSH SPRINGER SPANIELS WITH COAGULATION FACTOR VII DEFICIENCY FROM FINLAND
ESVCP-P-5	Garden	BREED-SPECIFIC HAEMATOLOGICAL AND SERUM BIOCHEMICAL PHENOTYPES IN THE DOMESTIC DOG

ESVCN – European Society of Veterinary Clinical Nutrition

ESVCN-P-1	Christmann	EFFECTIVENESS OF A NEW DIETETIC WEIGHT MANAGEMENT FOOD TO ACHIEVE WEIGHT LOSS IN CLIENT-OWNED OBESE DOGS
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SCH – Society of Comparative Hepatology**VBPS – Veterinary Blood Pressure Society****ESVONC – European Society of Veterinary Oncology**

ESVONC-P-1	Borschensky	CYSTIC PANCREATIC NEOPLASIA IN CATS
ESVONC-P-2	Queiroga	ROLE OF CYCLOOXYGENASE-2 IN PROGNOSIS OF CANINE MAST CELL TUMOURS
ESVONC-P-3	Kleiter	RADIOTHERAPY IN THE TREATMENT OF 32 CANINE SUBCUTANEOUS SOFT TISSUE SARCOMAS
ESVONC-P-4	Mclean	ONCEPT [®] VACCINATION IN 34 DOGS - THE SOUTH AFRICAN EXPERIENCE

ESVC – European Society of Veterinary Cardiology

ESVC-P-1	Szatmári	PUPPIES WITH AN INNOCENT CARDIAC MURMUR HAVE LOWER HEMATOCRIT
ESVC-P-2	Kvart	CARDIAC MURMURS IN NORMAL SIBERIAN HUSKY DOGS
ESVC-P-3	Swift	INVESTIGATION OF NT-PROBNP POINT-OF-CARE ELISA ASSAY IN CLINICALLY NORMAL FELINE PATIENTS
ESVC-P-4	Caivano	LEFT ATRIAL FUNCTION DETERMINED BY 2-DIMENSIONAL SPECKLE TRACKING ECHOCARDIOGRAPHY IDENTIFIES DOGS WITH CONGESTIVE HEART FAILURE SECONDARY TO MITRAL VALVE DISEASE
ESVC-P-5	Lehtinen	BREED-SPECIFIC REFERENCE RANGES FOR ECHOCARDIOGRAPHY IN SALUKIS
ESVC-P-6	Mocanu	ANALYSIS OF THE P WAVE DURATION IN ATRIAL REMODELING VS. AUTONOMIC NERVOUS SYSTEM DYSFUNCTION IN DOGS
ESVC-P-7	Quintavalla	TRANSTHORACIC ECHOCARDIOGRAPHY IN CLINICALLY HEALTHY ADULT NEWFOUNDLAND DOGS: REFERENCE VALUES FOR THE BREED

- ESVC-P-8 Roels CORRELATION BETWEEN THORACIC CT-SCAN ANGIOGRAPHY FINDINGS AND ECHOCARDIOGRAPHIC RIGHT PULMONARY VEIN TO PULMONARY ARTERY RATIO IN WEST HIGHLAND WHITE TERRIERS WITH IDIOPATHIC PULMONARY FIBROSIS
- ESVC-P-9 Wiley EFFECT OF A BASIC TRAINING PROGRAM ON EMERGENCY CLINICIAN ACCURACY TO SEMI-QUANTITATIVELY ASSESS THORACIC AND CARDIAC STRUCTURES USING FOCUSED CARDIAC ULTRASOUND (FOCUS)

ESVE – European Society of Veterinary Endocrinology

- ESVE-P-1 Horspool PREVALENCE OF HYPERTHYROIDISM IN PORTUGUESE CATS
- ESVE-P-2 Rito Brandao ESTABLISHMENT OF A PROTOCOL FOR THE ISOLATION OF PURE PANCREATIC ISLETS OF LANGERHANS IN CATS
- ESVE-P-3 Hambrook NESIDIOBLASTOSIS IN A CAT
- ESVE-P-4 Vilhena FELINE HYPERTHYROIDISM IN PORTUGAL
- ESVE-P-5 Brito-Casillas EVIDENCE OF AUTOIMMUNITY IN A POPULATION OF DIABETIC DOGS FROM THE CANARY ISLANDS
- ESVE-P-6 Hoffrogge COMPARISON OF THE PLASMA CORTISOL-DEHYDROEPIANDROSTERONE-RATIO IN HEALTHY DOGS AND DOGS WITH HYPERADRENOCORTICISM
- ESVE-P-7 Wehner PREVALENCE OF FELINE HYPERTHYROIDISM AND INTRINSIC RISK FACTORS IN A CLINIC POPULATION IN SOUTHERN GERMANY
- ESVE-P-8 Rosca ENHANCED DIAGNOSIS SYSTEM FOR FELINE DIABETES MELLITUS

ESCG – European Society of Comparative Gastroenterology

- ESCG-P-1 Steiner ANALYTICAL COMPARISON OF NEWLY INTRODUCED COMMERCIAL ASSAYS FOR THE MEASUREMENT OF CANINE AND FELINE PANCREATIC LIPASE IMMUNOREACTIVITY (CPLI AND FPLI) TO ESTABLISHED ASSAYS
- ESCG-P-2 Grellet EFFECT OF AGE, BREED SIZE AND ENTEROPATHOGEN INFECTION ON FECAL IMMUNOGLOBULIN A CONCENTRATIONS IN WEANING PUPPIES
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ESVNU-P-4	Harada	THE EVALUATION OF RENAL FUNCTION AFTER MITRAL VALVE REPAIR IN DOGS
ESVNU-P-5	Williams	VALIDATION OF A HIGH PERFORMANCE CAPILLARY ELECTROPHORESIS METHOD FOR MEASUREMENT OF GLOMERULAR FILTRATION RATE BY SERUM IOHEXOL CLEARANCE IN DOGS

ORALS

ESCG-O-1

EFFICACY OF SACCHAROMYCES BOULARDII IN THE TREATMENT OF DOGS WITH CHRONIC ENTEROPATHIES. RANDOMIZED DOUBLE BLIND PLACEBO-CONTROL STUDY. F. Bresciani, S. d'Angelo, F. Fracassi, R. Galuppi, A. Diana, N. Linta, G. Bettini, M. Morini, M. Pietra. University of Bologna, Ozzano dell'emilia (bo), Italy

Saccharomyces boulardii (Sb) is a non-pathogenic yeast used in the prevention and treatment of gastrointestinal disorders in human beings and horses. The aim of this study was to evaluate the effect of Sb in healthy dogs and dogs with chronic enteropathies (CE). Sb was formulated in 10×10^9 CFU capsules. Its concentration and viability within the capsules was controlled by yeast culture in subsequent steps until expiration date. Four healthy dogs (HD) and 18 dogs with CE (10 inflammatory bowel disease - IBD, 8 protein losing enteropathy - PLE) were included. In HD Sb was administered for 10 days (1×10^9 CFU/kg BID); daily clinical evaluation was performed to assess possible adverse effects and quantitative stool cultures for yeasts were performed before, during and after the administration. In dogs with CE a randomized double blind placebo-control study was performed, administering Sb (1×10^9 CFU/kg BID) or placebo (PI). Sb or PI administration was added to standard therapeutic protocols (diet, antibiotics and immunosuppressive drugs), to evaluate its efficacy for the treatment of IBD and PLE. Complete blood work, abdominal ultrasonography, gastro-duodenal and colon endoscopy and histopathological evaluation of intestinal samples were performed at diagnosis and after 60 days of treatment. Validated score system for the clinical signs (CECCAI), ultrasonography, endoscopy and histopathology were applied. Significance was set for $P < 0.05$. Results in HD showed the absence of Sb in the faeces before treatment, its presence after one day, its steady state (10×10^7 CFU/g) after 5 days and its complete elimination 4 days after withdrawal of treatment. No adverse effects were reported. In CE dogs the clinical score improved significantly in dogs receiving Sb compared to dogs receiving PI ($P = 0.009$). In PLE dogs the albumin concentration increased significantly ($P = 0.034$) in the group receiving Sb with respect to PI. The daily frequency of defecation in the Sb group was significantly lower with respect to PI after 45 ($P = 0.032$) and 60 ($P = 0.004$) days of treatment. No statistical differences were found between dogs receiving Sb and PI after treatment, based on the endoscopic evaluation of duodenum and colon. No statistical differences were found between the two groups on the duodenal ultrasonographic and histological evaluation after treatment. In conclusion, Sb can be safely used in dogs with CE, in addition to standard treatment, to achieve a better control of clinical signs and a significant increase in albumin concentration compared to the standard therapy alone.

Conflicts of interest: No conflicts of interest reported.

ESCG-O-2

EFFECT OF VSL#3 PROBIOTIC STRAINS ON THE INTESTINAL MICROBIOTA IN CANINE INFLAMMATORY BOWEL DISEASE. R.S. White¹, G. Rossi², T. Atherly³, C. Webb⁴, S. Hill⁵, J.M. Steiner⁶, J.S. Suchodolski⁶, A.E. Jergens¹. ¹Iowa State University, Ames, United States of America, ²University of Camerino, Camerino, Italy, ³USDA-ARS, Ames, United States of America, ⁴Colorado State University, Fort Collins, United States of America, ⁵Veterinary Specialty Hospital, San Diego, United States of America, ⁶GI Laboratory Texas A&M University, College station, United States of America

Canine inflammatory bowel disease (IBD) is an immune-mediated enteropathy likely triggered by environmental and immunoregulatory factors in genetically susceptible dogs. Previous studies suggest a pivotal role for gut bacteria in disease pathogenesis since luminal microbial composition is markedly altered (ie, dysbiosis) at diagnosis. Probiotic bacteria appear to be therapeutically effective in some forms of human IBD. Controlled studies evaluating the efficacy of probiotic therapy for canine IBD have not been previously reported. The aim of the present study was to characterize the mucosa-associated microbiota and determine

the clinical, microbiological, and mucosal homeostatic effects of orally administered VSL#3 probiotics in dogs with IBD. Twenty dogs diagnosed with moderate-to-severe IBD (CIBDAI score > 5) were randomized to receive standard therapy (ie, elimination diet and glucocorticoids) with or without probiotic VSL#3. The mucosal microbiota from endoscopic intestinal biopsies of IBD dogs and controls was evaluated by fluorescence in situ hybridization (FISH) targeting the 16S rRNA genes of total bacteria, group-specific organisms, and individual bacterial species shown to be relevant in human IBD. Epithelial tight junction protein (TJP) expression was studied using immunohistochemistry. Clinical signs and changes in mucosal microbiota and TJP expression were assessed before and after probiotic VSL#3 therapy. IBD dogs showed a reduction in GI signs following 8 weeks of probiotic therapy compared with baseline CIBDAI scores ($P < 0.05$). Adherent and sporadic invasive bacteria (EUB) were observed in the small intestines and colon of healthy dogs. The diseased canine duodenum was nearly bacteria-free. IBD dogs given probiotic VSL #3 had altered spatial redistribution of most bacterial groups in the mucus and adherent compartments of the colon. Subset analysis showed that Lactobacilli were significantly ($P < 0.05$) increased in the lumen and mucus post-VSL #3, while the number of mucus laden Bifidobacteria approached significance ($P = 0.08$). Expression of TJP showed that occludin was significantly lower in control intestines as compared to duodenal and colonic mucosa obtained from IBD dogs that received probiotic ($P = 0.008$ and $P = 0.01$, respectively). In contrast, claudin-2 expression in the colon was significantly higher ($P < 0.002$) in control dogs versus VSL #3 treated IBD dogs. Our data demonstrate that probiotic VSL#3 alters some of the mucosa-associated microbiota in dogs with IBD. These probiotic changes in bacterial composition are associated with up-regulated TJP expression indicative of enhanced epithelial barrier integrity, similar to VSL#3-induced disease protection seen in human IBD.

Conflicts of interest: The probiotics used in the trial were supplied free of charge by the manufacturer.

ESCG-O-3

TREATMENT WITH THE PROBIOTIC ENTEROCOCCUS FAECIUM IN DOGS WITH INFLAMMATORY BOWEL DISEASE: EFFECT ON MICROBIOME COMPOSITION. S. Schmitz¹, J.S. Suchodolski³, B.C. Guard³, J.M. Steiner³, D. Werling⁷, K. Allenspach¹. ¹Royal Veterinary College, Department of Clinical Sciences and Services¹, and Department of Pathology and Infectious Diseases², London, United Kingdom, ³Gastrointestinal Laboratory, Department of Small Animal Clinical Sciences, College Station, Texas, United States of America

Canine Inflammatory bowel disease (IBD) is thought to be partially caused by an aberrant immune response towards the intestinal microbiome. In humans and mice, administration of probiotics can alleviate IBD severity and/or prevent relapse by induction of a more "tolerant" microenvironment. The aim of this study was to investigate the effect of probiotic *Enterococcus faecium* NCIMB 10415 E1707 (EF) on intestinal microbiome composition. Dogs were recruited to receive EF at 1×10^8 cfu in a double-blinded, placebo-controlled manner in addition to an exclusion diet (hydrolysed protein). Seven dogs were included in the probiotics group and 5 dogs in the placebo group. All dogs improved clinically after treatment, however, there was no obvious effect on clinical severity in those that received probiotics. Fresh naturally voided faecal samples were collected from all dogs before and after treatment, snap-frozen in liquid nitrogen and stored at -80°C until further analysis. Genomic DNA was extracted from each faecal sample using the Mobio Power soil DNA isolation Kit (MoBio Laboratories), as recommended by the manufacturer. Next generation sequencing was performed on the Ion-Torrent [TRADEMARK] (Life Technologies) platform based upon the V1-V3 region (*E. coli* position 27-519) of the 16S rRNA gene with the following primers: forward 28F: GAG-TTTGATCNTGGCTCAG and reverse 519R: GTNTTACN CGGCKGCTG. Raw sequence data were screened, trimmed, filtered, and chimera depleted with default settings using the QIIME pipeline version 1.8 and UCHIME software, in which

microbiome composition between treatment groups before and after treatment was compared.

Microbiota composition was not significantly different between probiotic and placebo treatment groups, and did not change significantly before and after treatment. However, there was large individual variability in the microbiome composition. Species richness of faecal samples increased after treatment in both groups, but was only statistically significant in the probiotic treatment group.

In conclusion, probiotic treatment in dogs with IBD leads to a significantly increased richness of the faecal bacterial microbiome. A possible additional effect of the change of diet cannot be excluded. Further studies should investigate microbiomic changes in healthy dogs fed the same diet to assess if similar changes in the faecal microbiome occur due to dietary changes alone.

Conflicts of interest: This study is based on a PhD supported by Probiotics Ltd., Somerset, UK (the manufacturer of the probiotic product *Enterococcus faecium* mentioned in this study).

ESCG-O-4 RISK FACTORS FOR FAECAL COLONIZATION WITH ESCHERICHIA COLI PRODUCING EXTENDED-SPECTRUM AND PLASMID-MEDIATED AMPC BETA-LACTAMASES IN DOGS WITHOUT ANTIMICROBIAL PRESSURE. C. Pomba, A. Belas, A.S.S. Salazar, L. Telo Da Gama, N. Coufo. Faculty of Veterinary Medicine, Lisboa, Portugal

The aim of this study was to assess the prevalence and risk factors for faecal carriage of extended-spectrum beta-lactamases (ESBL) and plasmidic AmpC beta-lactamases (*pAmpC*) *E. coli* producers in healthy dogs.

A 3-month cross-sectional study was conducted at a private hospital in Lisbon, Portugal and 151 rectal swabs were obtained from healthy dogs. The dogs included in the study were healthy with no history of antimicrobial consumption in the previous month. ESBL and *pAmpC* genes were detected by PCR and were sequenced. Potential risk factors for ESBL- and *pAmpC*-producing *E. coli* faecal carriage were obtained through a questionnaire to the owner regarding reason for veterinary visit, hospitalisation and antimicrobial treatment within the last year, habitat (shelter, dog breeders and private owner), cohabitation with other animals, street access, kennel/hotel access, age and gender. Data were analysed by SAS software (version 9.3; SAS Institute Inc., Cary, N.C.) and logistic regression models were used.

Rectal swabs obtained from 151 healthy dogs yielded 131 positive samples for *E. coli*. About 15% of the isolates carried ESBL genes (*bla*_{CTX-M-32} *n* = 8, *bla*_{CTX-M-15} *n* = 5, *bla*_{CTX-M-1} *n* = 3, *bla*_{CTX-M-9-like} *n* = 4) and 20% carried *pAmpC* genes (*bla*_{CMY-2} *n* = 23, *bla*_{CMY-2-like} *n* = 2, *bla*_{DHA-1} *n* = 1). Thirteen dogs carried an *E. coli* isolate with both an ESBL and a *pAmpC* gene.

Dogs previously treated with antimicrobials within the last year were at higher risk of carrying at least one β -lactamase ($P = 0.003$; OR = 7.85; CI 95%: 1.99-30.89) or both β -lactamases ($P = 0.020$; OR = 5.18; CI 95%: 1.29-20.81) than non-treated dogs. Dogs in shelters/breeders tended to show a higher incidence of ESBL- producing *E. coli* ($P = 0.069$; OR = 3.17; CI 95%: 0.91-11.01) or at least one β -lactamase producing *E. coli* ($P = 0.050$; OR = 2.65; CI 95%: 1.00-7.03) than dogs from private owners. Males tended to be less likely to carry at least one β -lactamase (ESBL or *pAmpC*) ($P = 0.060$; OR = 0.42; CI 95%: 0.17-1.04) or a *pAmpC* enzyme ($P = 0.017$; OR = 0.28; CI 95%: 0.10-0.80) than females. This study suggests that dogs may act as reservoirs for resistant bacteria, namely for cephalosporin-resistant *E. coli*. Three potential risk factors associated with the carriage of ESBL- and/or *pAmpC*-producing *E. coli* by dogs were identified, which is important for the implementation of effective control measures and judicious antimicrobial therapy.

Conflicts of interest: Dr Pomba currently receives research funding from the government and national programmes (Fundação para a Ciência e a Tecnologia). In the past, she has occasionally received research support or honoraria for lectures from pharmaceutical companies including Zoetis and Atral Cípan. She is vice-chair of the Antimicrobial Working Party

(AWP) and member of the Antimicrobial Advice ad hoc Expert group (AMEG) of the European Medicines Agency (EMA).

ESCG-O-5 LONG-TERM RELAPSE RATE, COMPLIANCE AND CLINICAL SEVERITY IN DOGS DIAGNOSED WITH CHRONIC ENTEROPATHIES (29 CASES). K. Allenspach, S. Aflalo, F. Procoli. Royal Veterinary College, Department of Veterinary Clinical Sciences and Services, London, United Kingdom

There are few reports in the literature reporting long-term relapse rate, owner compliance and clinical severity of dogs with chronic enteropathies. The goal of this study was to compare clinical activity index (CCECAI), number of relapses and compliance rates 1-3 years after diagnosis.

Food-responsive disease (FRD) was defined as dogs that responded to elimination diet alone within 2 weeks after initiating therapy, whereas antibiotic-responsive disease (ARD) dogs had an unsuccessful dietary trial before and responded to metronidazole within 2 weeks after initiation of therapy, and steroid-responsive disease (SRD) dogs had an unsuccessful dietary and antimicrobial trial before, and required immunosuppressive therapy to control their clinical signs. CCECAI was extracted from the medical record database at 1-3 years after diagnosis. Relapse rate was obtained by requesting the medical records of the referring veterinarians and defined as number of return visits to the referring practice after diagnosis. Compliance data was obtained by telephone questionnaire to the owners.

The FRD group consisted of 21/29 dogs (72%), whereas the ARD and SRD groups consisted of 4 (10%) and 5 dogs (17%), respectively. There was a significant difference in CCECAI at follow-up between FRD and ARD, and FRD and SRD (median CCECAI 1.8 (range 0-5) for FRD, 5 (range 0-8) for ARD, and 4.5 (range 0-8) for SRD, $p = 0.01$). For the FRD dogs, 43% of owners stated that they deviated from the prescription diet on a daily basis, 24% once a week, and 5% once a month, with a median CCECAI at the time of deviation from the diet of 4.8 (range 2-8). Relapse rate was highest for the ARD group, when compared to FRD and SRD (10 for ARD, 1.7 for FRD, and 4.7 for SRD, $p = 0.004$). In the FRD group, 17/21 dogs had been kept on the prescribed diets, and 4 dogs had been changed to supermarket brands. All of the ARD dogs had been given immunosuppressive treatment in addition to antibiotics at the time of follow-up, while 4/5 SRD dogs were still on immunosuppressive treatments, with one dog being in remission with dietary treatment alone.

In conclusion, this pilot study indicates that compliance rate for FRD dogs is the lowest, with owners willing to tolerate the highest severity of clinical signs related to deviation from the prescription diet. ARD dogs had the highest relapse rate in this cohort, indicating poor response to treatment in the long-term.

Conflicts of interest: Dr Allenspach has received research funding from BBSRC, American Kennel Club, Comparative Gastroenterology Society, Probiotics Ltd UK, Laboklin GmbH Germany, and Bioiberica Sp. she has also undertaken paid consultancy work for Bioiberica Spain and Hoffmann-Laroche, Switzerland.

ESCG-O-6 THE USE OF MULTIFACTORIAL ANALYSIS FOR THE DETECTION OF PANCREATITIS IN DOGS. K.J. Slater¹, S. Tappin², R. Foale², I. Alexandrakis¹. ¹Avacta Animal Health, Wetherby, United Kingdom, ²Dick White Referrals, Six mile bottom, United Kingdom

Despite the high prevalence of canine pancreatitis in post-mortem studies and the introduction of new diagnostic tests, it is believed that the disease, particularly in its chronic form, remains under recognised due to the non-specific nature of presenting signs. Histology is considered to be the gold standard for diagnosis of canine pancreatitis, however, most clinicians are reluctant to take pancreatic biopsies due to significant risks to the patient.

Numerous serum markers have been reported to be elevated in canine pancreatitis, although most lack sensitivity or specificity. Consequently, confirmed diagnosis requires results from a range of tests including imaging, serum biochemistry and physical examination. We and others have previously shown in other diseases that performance of individual low specificity markers can be dramatically improved by combining data from multiple markers with clinical information using analytical algorithms. We therefore applied this approach to the detection of pancreatitis in dogs.

The activity of two non-specific biomarkers, amylase and lipase, was determined in 132 serum samples from dogs suspected of having pancreatitis by their veterinarian. Of these samples 42 were positive by virtue of their pancreatic lipase (cPLi) results (cPLi > 200 ug/l). The amylase and lipase data was then used to develop a series of algorithms using mathematical data mining and classification techniques. Additional algorithms were developed using extra parameters including age, sex, vomiting, diarrhoea and abdominal pain in addition to the two enzyme levels. The performance of the algorithms was assessed using 27 separate blinded serum samples taken from dogs which were scored clinically for acute pancreatitis according to the system described by McCord et al (J Vet Intern Med 2012;26:888-896). These cases presented for evaluation with vomiting, diarrhoea, inappetence and abdominal pain and were included if a clinical history, results of routine haematology, biochemistry, cPLi assay and abdominal ultrasound were available.

The results of the multifactorial analysis and cPLi assay results were compared to the clinical scores. Using amylase and lipase data alone, the algorithm gave a sensitivity of 81.8% and specificity of 93.3%, compared to cPLi results for the same samples of 63.6% and 86.7% respectively when both methods were referred to clinical scoring. When the presence of additional clinic data was also included into the algorithm, the sensitivity increased to 90.9% with specificity of 100%.

The data suggests that test performance for canine pancreatitis can be dramatically improved when multiple diagnostic parameters are combined using disease specific algorithms.

Conflicts of interest: The author receives a salary as Editor of the BSAVA journal Companion, and has undertaken unrelated paid consultancies for Bayer and Merial. The author also receives a salary from Avacta Animal Health, and duties involved working directly on this project.

ESCG-O-7

THE USE OF FAECAL LACTOFERRIN AS A MARKER OF INTESTINAL INFLAMMATION IN DOGS WITH CHRONIC ENTEROPATHY. M. Dunning¹, S. Hulme², D. Walker³, H.L. Jenkins⁴, N.H. Bexfield², R. Bettencourt⁵, J.H. Boone⁵. ¹University of Nottingham, Leicestershire, United Kingdom, ²School of Veterinary Medicine and Science, University of Nottingham, United Kingdom, ³Anderson Moores Veterinary Specialists, Winchester, United Kingdom, ⁴School of Veterinary Sciences, University of Bristol, United Kingdom, ⁵TechLab, Inc, Blacksburg, va, United States of America

Canine chronic enteropathy (CCE) can cause significant long-term morbidity. In some cases this is due to intestinal inflammation, resulting from idiopathic inflammatory bowel disease (IBD). Currently, the diagnosis of idiopathic IBD and assessment of disease severity relies on results of subjective clinical indices, laboratory data, diagnostic imaging and intestinal histopathology, whilst ruling out known causes of inflammation. In humans with IBD, a number of faecal biomarkers including lactoferrin, aid with diagnosis and determining disease activity. It may therefore be valuable to develop similar non-invasive objective methods to aid diagnosis and clinical assessment of disease severity in dogs with intestinal inflammation due to idiopathic IBD.

This pilot study aimed to measure faecal lactoferrin concentration (FLC) in dogs with CCE and histologically confirmed intestinal inflammation (HCII) and to compare this with control dogs. In addition, the FLC in dogs with HCII would be compared with the canine inflammatory bowel disease activity index

(CIBDAI) and WSAVA standard histopathological criteria for intestinal inflammation to determine whether there was correlation between these methods when assessing disease severity.

Faecal samples were obtained from dogs with HCII (n = 13) having undergone investigation for CCE (serum biochemistry, complete blood count, full faecal and urinalysis, serum cobalamin, quantitative cPLi, abdominal ultrasound and intestinal biopsies). The control population were dogs presented for reasons unrelated to CCE (n = 7). Analysis was carried out using a faecal lactoferrin ELISA previously validated in dogs (TechLab, USA).

The FLC in dogs with HCII (median 9.79 µg/g - range 0.47 to 150.2) was significantly higher than control dogs (median 0.75 µg/g - range 0.12-4.14) (P < 0.003). A cut-off FLC of 4.5 µg/g correctly identified 10/13 (77%) of dogs with HCII. Using this cut-off, there was no overlap between non-CCE dogs FLC and the HCII group; giving a sensitivity of 76% and specificity of 100%. Neither the presence of neutrophils nor the extent of inflammation on histopathology showed significant correlation with FLC. The CIBDAI showed moderate correlation with FLC in dogs with HCII (r = 0.79, P = 0.05).

The results of this pilot study suggest that FLC is able to discriminate between dogs with CCE due to HCII and dogs without CCE. It is possible that incorporating FLC into a panel of faecal biomarkers will enable non-invasive assessment of HCII and could serve as an adjunct to current measures of disease severity in dogs with idiopathic IBD.

Conflicts of interest: Ryan Bettencourt and James Boone are employees of TechLab, USA.

They provided the ELISA kits free of charge for this work.

There was no other incentive provided and the results have been openly discussed between all parties.

There has been no censorship placed on the results by TechLab and they have been supportive of the work and submission of this abstract.

There are no other conflicts to disclose.

ESCG-O-8

COMPARISON OF FAECAL S100A12 AND FAECAL CALPROTECTIN CONCENTRATIONS AS INDICATORS OF DISEASE ACTIVITY IN DOGS WITH CHRONIC DIARRHEA. A. Grellet¹, R.M. Heilmann², P. Lecointre³, A. Feugier¹, M.J. Day⁴, V. Freiche⁵, J. Hernandez⁶, J.S. Suchodolski², J.M. Steiner⁷. ¹Royal Canin, Aimagues, France, ²Gastrointestinal Laboratory, Texas A&M University, College station, United States of America, ³Cerisioz Veterinary Clinic, Saint-priest, France, ⁴University of Bristol, Bristol, United Kingdom, ⁵Alliance Veterinary Clinic, Bordeaux, France, ⁶CHV Frégis, Arcueil, France

Faecal S100A12 and faecal calprotectin concentrations have been described as biomarkers in dogs with chronic enteropathies [1]. However, to date there has been no direct comparison of these two markers in dogs with chronic diarrhea. The aim of this study was to evaluate the performance of these two markers in this situation.

Thirty one dogs presented for a history of chronic diarrhea were prospectively enrolled. The initial diagnostic workup for all patients included a serum biochemistry profile, fecal parasitology, abdominal ultrasound examination, and gastrointestinal endoscopy with collection of endoscopic biopsies. The severity of clinical signs was evaluated using the CCECAI scoring index and patients were grouped by having a CCECAI of < 12 or ≥ 12. Faecal calprotectin and S100A12 were quantified as previously described [1]. Correlations were evaluated with the Spearman rank correlation test. For both markers a receiver operating characteristics (ROC) curve was used to select cut-off value that allowed the best discrimination between dogs with a CCECAI < 12 and dogs with a CCECAI ≥ 12. Sensitivity and specificity were calculated.

Correlation analyses revealed a significant positive correlation between S100A12 and calprotectin (r = 0.77; p < 0.001). The optimal cut-off value for faecal calprotectin concentration was 49.1 µg/g, which was associated with a sensitivity of 53.3% and a specificity of 86.7% (AUC=0.698; P = 0.049). The optimal cut-off value for faecal S100A12 concentration was 173.7 ng/g, which

was associated with a sensitivity of 93.3% and a specificity of 60% (AUC=0.782; P = 0.001). The sensitivity for fecal S100A12 was higher than that for fecal calprotectin (p = 0.019). No significant difference was observed for the specificity of these two markers (p = 0.215). 20 out of the 30 dogs (67%) had concordant results for S100A12 and calprotectin tests. Among these 20 dogs, 11 presented with a CCECAI <12 and 8 of these 11 dogs had both markers below their cut-off values. Among the 9 dogs with a CCECAI ≥12, 8 dogs had both markers above their cut-off values. 66% of dogs (19/29) presented histologic signs of inflammation. Sensitivities for fecal calprotectin and S100A12 concentrations for histopathological intestinal inflammation were 42% and 68%, respectively, and specificities were 80% and 30%, respectively.

At least in this group of patients fecal S100A12 concentration was more sensitive (but less specific) to detect dogs with a CCECAI ≥ 12 or histopathologic intestinal inflammation than fecal calprotectin concentration.

1. Grellet et al. AJVR, 2013, 74, 5: 706-711.

Conflicts of interest: Financial support of Royal canin

ESCG-O-9

COBALAMIN CONCENTRATION IN GERIATRIC CATS WITH IDIOPATHIC MALABSORPTION, BUT CONCENTRATIONS DECREASE RAPIDLY FOLLOWING CESSATION OF SUPPLEMENTATION. D.A. Williams¹, G. Czarnecki-Maulden². ¹University of Illinois, Urbana, United States of America, ²Nestle-Purina Research, St. Louis, United States of America

Weight loss and malabsorption of fat, protein, cobalamin and tocopherol in the face of normal exocrine pancreatic function have been reported in up to 30-40% of cats older than 12 years of age fed a variety of nutritionally balanced dry and wet foods (Patil AP and Cupp CJ. Proc. Nestle-Purina Compan Anim Nutr Summit, Focus on Gastroenterology, 55-61, 2010). The objectives of this study were to determine if serum cobalamin concentrations increased after oral administration of a cobalamin supplement to affected cats, and the duration of any positive response following cessation of supplementation.

The study evaluated 14 cats older than 12 years of age with fat malabsorption demonstrated by either increased fecal fat (>20%) or subnormal fat digestibility (<80%), but without exocrine pancreatic insufficiency (EPI) as assessed by assay of serum trypsin-like immunoreactivity (fTLI). A commercially available solution of cobalamin containing 1 mg mixed with 1 mL of a liquid flavor enhancer was added to the food of each cat in a single dose each day for 2 months, after which supplementation ceased. Serum cobalamin (assayed by competitive binding assay performed through the GI Laboratory at Texas A&M University and evaluated using reference ranges derived by that laboratory) was determined immediately prior to initiation of supplementation, then weekly for 3 weeks, then monthly for 4 months.

At the start of the study serum cobalamin was subnormal (<290 ng/L) in 4 of the 14 cats (range <150 to 244 ng/L) and within the reference range (290 to 1499 ng/L) in the remaining 10 cats (334 to 1065 ng/L). Serum cobalamin was above the reference range in every cat (1892 to 13109 ng/L) after one week of supplementation and remained above the reference range in every sample collected during the supplementation period, with the exception of two cats with values within the reference range when supplementation was stopped. Serum cobalamin 1, 2 and 3 months after cessation of supplementation ranged from <150 to 1783, <150 to 1494, and <150 to 948 ng/L, respectively. At the end of the study serum cobalamin was subnormal in 5 of the 14 cats.

It is concluded oral cobalamin supplementation can effectively increase serum cobalamin concentrations in geriatric cats with idiopathic chronic enteropathy, but that following cessation of supplementation concentrations decrease rapidly and can become subnormal again within as few as 4 weeks.

Conflicts of interest: The primary author collaborated with Nestle-Purina on the work reported in this abstract, and a co-author is an employee of Nestle-Purina. The primary author

has previously received funding from Iams, Mars, Hills and Nestle-Purina. The author also acts as a paid consultant for the GI-Lab, Texas A&M University.

ESVC-O-1

LEFT ATRIAL FUNCTION DIFFERS BETWEEN DOGS WITH DIFFERENT SEVERITIES OF MYXOMATOUS MITRAL VALVE DISEASE. M. Rishniw¹, D. Dickson², D. Caivano³. ¹Veterinary Information Network, Davis, United States of America, ²Heartvets, Dursley, United Kingdom, ³University of Perugia, Perugia, Italy

Left atrial measurements are crucial in assessing severity of cardiac disease in dogs with myxomatous mitral valve disease (MMVD). However, linear and area dimensions might not provide a comprehensive assessment of patient status, and cannot differentiate between severe subclinical (B2) and clinical disease (CHF). Estimates of left atrial function could provide additional information to help categorize these patients.

We examined 87 dogs with MMVD (25 Normal, 9 B1, 40 B2 and 13 C) presented for cardiac evaluations by 2D echocardiography. Left atrial linear and area dimensions in right parasternal short and long axis views were obtained at 3 time points - early diastole (LA_{MAX}), just prior to mitral valve opening, at the onset of atrial systole (LA_P) and just prior to mitral valve closure (LA_{MIN}). We calculated 4 indices of LA function: total LA emptying fraction (LA_{TEF}), active LA emptying fraction (LA_{ACT}), passive LA emptying fraction (LA_{PAS}) and LA reservoir function (LA_{RES}) for all 4 sets of measurements. We examined the differences in selected LA function indices between different disease stages with a Kruskal Wallis Test with post-hoc multiple comparisons. We also examined the diagnostic accuracy of selected indices of LA function in differentiating dogs in Stage B2 and Stage C (CHF) using ROC analysis.

Three functional indices consistently differed across the various stages of MMVD - LA_{TEF}, LA_{ACT} and LA_{RES}. These differences were most apparent in the RPLA view for linear measurements and RPSA view for area measurements. Dogs with CHF had worse function than all other groups, which differed variably depending on the functional index being examined. LA_{areaACT} showed the best ability to discriminate between B2 and CHF dogs, with a 95% specificity, 69% sensitivity and an AUC of 0.84, but this was no better than use of LA:AO measurements.

Our data suggest that LA function differs between dogs with differing severities of MMVD, but does not provide a clear distinction between dogs with subclinical disease and CHF.

Conflicts of interest: No conflicts of interest reported.

ESVC-O-2

QUANTITATIVE EVALUATION OF CANINE MITRAL VALVE IN DOGS USING THREE-DIMENSIONAL ECHOCARDIOGRAPHY. G. Menciotti, M. Borgarelli, S. Wesselowski, J. Abbott. Virginia-Maryland Regional College of Veterinary Medicine, Blacksburg, United States of America

Real time three-dimensional echocardiography (3DE) has provided new insights into mitral valve (MV) morphology and function in human myxomatous mitral valve disease (MMVD).

Objective: Assess the feasibility of 3DE for evaluating the MV in dogs and describe the 3DE features of the MV in normal dogs and dogs with MMVD.

Materials and Methods: 3DE was used to evaluate 85 consecutive, non-sedated dogs that weighed more than 5 kg. The study population for the morphologic study included 15 normal dogs, and 41 dogs with ACVIM Stages B1 or B2-C MMVD. 3DE image data were digitally recorded and then analyzed offline, using commercially available software.

Results: 3DE image acquisition was feasible in 67/85 (78.8%) consecutive dogs. Patient anxiety (6), arrhythmias (6) and panting (5) explained failure to obtain a 3DE dataset. Forty-one of 67 (61.2%) datasets were of analyzable quality. Body

weight and heart rate were significantly lower in dogs for which it was possible to perform 3DE. Dogs with analyzable 3DE datasets were significantly older and weighed less than dogs in which 3DE could not be analyzed.

The mitral valve of normal dogs is saddle shaped (annulus height to commissural width ratio (AHCWR): 0.22 ± 0.06 [mean \pm SD]) and has an elliptical annulus (sphericity index (SI): 0.91 ± 0.07). The following measurements were significantly related to body surface area (BSA): antero-posterior diameter (APD) ($R^2 = 0.46$, $p < 0.01$), anterolateral-posteromedial diameter (ALPMD) ($R^2 = 0.38$, $p < 0.05$), annulus area (AA) ($R^2 = 0.39$, $p < 0.05$), anterior leaflet length (ALL) ($R^2 = 0.38$, $p < 0.05$), anterior leaflet area (ALA) ($R^2 = 0.38$, $p < 0.05$). These variables were indexed (i) to BSA for subsequent statistical analyses.

Dogs with MMVD had a significantly greater SI, non-planar angle, APDi, ALPMDi, ALAi and ALLi, while having a significantly lower posterior leaflet area (PLA), posterior leaflet length (PLL), annulus height (AH), tenting height (TH), tenting volume (TV), tenting area (TA), and AHCWR compared to normal dogs. AH, TV and TA were significantly greater in normal dogs, compared to dogs with MMVD. SI, APDi, ALPMDi, ALAi and ALLi were significantly greater in dogs with Stages B2-C MMVD, compared to normal dogs and those in Stage B1. PLL and PLA were significantly lower in B2-C dogs, compared to normal dogs. TH was significantly different between the three groups; greatest in normal dogs and lowest in dogs in Stages B2-C, suggesting that flattening of the MV occurs with disease progression.

Conclusions: 3DE assessment of the canine MV is feasible. Morphologic changes associated with MMVD progression are presented.

Conflicts of interest: This research is funded by CEVA Santé Animale.

ESVC-O-3

REAL-TIME 3- AND 2-DIMENSIONAL ECHOCARDIOGRAPHIC ASSESSMENT OF EFFECTIVE REGURGITANT ORIFICE AREA IN DOGS WITH MYXOMATOUS MITRAL VALVE DISEASE. A. Tidholm¹, K. Höglund², J. Hågström³, I. Ljungvall³. ¹Albano Animal Hospital, Danderyd, Sweden, ²Dept of Anatomy, Physiology and Biochemistry, Faculty of Veterinary Medicine, Uppsala, Sweden, ³Dept of Clinical Sciences, Faculty of Veterinary Medicine, Uppsala, Sweden

Effective regurgitant orifice area (EROA), calculated from a 1-dimensional measurement of the width of vena contracta (VC) as the narrowest portion of the proximal regurgitant jet, might be used to estimate severity of mitral regurgitation (MR). However, this simplified assumption only holds when the EROA is circular, which might not be true in dogs with myxomatous mitral valve disease (MMVD). The aim of the study was to compare measured EROA using color Doppler real-time 3 dimensional echocardiography (RT3D) with calculated EROA estimated by 2 dimensional echocardiography (2D) in 4 chamber (4ch) and 2 chamber (2ch) views of the left ventricle (LV) in dogs with MMVD.

Ninety-three privately owned dogs of 32 breeds diagnosed with naturally acquired MMVD were examined using 2D and RT3D. According to the ACVIM classification of congestive heart failure (CHF), 23 dogs were classified with CHF (2 in class C1 and 21 in class C2) and 70 dogs without CHF (65 dogs in class B1 and 5 dogs in class B2). Age ranged from 1 to 15 years (median 10 years), and body weight ranged from 2.5 to 35 kg (median 10 kg). Fifty-nine males (63%) and 34 females (37%) were included, and heart rate ranged from 80 to 222 beats/minute (median 126 b/min).

EROA was calculated from 2D measurements of VC diameter, in the 4ch view only (assuming a circular regurgitant orifice), and from measurements of VC diameter in both 4ch and 2ch views (assuming an elliptical regurgitant orifice) of LV. Bland-Altman plots were used to compare EROA measured by RT3D with calculated EROA obtained from 2D 4ch and 4ch/2ch LV views.

None of the 2D estimations of EROA showed good agreement with the measured RT3D EROA when corrected for BSA, and the difference between methods increased with increasing EROA. The difference between RT3D and 2D methods normalized to the mean EROA value did not increase with increasing EROA, but showed a systematic underestimation of EROA by 60% (4ch) and 40% (4ch/2ch), respectively, compared to RT3D. The beat-to-beat variation of EROA assessed by RT3D ($n = 56$) had a coefficient of variation ranging from 2.8% to 68% (median 30%).

In conclusion, substituting assessment of EROA with a measurement of VC in 1 or 2 dimensions might underestimate the MR severity in dogs with MMVD. In some dogs, the beat-to-beat variation of the EROA was large, thereby necessitating the need for several consecutive measurements.

Conflicts of interest: No conflicts of interest reported.

ESVC-O-4

MICRORNAS EXPRESSION IN CANINE MYXOMATOUS MITRAL VALVE DISEASE. C.C.L. Lu, D.J.A. Argyle, B.M.C. Corcoran. The Roslin Institute, University of Edinburgh, Edinburgh, United Kingdom

MicroRNAs (miRNA) are short (19-22 nucleotides), single-stranded, non-coding RNAs that specifically anneal with complementary sequences in multiple mRNA targets, and they silence mRNAs and suppress downstream protein translation. A miRNA can act as a fine-tuner of gene expression or an on/off switch. These features highlight the potential of miRNAs as therapeutic targets. The role of miRNAs in myocardial fibrosis and hypertrophic cardiomyopathy has been widely studied in human patients. However, there is no data available for canine and human myxomatous mitral valve disease (MMVD). The aim of this study was to investigate miRNA transcriptomics in canine MMVD by using global transcriptional profiling, miRNA target prediction software (DIANA Tool, TargetScan 6.2) and network analysis software (BioLayout Express^{3D}).

Four myxomatous mitral valves (CKCS) and 4 controls valves were profiled using the Affymetrix Canine Gene 1.0 ST Array. In total 29 out of 302 miRNAs were found to be statistically significantly differentially expressed (down-regulated) based on the false discovery rate, p-value, and fold-change. Expression of three miRNA (cfa-miR-23b, cfa-miR-29c, cfa-miR-218) were also validated by quantitative polymerase chain reaction (Q-PCR, TaqMan), and the results were in agreement with the microarray findings. For network analysis and visualization, Markov clustering algorithms were conducted in BioLayout Express^{3D}, and major clusters of miRNAs were exported and uploaded to the DIANA-miRPath (KEGG pathway) web-server. The pathways identified in the main cluster were attributed to the biological functions of focal adhesion, cytoskeleton (actin) regulation, TGF- β signalling, glycosaminoglycan biosynthesis, osteoclast differentiation, NOTCH signalling and VEGF signalling.

The most significantly down-regulated miRNA in MMVD was cfa-miR-218, which is an endothelial specific miRNA shown to regulate endothelial migration and vessel patterning. The top predicted target of cfa-miR-218 is glucuronic acid epimerase (GLCE) which is the main enzyme controlling heparan sulphate biosynthesis. Other interesting findings were down-regulation of cfa-miR-29 and members of the cfa-miR-23 family. Cfa-miR-29 targets multiple extracellular matrix transcripts, such as collagens, elastin, integrin, laminin, MMP (matrix metalloproteinase) and ADAMTS (a disintegrin and metalloproteinase with thrombospondin motifs), whereas cfa-miR-23 targets hyaluronic acid synthase 2 (Has2). Since the major pathology of MMVD is aberrant turnover of extracellular matrix proteins, this may be linked to miRNA regulation. Dysregulation of valve miRNAs might be potential therapeutic targets in the treatment of canine MMVD.

Conflicts of interest: No conflicts of interest reported.

ESVC-O-5
INCREASED LEFT HEART SIZE PREDICTS RISK OF CONGESTIVE HEART FAILURE IN CAVALIER KING CHARLES SPANIELS WITH MITRAL REGURGITATION CAUSED BY MYXOMATOUS VALVE DISEASE. S. Eriksson-Palojarvi¹, K. Hansson², H. Duelund Pedersen³, J. Haukka¹, J. Häggström². ¹University of Helsinki, HELSINKI, Finland, ²Swedish University of Agricultural Sciences, UPPSALA, Sweden, ³Novo Nordisk, Denmark

Mitral regurgitation (MR) progresses slowly, but dogs living long enough often develop congestive heart failure (CHF). However, tools to predict onset of CHF are sparse.

225 echocardiographic examinations in 78 dogs were performed in a longitudinal, multicenter study with a surveillance time of up to 4.5 years. Client-owned dogs were enrolled at the University Hospitals in Finland, Sweden and Denmark (subset to the SVEP study).

Left ventricular end diastolic (LVIDd) and systolic (LVIDs) diameters, fractional shortening (FS), left atrial (LA) and aortic root (Ao) diameters were estimated. Values were normalized for body size (nLVIDd, nLVIDs, and nLA, respectively) and, for comparison, ratios to aortic root were calculated (LVIDd/Ao, LVIDs/Ao and LA/Ao, respectively).

A Cox's proportional hazard analysis with a counting process approach was used. Spline smoothed graphical models were constructed to evaluate linearity of hazards. Curves were then used to find cut-off values for interval hazard ratios (HRs).

The HR for nLVIDd, nLVIDs and nLA (per 0.1 unit, 95% confidence intervals), were 1.5 (1.32-1.70, $p = 0.00034$), 1.3 (1.05-1.62, $p = 0.016$), and 1.5 (1.28-1.83, $p = 0.0039$), respectively. The HRs for LVIDd/Ao, LVIDs/Ao and LA/Ao (0.1 unit increase) were 1.3 (1.14-1.55, $p = 0.0025$), 1.1 (0.99-1.32, $p = 0.07$), and 1.4 (1.24-1.66, $p = 0.0041$), respectively. The HR for FS was 1.1 (1.05-1.19, $p = 0.00037$).

The relative hazard plot presented a steep increase for FS values above 31%. HRs for intervals $31 < 36\%$, $36 < 40\%$, and $\geq 40\%$ were 1.0 (0.2-5.1, $p = 0.99$), 4.8 (1.2-18.4, $p = 0.023$), and 9.1 (2.4-33.7, $p = 0.00098$), respectively. The HR for nLVIDd increased linearly. HRs for intervals $1.73 < 1.86$, $1.86 < 1.92$, $1.92 < 2.1$ and ≥ 2.1 were 0.7 (0.1-6.6, $p = 0.7$), 6.0 (1.3-26.7, $p = 0.02$), 6.8 (1.9-25.1, $p = 0.0037$), and 12.5 (3.8-40.1, $p = 0.00088$), respectively. In contrast, the hazard for nLVIDs remained stable until 1.4, whereafter it increased. The HRs for nLVIDs ($1 < 1.33$, $1.33 < 1.68$, $1.68 < 1.86$ and ≥ 1.86) were 1.6 (0.6-4.6, $p = 0.35$), 3.4 (1.0-11.5, $p = 0.048$), 78 (9.6-634, $p = 0.030$), and 47.6 (7.1-316, $p = 0.044$), respectively. HRs for values normalized to Ao diameter behaved in a parallel way.

We conclude that FS, left ventricular and atrial size may be used to predict CHF. However, because the value of a HR is dependent on the unit used and, more essentially, does not account for nonlinear change in hazard, interpretation of hazards is challenging. In contrast, interval hazards are only dependent on the reference interval used. Therefore they are easier to implement in every day clinical work.

Conflicts of interest: No conflicts of interest reported.

ESVC-O-6
LONGITUDINAL STUDY OF SYSTOLIC ARTERIAL BLOOD PRESSURE IN DOGS WITH VARIOUS STAGES OF MITRAL VALVE DISEASE. C. Koutinas, Z. Polizopoulou, A. Dasopoulou, A. Koutinas. Faculty of Veterinary Medicine, Aristotle University of Thessaloniki, Thessaloniki, Greece

Systemic arterial hypertension is not frequently recognized in dogs with mitral valve degeneration (MVD), although borderline hypertension is difficult to assess, mainly because of different measurement techniques, inter-operator variability and, most importantly, examination-related stress.

The object of this study was to evaluate systolic arterial blood pressure (SBP) at initial presentation and at regular intervals in dogs with various clinical stages of MVD.

Fifty six dogs with MVD that had not received any heart medication prior to admission, were included in the study. Based on the ISACHC staging system, 22 were assigned to class I (Group

A), 18 to class II (Group B) and 16 to class III. Small-breed dogs and miniature Poodles, in particular, were overrepresented. Comorbidities that could affect SBP were ruled out prior to enrollment. SBP was measured using a commercially available veterinary oscillometric device, by applying the proper cuff on the cephalic artery. Dogs were left to acclimate for 10 - 15 minutes and measurements were always taken by the same investigator, before any other examination was performed, with the dog sitting on the owner's lap. A total of 5 readings were taken, outlier values were discarded and the mean of the 3 remaining measurements was documented. After initial consultation, treatment was customized according to the clinical stage. SBP was then measured every 2 months, up to 6 months after initial admission. At presentation, all class I dogs had SBP > 140 mm Hg, with only 5/22 having SBP ≥ 150 mm Hg, whereas all class II dogs had SBP < 140 mm Hg. Of class III dogs, 6 had SBP > 140 mm Hg, and 2 had SBP ≥ 150 mm Hg. A linear mixed effects model was used to assess the temporal variability of the measured parameters between groups.

Groups were matched for gender, age and body weight. Blood pressure measurements, for the duration of the study, were higher in Group A dogs, compared to groups B and C ($P < 0.001$). At the same time, Group C had significantly higher SBP values than Group B dogs ($p < 0.001$).

Asymptomatic MVD dogs seem to have higher SBP measurements, compared to those with clinical evidence of heart failure. Whether this difference is stress-related, a maladaptive mechanism of sympathetic and RAAS activation to MVD or idiopathic remains to be elucidated.

Conflicts of interest: No conflicts of interest reported.

ESVC-O-7
CAN THE CALCIUM UPTAKE GENES EXPRESSED IN BLOOD REFLECT THE MYOCARDIAL DYSFUNCTION IN CHRONIC HEART FAILURE? J. Lee, M. Mizuno, T. Mizuno, K. Harada, T. Sawada, A. Shinoda, S. Uchida, M. Uechi. NIIHON UNIVERSITY, Fujisawa, Japan

Sarcoplasmic reticulum (SR) Ca^{2+} -ATPase and its regulatory proteins are pivotal determinants of myocardial active relaxation via calcium uptake against the SR-cytoplasmic gradient. The lowered density of the SR Ca^{2+} -ATPase has been well demonstrated in many species during chronic hemodynamic overload. The genes linked to SR calcium uptake were reported not only being expressed in peripheral blood but serving as potential cardiac biomarkers in dogs with chronic mitral regurgitation, such as SR Ca^{2+} adenosine triphosphatase isoform 2 α (*SERCA2 α*), phospholamban (*PLN*), and HS-1 associated protein X-1 (*HAX-1*). The aim of this study is to determine whether the target genes expressed in the blood will be translatable to the myocardial setting as cardiac biomarkers.

The mRNA expression levels of the target genes (*SERCA2 α* , *PLN*, *HAX-1*) from biopsied left ventricle (LV) and peripheral white blood cells (PWBC) in 129 surgical mitral valve repair cases were estimated with quantitative real-time PCR using comparative Ct method with GAPDH. The gene expression levels in LV and PWBC were compared and their clinical relationships were evaluated. The diagnostic power of the genetic expressions in PWBC was analyzed by comparing to those of 33 normal dogs.

The levels of all target genes expressed in LV and PWBC were highly correlated each other in linear regression analysis ($p < 0.0001$; *SERCA2 α* , $r = 0.7425$, $R^2 = 0.5513$; *PLN*, $r = 0.7720$, $R^2 = 0.5959$; *HAX-1*, $r = 0.6598$, $R^2 = 0.4353$), although LV and PWBC showed different expression levels in a paired comparison ($p < 0.0001$). According to the severity of the heart failure (ISACHC), the expression levels of all genes were gradually and significantly reduced in both LV and PWBC ($p < 0.01$). Especially, the *SERCA2 α* and *PLN* expressed in PWBC could clearly discriminate all ISACHC groups from the control ($p < 0.0001$). Multivariate regression adjusted by age and body weight revealed that *SERCA2 α* and *PLN* in LV were negatively associated with LV internal systolic dimension ($p = 0.0251$, adjusted $R^2 = 0.524$ and $p = 0.012$, adjusted $R^2 = 0.530$, respectively). *PLN* was also negatively related with LV internal

diastolic dimension ($p = 0.037$, adjusted $R^2 = 0.573$). Additionally, receiver-operating characteristic analysis using PWBC showed high area under the curve (AUC) values for all target genes on overall ISACHC groups ($p < 0.0001$; *SERCA2 α* , AUC=0.9212; *PLN*, AUC=0.8936; *HAX-1*, AUC=0.8797).

In conclusion, the transcriptional changes of the calcium uptake related genes in PWBC may be able to reflect myocardial hemodynamic stress as well as to be utilized as promising cardiac biomarkers.

Conflicts of interest: No conflicts of interest reported.

ESVC-O-8

INCREASED NORMALIZED PULMONARY TRANSIT TIMES AND PULMONARY BLOOD VOLUMES IN CARDIOMYOPATHIC CATS WITH OR WITHOUT CONGESTIVE HEART FAILURE. P. Modler¹, A. Streitberger¹, J. Haggström². ¹Traunkreis Vet Clinic, Saftledt, Austria, ²Swedish University of Agricultural Sciences, Faculty of Veterinary Medicine, Uppsala, Sweden

The aim of this study was to estimate heart-rate normalized pulmonary transit times (nPTT) in cardiomyopathic cats with or without congestive heart failure (CHF), to assess potential associations of echocardiographic variables and nPTT, and to evaluate nPTT as a test for presence of CHF.

48 privately owned cats were included. nPTT was measured using echocardiography and the ultrasound contrast media SonoVue[®] in 3 groups of cats: healthy cats (Group 1), cats with cardiomyopathy (CM) but without CHF (Group 2), and cats with CM and CHF (Group 3). Receiver operating characteristic curves (ROC) were created for nPTT, left atrial diameter (LAD) and the left atrial to aortic root ratio (LA:AO) to assess and compare their usefulness as tests for presence of CHF. Interrelations between pulmonary blood volume (PBV), nPTT, stroke volume (SV) and echocardiographic variables were investigated by means of uni- and multivariate analysis.

nPTT values in group 1, group 2 and group 3 were 3.63 (interquartile range (IQR) 3.20-4.22), 6.09 (IQR 5.0-7.02), and 8.49 (IQR 7.58-11.04), respectively. Values were significantly different between all 3 groups. Pulmonary blood volumes in group 1, group 2, and group 3 were 27.94 ml (IQR 21.02 ml-33.17 ml), 42.83 ml (IQR 38.46 ml-50.36 ml) and 49.48 ml (IQR 38.84 ml-64.39 ml). SV, PBV and shortening fraction <30% were significant predictors of nPTT. nPTT and LA:AO ratio, not SV were the main predictors of PBV. Analyzing ROC for nPTT as a clinical test for CHF yielded an AUC of 0.956 which was similar for LA:AO ratio.

nPTT may be useful test for the presence of CHF in cats with CM and as a measure of cardiac performance. nPTT and LA:AO ratios predict CHF with equal accuracy. Increased PBV is significantly associated with higher nPTT and LA:AO ratios. Both decreased SV and increased PBV explain the increased nPTT in cardiomyopathic cats.

Conflicts of interest: The author received a travel scholarship from Zoetis to attend this congress.

ESVC-O-9

DIFFERENCE IN PERIPHERAL AND CENTRAL VENOUS BLOOD GLUCOSE CONCENTRATIONS IN ACUTE ARTERIAL THROMBOEMBOLISM IN CATS AND DOGS. I. Aroch, E. Kelmer, G. Segev, S. Klainbart. Hebrew University of Jerusalem, Rehovot, Israel

Acute arterial thromboembolism (AATE) occurs commonly in cats, and less frequently in dogs, mostly resulting in limb paresis or paralysis. Diagnosis is based typically on physical examination and advanced imaging. Diminished affected-limb peripheral blood flow induces changes in several analytes concentrations in affected limb venous samples, compared to their peripheral venous concentration. We hypothesized that in AATE, local, affected-limb venous glucose concentration decreases below reference interval, while systemic glucose

concentration remains unaffected. The study included 3 groups for each species: paralytic AATE cases, non-ambulatory controls with limb paralysis of orthopedic or neurologic disorders, and ambulatory controls diagnosed with various diseases. Systemic and peripheral, affected-limb blood glucose concentrations were measured. Group absolute (Δ Glu) and relative (% Δ Glu) differences were compared. No procedure-associated complications or pain were noted. Peripheral blood glucose concentrations were decreased ($P \leq 0.006$) only in cats and dogs with AATE. Δ Glu and % Δ Glu were higher in the AATE groups in both cats and dogs compared to their respective control groups ($P < 0.0001$, $P < 0.001$, respectively), with no differences between the control groups. Receiver operator characteristics analysis of Δ Glu and % Δ Glu as predictors of AATE in cats had areas under the curve of 0.96 and 1.00, respectively, and 0.99 and 1.00, in dogs, respectively. Δ Glu cut-offs of 30 mg/dL and 16 mg/dL, in cats and dogs, respectively, corresponded to sensitivity and specificity of 100% and 90% in cats, respectively, and 100% in dogs. Δ Glu and % Δ Glu are extremely accurate, readily-available, simple diagnostic markers of AATE in cats and dogs.

Conflicts of interest: No conflicts of interest reported.

ESVC-O-10

PERIPHERAL GLYCAEMIA IN DOGS WITH LIMB THROMBOSIS: A PROSPECTIVE STUDY. M. Dolera, L. Malfassi, R. Vailati Facchini, S. Finesso, G. Mazza, S. Marcarini. La Cittadina Fondazione Studi e Ricerche Veterinarie, Romano, cr, Italy

Glycaemia determination is usually included in routine biochemistry panels. No works are devoted to the evaluation of peripheral glycaemia in animals suffering from arterial thrombosis. The aim of this study was to document the peripheral glycaemia variations in hypoperfused limbs of patients affected by MRI-confirmed arterial thrombosis.

Eleven dogs referred for monoparesis or paraparesis were recruited. Inclusion criteria were a clinical examination supportive of limb hypoperfusion and availability of blood cell count, biochemical profile and urine analyses. Before MRI examination, peripheral glycaemia was tested. Two blood samples were obtained, one from the affected limbs and one from a healthy limb. Plasma glycaemia was measured using an automated glucose analyser. All the patients underwent a total body MRI (MRI Intera 1.5T, Philips Medical Systems) that provided the final diagnosis. The arterial thrombosis location was documented and the entity was scored. All the eleven patients were diagnosed with a peripheral thrombosis involving an arterial vessel and in some cases the relative branches. The thrombus was located: in the abdominal aorta (7/11), in the subclavian artery (1/11), in the axillary artery (1/11), in the iliac arteries (2/11). Of the total amount of abdominal aortic thrombosis, 3/7 involved also the internal iliac arteries, 2/7 the external ones and 2/7 both internal and external. The extent of the thrombosis was classified as grade 1 (G1), when the greatest portion of the thrombus did not reach half of the vessel lumen (1/11 patients); grade 2 (G2), when the greatest portion of the thrombus was between 1/2 and 2/3 of the vessel lumen (7/11); grade 3 (G3), when the thrombus exceeded 2/3 of the lumen (3/11). A substantial decrease in peripheral glycaemia values was found in sampling arising from the thrombosis-affected limbs. Comparing thrombosis-affected limbs values with healthy limbs measurements from the same patient, the reduction was found from 17.65% to 34.41%. Accounting only the G3 scored patients, the percentage of reduction was found up to the 28.34% suggesting a proportional decrease related to the grade of occlusion.

Results from this study suggest that peripheral glycaemia values are affected by limb hypoperfusion disorders. If an arterial thrombosis is suspected, samples from the affected limbs and the healthy ones could be used to compare glycaemia values and to support the early stage therapy in anticipation of diagnostic imaging. Further studies are needed to confirm the proportional relation of the decrease with thrombus entity.

Conflicts of interest: No conflicts of interest reported.

ESVC-O-11
RIGHT PULMONARY VEIN TO PULMONARY ARTERY RATIO: A NEW ECHOCARDIOGRAPHIC INDEX OF PULMONARY HYPERTENSION IN WEST HIGHLAND WHITE TERRIERS WITH IDIOPATHIC PULMONARY FIBROSIS.
 E. Roels¹, A.C. Merveille¹, E. Krafft¹, F. Farnir¹, S. Gomart², C. Clercx¹, K. Mc Entee². ¹ULG, Liege, Belgium, ²ULB, Brussels, Belgium

Canine idiopathic pulmonary fibrosis (CIPF) is a progressive interstitial lung disease mainly affecting West Highland white terriers (WHWT). Pulmonary hypertension (PH) may develop secondary to hypoxic vasoconstriction and/or pulmonary parenchymal infiltration. In the absence of measurable tricuspid regurgitation (TR), this co-morbid condition may be difficult to diagnose non-invasively. The degree of cardio-pulmonary impairment in CIPF dogs can be evaluated through blood gas analysis (BGA) and 6 minute walking test (6MWT). A new echocardiographic index, the right pulmonary vein to pulmonary artery ratio (PV/PA) has been described for the detection of pulmonary venous hypertension. The aim of this study was to investigate PV/PA in CIPF in order to determine its utility in the detection of PH and in the assessment of cardio-pulmonary disease severity. This prospective clinical cohort study included 10 WHWT with CIPF (Group A), 9 healthy WHWT (Group B) and 25 healthy dogs from other breeds (Group C). Diameters of right PV and PA were measured, in bi-dimensional (BD) and M-modes (MM), in a parasternal right long axis view, at the end of the T wave. Other echocardiographic parameters for evaluation of PH were also measured: speed of TR, acceleration time to ejection time ratio of the pulmonary flow (AT:ET) and pulmonary artery to aorta ratio (PA/Ao). BGA was performed in 14 dogs (9, 1 and 4 in groups A, B and C) and 6MWT in 17 dogs (8, 6 and 3). Values are given as mean±SD. In BD and MM mode, the PV/PA ratio was lower in group A (MM: 0.62 ± 0.25 , BD: 0.51 ± 0.20) compared to group B (MM: 0.98 ± 0.17 , BD: 0.93 ± 0.12 , $P \leq 0.01$) and group C (MM: 1.03 ± 0.13 , BD: 1.00 ± 0.10 , $P \leq 0.0001$). The changes in PV/PA were both due to an increase of PA ($P \leq 0.01$) and a decrease of PV ($P \leq 0.05$). TR was found in 60% of dogs with CIPF; mean pressure gradient was 34.03 ± 16.90 mmHg. AT:ET was lower in group A (0.42 ± 0.08) compared to group C (0.50 ± 0.04 , $P = 0.002$) and tended to be lower compared to group B (0.48 ± 0.07 , $P = 0.09$). PA/Ao was not statistically different between groups. PV/PA was correlated with arterial pO_2 values (B mode: $r = 0.870$, $P = 0.0001$) and results of the 6MWT (B mode: $r = 0.791$, $P = 0.0003$). PV/PA was also correlated with AT:ET and the speed of TR, but not with PA/Ao. In conclusion, in WHWT affected by CIPF, PV/PA is a useful indicator of PH and could serve in the assessment of disease severity.

Conflicts of interest: No conflicts of interest reported.

ESVC-O-12
CHARACTERIZATION AND LONG-TERM OUTCOME OF VENTRICULAR SEPTAL DEFECTS IN DOGS AND CATS.
 R. Abrantes, J. Novo Matos, N. Summerfield, T. Glaus. University of Zurich, Zurich, Switzerland

Ventricular septal defect (VSD) is the fourth most common congenital cardiac defect in dogs and the most common in cats. The aim of this study was to evaluate the long-term outcome in VSD patients. Case records of 95 animals were reviewed, 46 of these re-evaluated echocardiographically and 49 followed up by phone interview only. Out of 53 dogs Pug was the most common breed (11%) followed by Border Terrier (8%). Out of 42 cats Domestic Short Hair was most common (55%) followed by Main Coon (19%). Isolated VSDs were present in 29 dogs and 26 cats. Complex defects (CDs) were present in 40 cases, most frequent anomalies being sub-aortic stenosis (9 dogs, 1 cat), pulmonic stenosis (5 dogs, 3 cats), Tetralogy of Fallot (3 dogs, 2 cats), cushion defects (5 cats) and double-chambered right ventricle (DCRV) (5 dogs, 4 cats; in 2/5 dogs not present initially supporting the cause-and-effect theory). Eisenmenger was observed in 1 dog and 2 cats. Aortic insufficiency, not considered a CD, was noted in 10 dogs. In 3 dogs and 6 cats (15% of isolated VSDs

and 1 cat with a DCRV) the defect closed spontaneously. Nine dogs and 9 cats (22%) died of non-cardiac causes with an age at death of 5 to 204 (mean 63.8) months; 7 dogs and 8 cats died due to cardiac causes with an age at death of 0.25 to 204 (mean 37.5) months. Cardiac deaths were sudden (2 dogs with CDs) or euthanasia for left sided congestive heart (CHF) failure associated with CDs (3 dogs, 1 cat); right sided CHF associated with CDs (2 cats); biventricular failure (1 cat with CD); weakness (Eisenmenger, 1 dog and 1 cat; Fallot 1 dog; CD 1 cat). Two cats developed CHF due to unrelated HCM. Only one dog with an isolated VSD was euthanized for CHF.

These results indicate that spontaneous VSD closure occurs more often than previously thought, most patients with isolated restrictive VSDs live a normal life span without surgical intervention, but non-restrictive VSDs or complex defects can be associated with significant morbidity and mortality. Echocardiography early in life is crucial to identify the anomaly and CDs, as well as useful to prognosticate long-term outcome and to identify patients where a surgical intervention should be considered if available. Follow-up echocardiography is indicated to corroborate the prognosis, to detect complications due to the VSD and to detect unassociated acquired cardiac diseases.

Conflicts of interest: No conflicts of interest reported.

ESVC-O-13
TISSUE DOPPLER AND STRAIN IMAGING ALTERATIONS IN AGING PTPLA-DEFICIENT LABRADOR RETRIEVERS WITHIN A FRENCH PEDIGREE SEGREGATING CENTRONUCLEAR MYOPATHY. V. Gouni¹, V. Chetboul¹, J. Blondelle², E. Trehiou-Sechi¹, C. Misbach¹, R. Tissier³, J.L. Pouchelon¹, I. Barthelemy⁴, P. Aguilar⁴, N. Blanchard-Gutton⁴, J.L. Thibaud⁴, H.P. Lefebvre⁵, F. Pilot-Storck², S. Blot⁴, L. Tiret². ¹UCA, CHUVA, Ecole Nationale Vétérinaire d'Alfort, Université Paris-Est, Maisons-alfort, France, ²CNM Project, Université Paris-Est, Ecole Nationale Vétérinaire d'Alfort, Maisons-alfort, France, ³INSERM, Unité Mixte de Recherche U955, Equipe 03, Créteil, France, ⁴UPR de Neurobiologie, Université Paris-Est, Ecole Nationale Vétérinaire d'Alfort, Maisons-alfort, France, ⁵Unité de Recherche Clinique, INP, Ecole Nationale Vétérinaire de Toulouse, Toulouse, France

Centronuclear myopathy (CNM) is the most prevalent congenital inherited disorder affecting skeletal muscles in Labrador Retrievers. This disabling condition segregates worldwide and a recessive loss-of-function founder mutation was identified in the *Protein Tyrosine Phosphatase-Like, member A* gene (*PTPLA/HACDI*).

The objectives of this study were 1) to describe *PTPLA* expression pattern in hearts from homozygous wild type (WT), heterozygous (HET) and homozygous mutated (CNM) dogs, 2) to assess and compare the left myocardial function in aging WT, HET and CNM dogs.

For this purpose, seven WT, four HET and eleven CNM dogs were included in the study. *PTPLA* mRNA levels were assessed by RT-PCR and RT-qPCR. All dogs were examined using conventional echocardiography, 2D color Tissue Doppler Imaging (TDI) and TDI-derived strain imaging.

We found that the expression of the two wild type *PTPLA* splice isoforms increased post-natally in WT dogs. Their levels were halved in HET dogs and drastically reduced in CNM dogs. In both HET and CNM dogs, a slight left ventricular hypertrophy was detected using conventional echocardiography. TDI and strain imaging revealed that the left ventricular myocardial function was significantly altered in both HET and CNM dogs compared to WT dogs. Moreover, these functional defects were associated with significantly higher values of systemic arterial blood pressure, although maintained within normal ranges.

In conclusion, subclinical myocardial alterations were detected in both HET and CNM aging dogs from our French pedigree, suggesting a role for *PTPLA* in long-term cardiovascular homeostasis. These findings prompt globalized confirmation in additional *PTPLA* deficient dogs, which may thus be considered as a new large-size model for human left ventricular sub-clinical myocardial dysfunction.

Conflicts of interest: No conflicts of interest reported.

ESVC-O-14

MACROSCOPIC EVALUATION OF THE MITRAL VALVE CHORDAE TENDINEAE DURING MITRAL VALVE REPAIR. T. Mizuno¹, H. Katagiri², M. Mizuno¹, K. Harada¹, M. Furukoshi¹, M. Uechi¹. ¹Japan animal specialty medical research institute, Yokohama, Japan, ²Nihon University, Fujisawa, Japan

Mitral regurgitation (MR) secondary to degenerative mitral valve disease (DMVD) is the most common heart disease in dogs. In dogs with MR, mitral valve prolapse caused by degeneration of the mitral valve leaflet, chordae tendinae extension and/or rupture and mitral annulus dilation are observed. However, limited data are available on morphological changes in dogs with MR. Currently, there are no studies confirming the anomaly of the mitral complex via direct observation in living dogs with mitral regurgitation. At our institution over the last ten years, approximately 300 dogs have undergone mitral valve repair. During surgery, the anomaly of mitral complex can be observed macroscopically (directly visualized). To our knowledge, this is the first study evaluating the anomaly of the mitral valve leaflet and chordae tendinae in dogs undergoing mitral valve repair.

Animals: Dogs that underwent mitral valve repair with CPB at Nihon University between February 2010 and June 2013 were included in this study.

Methods: Confirmation of chordae tendinae rupture was visually confirmed during surgery. The sites of chordae tendinae rupture were also recorded at that time. Septal chordae and mural chordae were divided three division depend on the site (S1, S2, S3 and M1, M2, M3 respectively).

Results: Ninety eight dogs were included in this study. The mean age and body weight were 8.8 ± 1.8 years and 5.15 ± 3.24 kg, respectively. Of the 98 dogs, ruptured chordae was observed in 80 dogs (81.6%). Septal leaflet chordae was ruptured in 73 dogs (74.5%) and mural leaflet chordae was ruptured in 25 dogs (25.5%). Chordae of both leaflets were ruptured in 18 dogs (18.4%). No chordal rupture was observed in 18 dogs (18.4%). In the dogs with ruptured septal chordae, the chordae between S2 and S3 was most often ruptured ($n = 37$, 51%).

Conclusion: In this study, rupture of the septal chordae tendinae was most commonly observed. This is the first pilot study to visually evaluate the anomaly of the mitral valve leaflet and chordae tendinae in dogs undergoing mitral valve repair. Future studies comparing pathological changes and molecular biological analysis to gross findings of mitral chordae tendinae in dogs undergoing mitral valve repair may be useful in advancing the understanding of the disease.

Conflicts of interest: No conflicts of interest reported.

ESVC-O-15

TWO-DIMENSIONAL AORTIC VALVE MEASUREMENTS DIFFER THROUGHOUT DIASTOLE. D.P. Dickson¹, D. Cavanaugh², M. Rishniw³. ¹HeartVets, Gloucestershire, United Kingdom, ²Department of Veterinary Medicine, University of Perugia, Umbria, Italy, ³Department of Clinical Sciences, Cornell University, Ithaca, ny, United States of America

Echocardiographic aortic valve (Ao) measurements are routinely obtained during cardiac evaluation of patients. Cardiologists commonly use diastolic Ao measurements to obtain ratiometric weight-independent estimates of dimensions of other cardiac structures, most commonly the left atrium (LA). However, no consensus exists about the point in diastole at which Ao measurements should be obtained - immediately after closure of the aortic valve, when LA size is largest (Ao_{MAX} , but often with least distinct margins), during the P-wave of the ECG (Ao_P) and at the onset of ventricular electrical systole, when LA size is smallest (Ao_{MIN}). We examined the linear and area dimensions of the Ao (AoD and AoA) to determine if clinically significant differences exist at 3 distinct diastolic time-points, or if these measurements could be interchangeable.

We examined 130 patients (114 dogs and 16 cats) presented for cardiac evaluations by 2D echocardiography. Three replicates of each time-point linear and area measurement (Ao_{MAX} , Ao_P , Ao_{MIN}) were obtained in each patient and averaged for

analysis. Only patients with aortic valve disease and those with atrial fibrillation were excluded from analysis. Beat-to-beat variability of the Ao measurements was determined. Standard and normalized limits of agreement (LoA) plots were generated for each pairwise comparison. The frequency of each Ao measurement being the largest or smallest within-patient measurement was determined, and compared via repeated measures ANOVA.

All pairwise agreement plots of both AoD and AoA demonstrated heteroscedasticity; normalized AoD plots showed 95% LoA to be 15% of the mean AoD measurement, with a bias of approximately 6.5% for $AoD_{MAX}-AoD_{MIN}$, 4% for $AoD_{MAX}-AoD_P$, and 3% for AoD_P-AoD_{MIN} . Normalized AoA plots showed 95%LoA to be 20% of the mean AoA measurement, with a bias of approximately 12% for $AoA_{MAX}-AoA_{MIN}$, 5% for $AoA_{MAX}-AoA_P$, and 7% for AoA_P-AoA_{MIN} . AoD_{MAX} was the largest measurement in 93/131 (71%) patients and AoD_{MIN} was the smallest measurement in 90/131 (69%) patients; AoA_{MAX} was the largest measurement in 96/131 (73%) patients and AoA_{MIN} was the smallest measurement in 95/131 (73%) patients. RMANOVA confirmed that $Ao_{MAX} > Ao_P > Ao_{MIN}$ ($p < 0.0001$). Median within-patient within-measurement variability was 5% for AoD and 9% for AoA measurements.

Our data suggest that Ao measurements differ throughout diastole, with $Ao_{MAX} > Ao_P > Ao_{MIN}$. The disparity is greater for area than linear estimates. The degree of disagreement between Ao_{MAX} and Ao_P is small and similar to the within-measurement variability. Thus, using either Ao_{MAX} or Ao_P measurements should result in similar ratiometric estimates of cardiac dimensions.

Conflicts of interest: No conflicts of interest reported.

ESVC-O-16

CARDIAC MITOCHONDRIAL AND CYTOSKELETAL CHANGES IN FELINE HYPERTROPHIC CARDIOMYOPATHY: AN ULTRASTRUCTURAL STUDY. L.B. Christiansen, P. Hyttel, J. Koch. University of Copenhagen, Frederiksberg, Denmark

Feline hypertrophic cardiomyopathy (fHCM) is the most common heart disease in cats. HCM is considered an inherited disease of the sarcomere and fHCM has been linked to mutations in one sarcomere protein i.e. MYBPC3. However, the pathophysiologic mechanisms behind disease development and progression are largely unknown. In this study we investigate whether mitochondrial morphological changes in the myocardium accompany mitochondrial dysfunction and enhanced oxidative stress formation that we recently found in fHCM.

Myocardial tissue from the left ventricle (LV) was obtained immediately after euthanasia from 7 cats diagnosed with primary HCM on echocardiography (3 Maine Coon, 2 British Shorthair, 1 Exotic Shorthair, 1 Norwegian Forest cat) and 8 age-matched control cats (5 Maine Coon, 3 Norwegian Forest cats). Ultrastructural examination was performed by the use of transmission electron microscopy.

In HCM cats, marked ultrastructural changes of the cardiomyocytes were observed. The population of subsarcolemmal mitochondria (SSM) was absent in large cellular areas in cats with moderate and severe LV hypertrophy. Flattening of the sarcolemma was a common finding, causing disorganization of the T-tubular system. Interfibrillar mitochondria (IFM) were disorganized but not depleted. Additional changes in cardiomyocytes from cats diagnosed with fHCM included remodeling of sarcomeres, disorganization of myofibrils, convolution of gap junctions, accumulation of intracellular Z-disc material, perinuclear lipofuscin granula and extensive extracellular deposits of collagen.

In healthy mammalian cardiomyocytes, the T-tubular system upholds cellular structure, prevents mitochondrial reticulum formation and provides calcium, oxygen and substrates, necessary for normal functioning muscle. Disorganization of the sarcolemma and T-tubular system may cause the depletion of SSM. Possible mechanisms are atrophy or disruption of the mitochondria or altered fusion-fission dynamics.

Calcium cycling and substrate supply are likely to be compromised by the observed structural changes. We propose this to be related to mitochondrial dysfunction and oxidative stress formation that occurs in fHCM, however a causative relationship remains unknown.

In conclusion, morphological changes of mitochondria and extra-sarcomeric structures are common in fHCM, regardless of breed, genotype and phenotypic disease expression. Moreover, mitochondrial subpopulation-specific changes occur in fHCM with depletion of SSM.

Ultrastructural and functional changes of cardiac muscle mitochondria are considered important molecular mechanisms, responsible for the development and progression of fHCM and may be relevant future treatment targets.

Conflicts of interest: No conflicts of interest reported.

ESVC-O-17

CARDIAC MECHANICS IN DOGS WITH PATENT DUCTUS ARTERIOSUS: A SHORT TERM SPECKLE-TRACKING ECHOCARDIOGRAPHY STUDY. I. Spalla¹, C. Locatelli¹, G. Riscuzzi¹, P. Brambilla¹, C. Bussadori². ¹Università degli Studi di Milano, Milano, Italy, ²Clinica Veterinaria Gran Sasso, Milano, Italy

Patent ductus arteriosus (PDA) is one of the most common congenital cardiac defect in the dog. Ductal patency is associated with pulmonary overcirculation, left ventricular volume overload and can rapidly determine congestive heart failure if untreated. Several devices to close the PDA have been used, with Amplatzer Canine Duct Occluder (ACDO©) being considered the safer device with lowest complication rates.

Echocardiography represents the cornerstone of PDA diagnosis, but its role has been recently expanded to wider field of application: device sizing and intraoperative monitoring, as well as a tool to quantify cardiac morphology and function. Speckle-tracking echocardiography (STE) has been used to evaluate cardiac function in a wide variety of diseases in human and veterinary patients, however no study has evaluated its usefulness in dogs affected by PDA both before and after percutaneous closure of PDA.

The aim of our study was therefore to assess standard M/B-mode derived parameter of cardiac function and STE derived longitudinal, radial and circumferential strain and strain rate before and after PDA closure.

Twenty-five dogs of different breeds, age and weight were prospectively recruited and a complete echocardiographic evaluation was performed before and 24 hours after PDA closure. End diastolic and systolic diameters indexed for body surface area (EDVI/ESVI) both derived by M-mode and B-mode views, allometric scaling derived AlloD and AlloS, Sphericity Index (SI) and pulmonary to systemic flow ratio (Qp/Qs) were assessed both pre and postoperatively. STE derived parameters assessed were longitudinal, radial and circumferential strain and strain rate. A statistically significant difference was found in all standard parameters of cardiac function before and after PDA closure ($p < 0.002$), with a general decrease in values 24 hours postoperatively. STE derived parameters of cardiac function showed a trend toward a decrease back to normal values, which was statistically significant ($p < 0.01$) for circumferential and radial strain and strain rate, while longitudinal strain and strain rate did not reach statistical significance.

Based on our results, no cardiac dysfunction was identified by the use of STE derived parameters both before and after PDA closure, with an increased contractility as identified by higher than normal STE values before PDA closure and a decrease back to normal strain and strain rate values for both circumferential and radial immediately after percutaneous closure. Longitudinal strain persists on higher than normal values, refusing the hypothesis of systolic dysfunction after PDA closure and suggesting a longer reverse remodeling process after PDA closure.

Conflicts of interest: Dr Bussadori receives royalties from ESAOTE (Florence, Italy) related to an European patent (nr 071129712) he developed for Xstrain software. The study was not funded by a research grant.

ESVC-O-18

PREVALENCE AND PROGNOSIS OF CARDIAC CACHEXIA IN DOGS AFFECTED WITH CHRONIC DEGENERATIVE MITRAL VALVE DISEASE. I.B.Y. Yu, H.P. Huang. The Institute of Veterinary Clinical Science, National Taiwan University, Taipei city, Taiwan

Cardiac cachexia which is characterized by progressive weight loss and depletion of lean body mass, is an independent predictor of survival in human patients with congestive heart failure. Chronic degenerative mitral valve disease (CDMD) is one of the most common cardiac diseases in dogs. The aims of this study were to evaluate the prevalence and the effects of cardiac cachexia in survival of dogs with CDMD.

Medical records of 114 client-owned dogs with CDMD were reviewed. The mean age at entry was 11.1 ± 3.2 years; 73 were females, and 41 were males. Data obtained from the records including breed, sex, body weight, age at diagnosis, complete blood counts, biochemical profiles, urinalysis, systemic blood pressure, thoracic radiographs, electrocardiograms, ultrasonography and echocardiographic examinations at initial visit and survival time. Diagnosis of CDMD was based on echocardiographic characteristics and categorized by modified New York Heart Association (NYHA) functional classification. Cardiac cachexia was defined as presence unintentional weight loss ($> 5\%$ within 12 months after diagnosis) together with anorexia and muscle weakness, anemia (red cell count $< 5.5 \times 10^6/\mu\text{L}$, hemoglobin $< 12 \text{ g/dL}$, or both), hypoalbuminemia (plasma albumin $< 3.0 \text{ g/dL}$), and azotemia (blood urea nitrogen $> 26 \text{ g/dL}$, creatinine $> 1.6 \text{ g/dL}$, or both). Dogs with other cardiac disorders and other systemic disorders those would cause anemia and hypoalbuminemia were excluded from this study.

Prevalence of cardiac cachexia, anemia and azotemia was 32.5%, 15.8% and 45.6%, respectively. These conditions were the most prevalent in NYHA class 4, followed by NYHA classes 3 and 2. The prevalence of hypoalbuminemia was not significantly different among classes. The one-year body weight change was found in the NYHA classes 2 (increased $2.7 \pm 10.7\%$), 3 (decreased $1.3 \pm 12.3\%$) and 4 (decreased $6.1 \pm 9.5\%$). The difference between classes 4 and 2 was significant. Results of the Cox proportional hazard model indicated that survival time was significantly positively associated with NYHA functional severity at diagnosis ($P < 0.001$), presence of cardiac cachexia, weight loss, anemia, hypoalbuminemia and azotemia ($P < 0.001$, $P = 0.003$, $P = 0.0033$, $P = 0.003$ and $P = 0.019$, respectively).

The prevalence of cardiac cachexia was common in advanced CDMD dogs, and the parameters of cardiac cachexia, namely weight loss, anemia, hypoalbuminemia and azotemia were strong prognostic factors associated with survival.

Conflicts of interest: No conflicts of interest reported.

ESVC-O-19

EPIDEMIOLOGY OF MITRAL VALVE DISEASE IN MALTESE DOGS IN TAIWAN. T.L. Lu, T.W. Yu, C.H. Chang, Y.W. Hung. Cardiospecial veterinary hospital, Taipei, Taiwan

Mitral valve disease (MVD) is the most common cardiovascular disease in dogs. It's characterized by myxomatous degeneration, which causes mitral valve prolapse (MVP), mitral regurgitation (MR) and a left apical systolic murmur (LASM). MVD affects small breed dogs with a very high prevalence in Cavalier King Charles Spaniels (CKCS). The main goal of this study was to determine the prevalence of LASM, MVP and MR in the Maltese, the most presented breed among dogs with MVD in Taiwan. The correlation between these 3 measurements and the influence of age, gender, reproductive state, and body weight were also investigated. Study results were compared to other MVD prevalence studies in Europe and North-America. 162 client-owned Maltese dogs (75 males and 87 females; body weight 1.35-7.15 kg; age 2-15 yrs) with no signs of heart failure were recorded. The intensity (grade 1-6) of LASM was recorded. Grade of MVP (mild/severe) and MR severity (mild/moderate/severe) were evaluated by echocardiography. Logistic regression

was used to determine the correlation between age and presence of LASM, MVP and MR. A Chi-square test was used to evaluate whether sex and reproductive-status were related to prevalences of LASM, MVP and MR. Spearman's correlation coefficient was used to assess the relationships between age, body weight, LASM intensity, grade of MVP and severity of MR. The prevalence of LASM, MVP and MR were 28.4%, 34% and 41.4%, respectively. All have positive correlation with age ($p = 0.000$). The age at which 50% of the dogs had LASM, MVP and MR was 7.7, 7.5, and 5.6 years, respectively. The LASM intensity, MVP grade and MR severity were all positively correlated to age (all $p = 0.000$) and had no correlation with BW and reproductive status. Females had a significantly higher prevalence of LASM than males (36% vs. 21.8%, $p = 0.046$). Maltese dogs in Taiwan have a very high prevalence and an early development of MVD as compared to other small breed dogs, similar to MVD in CKCS in other countries. Since we only recruited asymptomatic Dogs, this study may underestimate the prevalence of MVD in the whole Maltese population. To our knowledge, this is the first report to document the high prevalence of MVD in Taiwanese Maltese. The Maltese may be a new canine model for genetic, pathology, and natural history studies in MVD.

Conflicts of interest: Boehringer-Ingelheim sponsored the author's accommodation costs for this congress.

ESVC-O-20

CARDIORENAL SYNDROME IN DOGS WITH CHRONIC VALVULAR HEART DISEASE: A RETROSPECTIVE STUDY. E. Martinelli¹, P. Scarpa², C. Quintavalla¹, C. Locatelli², P. Brambilla¹. ¹University of Parma, Parma, Italy, ²University of Milan, Milan, Italy

In human medicine, primary disorders of the heart often result in secondary dysfunction or injury to the kidneys. The coexistence of the two problems in the same patient is referred as cardiorenal syndrome (CRS). Just little information about CRS is available in veterinary medicine.

The aim of this study was to define the prevalence of chronic kidney disease (CKD) complicating chronic valvular heart disease (CVHD) in dogs and to investigate the relationship between class of cardiac insufficiency (ACVIM) and class of renal insufficiency (IRIS).

Medical records of dogs presented at the Cardiology Service of the Department of Veterinary Science and Public Health, University of Milan, between January 2003 and December 2012 were retrospectively evaluated. Dogs with a complete physical examination, thoracic radiographs, a CVHD diagnosis based on echocardiographic examination, and a serum biochemical panel, including assessment of serum creatinine (sCr) and serum urea (BUN), were included in the study. Dogs with other heart disease, neoplasm or systemic diseases were not included in the study. One hundred eighteen dogs of both genders (73 males and 45 females), 5 to 18 years of age (11.64 ± 2.66 years), 3 to 48 kg of bodyweight (11.38 ± 8.84 kg) fulfilled the inclusion criteria. The 20% of males and the 37% of females were neutered. The most represented breeds were mongrel (44%), miniature Poodle (9.32%), York Shire Terrier (8.5%), Shih -Tzu (4.24%), Pinscher (4.24%) and Dachshund (3.38%). Dogs were classified as follow: 0% ACVIM A, 23% ACVIM B1, 9% ACVIM B2, 59% ACVIM C and 9% ACVIM D. While the 73% of the dogs were normoazotemic (sCr < 1,4 mg/dl), 16% were staged in IRIS 2, 11% in IRIS 3 and 0% in IRIS 4. Statistical analysis was performed using JMP 7.0 (SAS Institute Inc.). A p value <0,05 was considered significant. The prevalence of CKD associated with azotemia in dogs affected by CVHD was 27%. There was a statistically significant direct correlation between ACVIM and IRIS class (Pearson test $p = 0.0114$). Unexpectedly, the 58% of dogs receiving drugs for medical management of heart failure (ACVIM class C and D) were normoazotemic. Despite a definite conclusion about the role of CVHD on the induction and/or progression of CKD cannot be drawn from this cross-sectional study, these results suggest that there is a direct correlation between the severity of CKD and CVHD.

Conflicts of interest: No conflicts of interest reported.

ESVC-O-21

BREED DIFFERENCES IN CONCENTRATIONS OF NEUROENDOCRINES AND CORTISOL IN HEALTHY DOGS.

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There is growing evidence of breed differences in concentrations of several blood variables in dogs. The aim of the study was to investigate breed differences in plasma concentrations of components of the renin-angiotensin-aldosterone system (RAAS), endothelin-1 (ET-1) and serum cortisol concentration in healthy dogs.

535 healthy, privately-owned dogs of nine breeds were examined at five centers as part of the European LUPA-project. Absence of cardiovascular or other clinically relevant organ-related or systemic disease was ensured by thorough clinical investigations. Plasma concentrations of ET-1 and aldosterone, renin activity, and serum concentration of cortisol were measured by RIA or ELISA assays.

Overall significant breed differences were found ($P < 0.0001$ for all 4 variables). Bonferroni-corrected pair-wise significant differences between breeds were found in 67% of comparisons for ET-1, 22% for cortisol, 19% for renin and 11% for aldosterone.

For ET-1, the highest median concentration was found in Newfoundlands with values >3 times higher than most other breeds, while renin was highest in Dachshunds, >2 times higher than in Newfoundlands and Boxers, which had the lowest concentrations. Aldosterone was especially low in Belgian Shepherds with median concentration <10 times than the other breeds. Cortisol was highest in Finnish Lapphunds, almost 3 times higher than Boxers with the lowest concentration.

In conclusion, considerable inter-breed variation in concentrations of ET-1, components of RAAS and cortisol was found in healthy dogs. These differences are likely influenced by genetic factors and should be taken into account when designing clinical trials and tests. Breed-specific reference ranges might be necessary.

Conflicts of interest: No conflicts of interest reported.

ESVCN-O-1

THE KINETICS OF WEIGHT LOSS IN OBESE CLIENT-OWNED DOGS. G. Deagle¹, S.L. Holden¹, V. Biourge², S.L. Serisier², A.J. German¹. ¹University of Liverpool, Neston, United Kingdom, ²Royal Canin Research Center, Aimargues, France

Most studies that assess weight management in obese dogs only examine the early stages of weight loss, and this may not properly reflect a complete weight management regime. The aim of the current study was to examine the kinetics of a complete weight management cycle in obese client-owned dogs.

Dogs referred to the Royal Canin Weight Management Clinic, University of Liverpool, for the management of obesity, were eligible for inclusion. All dogs were followed until they had either completed (i.e. reached target weight) or the programme was discontinued. Rate of weight loss, percentage weight lost, and energy were assessed at different time points.

A total of 149 dogs were included, with a range of breeds, ages and sexes represented. Rate of weight loss steadily decreased throughout the weight loss period (d28: $1.2 \pm 0.67\%/wk$; d56: $0.8 \pm 0.6\%/wk$; d84: $0.7 \pm 0.5\%/wk$; d168: $0.5 \pm 0.4\%/wk$; d252: $0.4 \pm 0.3\%/wk$; d672: $0.1 \pm 0.1\%/wk$; $P < 0.001$). The energy intake required to maintain weight loss also progressively decreased ($P < 0.001$). By day 84, mean \pm sd weight loss was $11 \pm 4.9\%$, and compliance was good, but most had not com-

pleted (1% completed, 86% ongoing, 13% discontinued). Thereafter, more dogs completed, but the number of discontinuing also increased (d252: $20 \pm 7.7\%$ weight loss, 32% completed, 41% ongoing, 27% discontinued; d672: $25 \pm 14.6\%$ weight loss; 59% completed, 4% ongoing, 37% stopped).

Initial weight loss is good in obese dogs but, thereafter, steadily worsens. Thus, studies examining only the first few months of weight loss are not fully representative of the entire weight loss process.

Conflicts of interest: The following conflicts of interest apply: The diet used in this study is manufactured by Royal Canin. whilst VB is employed by Royal Canin. VB and SS are employed by Royal Canin. AJG's Readership is funded by Royal Canin.

ESVCN-O-2

METABOLIC AND HORMONAL RESPONSE TO A FEED-CHALLENGE TEST IN LEAN AND OVERWEIGHT DOGS. J. Söder, R. Hagman, K. Höglund, K. Malmjöf, S. Wernersson. Swedish University of Agricultural Sciences, Uppsala, Sweden

Obesity and obesity-related metabolic dysfunctions are increasing in humans as well as in dogs. Obese dogs become affected by chronic diseases at young age, have a decreased quality of life and a shorter life-span. The aim of the study was to describe the metabolic and hormonal response to a feed-challenge test in lean and overweight dogs.

Twenty-eight healthy intact male Labrador retrievers aged 5.2 ± 1.5 years with varying body condition score (BCS, scale 1-9) were included. Twelve dogs were classified as lean (BCS 4-5), ten as slightly overweight (BCS 6) and six as overweight (BCS 6.5-8). An overnight fasting period and blood sample collection was followed by a high fat meal. After food intake, blood samples were collected hourly for four hours. A Glucagon ELISA was validated for use in dogs.

The assigned BCS was supported by positive association with serum leptin concentrations. Postprandial triglyceride concentration was significantly higher in the overweight group. A tendency to higher cholesterol concentration was seen in the overweight group but cholesterol was not affected by food intake. Glucagon concentration rose after food intake and resembled the response seen in humans after a mixed meal. Glucose and insulin concentrations followed the same pattern while free fatty acids had declined one hour after the meal.

In this study, the metabolic and hormonal response to a high fat meal was similar between lean and slightly overweight dogs, whereas the response of overweight dogs differed. Studies on the health significance of postprandial hypertriglyceridemia in dogs are warranted.

Conflicts of interest: The study was financially supported by The Swedish Veterinarian Federation, The Companion Animal Research Foundation, and The Foundation of Thure F. & Karin Forsberg.

ESVCN-O-3

EFFECTIVENESS OF A NEW DIETETIC WEIGHT MANAGEMENT FOOD TO ACHIEVE WEIGHT LOSS IN CLIENT-OWNED OBESE CATS. U. Christmann¹, I. Becvarova², S. Werre¹, H. Meyer². ¹Virginia-Maryland Regional College of Veterinary Medicine, Blacksburg, Virginia, United States of America, ²Hill's Pet Nutrition - Europe, Middle East & Africa, Prague, Czech Republic

Feline weight-loss programs are often hindered by compliance issues and sedentary lifestyle. The purpose of this study was to assess the effectiveness of a new dietetic weight management food (NDWMF)* in achieving weight loss in overweight/obese, client-owned cats. The objectives were 1) to evaluate weight loss parameters in cats fed the NDWMF* and 2) to describe the owner's perception of the cat's quality of life. Overweight/obese, otherwise healthy, client-owned cats (>3/5 body condition score - BCS) were enrolled in the study (n = 132). Initial veterinary evaluation comprised a physical examination, nutritional assess-

ment, determination of ideal body weight (IBW), and development of weight loss feeding plan. Daily energy requirement (DER) for weight loss was calculated as $DER = 0.8 \times (70 \times IBW_{kg}^{0.75})$. Initial and follow-up evaluations (monthly for 6 months) consisted in determination of body weight (BW), BCS, body fat index (BFI), muscle condition score (MCS), and current feeding practices. Quality of life assessment by owners included cat's level of energy, happiness, appetite, begging behavior, flatulence, stool volume, and fecal score. Statistical analysis encompassed scatterplots, regression analysis, summary statistics as appropriate for the type of analyses (continuous or categorical variables, distribution), a mixed model ANOVA was used to assess changes over time (statistical significance at $p < 0.05$). Eighty three percent of the cats (n = 110) lost weight with an average weight loss of 11% (SEM, 1.8%) over 6 months and an average weekly weight loss rate of 0.45% (SEM, 0.03%). A significant decrease in BCS from week 12-24 and in BFI from week 8-24 compared to baseline was observed. MCS did not change. Average duration of weight loss was 134 days (SEM, 4.8 days) with 32 days (SEM, 0.5 days) between visits. Fourteen percent of cats achieved IBW (0.14, CI: 0.08-0.22). Seventy nine percent of cats ate more than the recommended DER (median fed above DER=6%), and the majority of these cats still lost weight. Owners perceived a significant increase in energy and happiness (>week 12) compared to baseline in the cats that lost weight without changes in appetite or begging behavior. No significant changes were seen in scores for flatulence, stool volume, and fecal score. In conclusion, this clinical study showed that feeding the NDWMF* to client-owned, overweight/obese cats resulted in weight loss. Owners reported significant improvements in cat's quality of life without negative side effects.

*Hill's [TRADEMARK] Prescription Diet [TRADEMARK] Feline Metabolic Advanced Weight Solution, dry (caloric distribution: protein=39%, fat=31%, carbohydrate=30%)

Conflicts of interest: Becvarova and Meyer are employees of Hill's Pet Nutrition Manufacturing s.r.o.

The study was sponsored by Hill's Pet Nutrition.

ESVCP-O-1

FELINE ACUTE INTERMITTENT PORPHYRIA IN NEW BRUNSWICK, CANADA: CLINICAL TO MOLECULAR GENETIC CHARACTERIZATION. U. Giger¹, S. Clavero², K. Raj³, E. O'Neil⁴, S. Burton¹, P. Hanna¹, M.E. Haskins³, D.F. Bishop², R.J. Desnick². ¹Section of Medical Genetics, University of Pennsylvania, Philadelphia, United States of America, ²Department of Genetics and Genomic Sciences, Mount Sinai School of Medicine, New York, United States of America, ³University of Pennsylvania, Philadelphia, United States of America, ⁴Atlantic Veterinary College, University of Prince Edward Island, Charlottetown, Canada

Porphyrias are a group of inborn errors of metabolism resulting from accumulation of porphyrins due to deficient activities of specific enzymes in heme biosynthesis. In humans, they are clinically classified as either erythroid with cutaneous involvement or hepatic with acute neurovisceral attacks. Here we describe the clinical, biochemical, and molecular genetic studies in porphyric cats from New Brunswick, Canada.

From 2008 to 2014, three separately identified adult domestic shorthair cats from the city of Saint John in New Brunswick were found to have erythrodontia (brown discolored teeth which fluoresced pink) and pigmenturia. A mild compensated hemolytic disorder with numerous small dark blue irregularly shaped erythrocyte inclusions was noted. There was no evidence of acute life-threatening neurovisceral attacks or cutaneous lesions. Necropsy of one cat revealed massive deposition of porphyrins in all bones and teeth. Urine and EDTA blood samples from one cat were metabolically studied, while molecular genetic studies were performed in all cats either from EDTA blood or a formalinized splenic tissue block.

Urinary δ -aminolevulinic acid, porphobilinogen, uroporphyrin I, and coproporphyrin I concentrations were increased in the cat studied, suggesting an acute intermittent porphyria (AIP). The erythrocytic hydroxymethylbilane synthase (HMBS) activity

in erythrocytes was approximately half normal suggesting a dominant enzymopathy, while the erythrocyte uroporphyrinogen III-synthase activity was normal. Sequencing the feline *HMBS* gene revealed a heterozygous intronic 12 base deletion (c.772-13_-2del) which results in an insertion in the mRNA and would predict a truncated protein.

In conclusion, these three domestic shorthair cats had the same *HMBS* mutation causing an autosomal dominantly inherited AIP. Cats with discolored teeth and normal or mild hemolysis may have either acute intermittent porphyria or congenital erythroid porphyria. Interestingly, seven disease-causing mutations have now been found by us in the *HMBS* gene - more than in any other gene in cats. The biochemical and molecular characterization facilitates clinical screening of affected cats to reach a specific diagnosis.

Supported in part by NIH OD 010939.

Conflicts of interest: Urs Giger and Raj Karthik are also part of the laboratory that offers DNA testing for this mutation.

ESVCP-O-2

HAEMOSTATIC FINDINGS OF PLEURAL FLUID: A CROSS-SECTIONAL STUDY IN 33 DOGS. A. Zoia¹, M. Drigo², C.J. Piek³, P. Simioni⁴, M. Caldin⁵. ¹San Marco Veterinary Clinic, Padua, Italy, ²Sanità Pubblica Veterinaria, Padua University, Padua, Italy, ³Dep. of Clinical Sciences of Companion Animals, Faculty of Veterinary Medicine, Utrecht, The Netherlands, ⁴Department of Cardiologic, Thoracic and Vascular Sciences, University of Padua, Padua, Italy, ⁵Laboratorio d'Analisi Veterinarie San Marco, Padua, Italy

Fibrinogen decreases when coagulation is activated to form fibrin, while FDPs and D-dimers represent the products of fibrinolysis. In humans, activation of coagulation and fibrinolysis develops in all type of ascites and it is also associated with signs of systemic fibrinolysis. These results have led to the suggestion that ascitic fluid is inherently fibrinolytic. Preliminary studies showed similar results also in dogs (JAVMA Nov. 2012, ECVIM proceedings 2013). In addition, in an old experimental study conducted in dogs, inoculation of blood or of a solution containing fibrinogen and thrombin into the pleural cavity resulted in the activation of the coagulation system followed by fibrinolysis. Therefore, the objective of the present study was to determine whether the activation of coagulation and fibrinolysis (i.e. low fibrinogen and elevated FDPs and D-dimer) occurs not only in the ascitic fluid, as already been demonstrated, but also in all type of pleural effusions in dogs. Thirty-three dogs referred to the San Marco Veterinary Clinic with pleural effusion, but without ascites, were studied. Fibrinogen, FDPs, and D-dimer concentrations were measured and then compared in both pleural fluid and venous blood via Wilcoxon signed ranks test. The dog's pleural effusions were categorized based on pathophysiology of fluid formation into 5 dogs with transudate (4 due to increased hydrostatic pressure and 1 due to decreased osmotic pressure), 23 with an exudate (of which 11 due to septic causes), 4 with a haemorrhagic pleural effusion, and 4 with a chylous effusions. The fibrinogen concentration in the pleural effusion (median: 59 mg/dL; range: 59-59) was significantly lower ($p < 0.0001$) than the plasma fibrinogen concentration (median: 419 mg/dL; range: 131-1406). In all dogs, the fibrinogen pleural fluid concentration was lower than the plasma concentration. The FDP concentration in the pleural effusion (median: 151 mg/dL; range: 0.69-151) was significantly ($p < 0.0001$) higher than plasma FDP concentrations (median: 5.55 mg/dL; range: 0.83-108.47). In 1 case, the FDPs pleural fluid concentration was lower than the plasma concentration and in 32 cases the pleural fluid concentration was higher. The D-dimer concentrations were significantly ($p < 0.0001$) higher in the pleural effusion (median: 3.84 $\mu\text{g/mL}$; range: 0.05-9.61) than in the plasma (median: 0.07 $\mu\text{g/mL}$; range: 0.01-7.67). In one case, the D-dimer pleural fluid concentration was lower than the plasma concentration and in 32 cases was higher. These findings support the hypothesis that activation of coagulation followed by fibrinolysis occurs in all type of pleural effusions.

Conflicts of interest: No conflicts of interest reported.

ESVCP-O-3

ASSOCIATION BETWEEN PLEURAL EFFUSIONS AND PRIMARY HYPERFIBRINOGENOLYSIS: A CASE CONTROL STUDY IN 99 DOGS. A. Zoia¹, M. Drigo², P. Simioni³, M. Caldin⁴, C.J. Piek⁵. ¹San Marco Veterinary Clinic, Padua, Italy, ²Sanità Pubblica Veterinaria, Padua University, Padua, Italy, ³Department of Cardiologic, Thoracic and Vascular Sciences, University of Padua, Padua, Italy, ⁴Laboratorio d'Analisi Veterinarie San Marco, Padua, Italy, ⁵Dep. of Clinical Sciences of Companion Animals, Faculty of Veterinary Medicine, Utrecht, The Netherlands

During primary hyperfibrinolysis (PHF), FDPs production is increased but production of D-dimer is not. Therefore, elevated FDPs and normal D-dimer are considered an indicator of PHF. In humans and dogs, activation of coagulation and fibrinolysis develops in all type of ascites and it is associated with systemic PHF, suggesting that ascites is inherently fibrinolytic. Preliminary data have shown that activation of coagulation followed by fibrinolysis occurs also in all type of pleural effusions (PE). The objective of this study was to determine if systemic PHF occurs also in dogs with PE. Thirty-three dogs referred to the San Marco Veterinary Clinic with PE, but without ascites, were studied (group 1). From the electronic data-base of the clinic dogs for inclusion in control groups 2 (healthy dogs) and 3 (sick dogs without PE or ascites) were randomly selected and individually matched to group 1 dogs for age, sex, and breed. Fibrinogen, FDPs, D-dimers, C-reactive protein (CRP), fibrinogen/CRP ratio, and prevalence of PHF (i.e., dogs with elevated plasma FDPs and normal D-dimer) were determined. Differences between the 3 groups were analyzed using ANOVA (fibrinogen), Chi-Square (FDPs and prevalence of PHF) and Kruskal-Wallis test (CRP, fibrinogen/CRP ratio, and D-dimer). Post-test analysis were performed by Tamhane and Mann-Whitney test. Fibrinogen concentration in group 1 was significantly increased compared to group 2 ($p < 0.0001$), but not compared to group 3 ($p = 0.504$). FDPs concentration in group 1 was significantly increased compared to groups 2 ($p < 0.0001$), but not compared to group 3 ($p = 0.148$). D-dimers concentration in group 1 was significantly increased compared to group 2 ($p < 0.0001$), but not compared to group 3 ($p = 0.964$). CRP was significantly increased in group 1 compared to group 2 and 3 ($p < 0.0001$ for both comparison). Fibrinogen/CRP ratio was significantly decreased in group 1 compared to group 2 and 3 ($p < 0.0001$ for both comparison). Prevalence of PHF was significantly higher in group 1 compared to groups 2 ($p = 0.004$), but not compared to group 3 ($p = 0.186$). These results support the hypothesis that PHF occurs significantly more often in dogs with PE compared to healthy dogs. Despite there was a trend of increased PHF also in dogs with PE compared to sick dogs, this difference did not reach significance. Nevertheless, the decreased in fibrinogen/CRP ratio in group 1 compared to group 3, in the face of a similar D-dimer concentration, would suggest that PHF is also more prevalent in dogs with PE compared to sick control dogs.

Conflicts of interest: No conflicts of interest reported.

ESVCP-O-4

INFLAMMATORY CYTOKINES AND C-REACTIVE PROTEIN IN CANINE SYSTEMIC INFLAMMATORY RESPONSE SYNDROME. K. Gommeren¹, I. Desmas¹, A. Garcia¹, N. Bauer², A. Moritz², J. Roth², D. Peeters¹. ¹Liège University, Liège, Belgium, ²Justus-Liebig-University Giessen, Giessen, Germany

The systemic inflammatory response syndrome (SIRS) refers to clinical signs of systemic inflammation in response to (non-) infectious insults. Current diagnosis of SIRS is based on clinical and basic laboratory data and is a sensitive screening to identify patients at risk. C-reactive protein (CRP) is a major canine acute phase protein with concentrations related to disease severity and underlying cause. CRP rises in response to proinflammatory cytokines, mainly interleukin (IL)-6 and tumor necrosis factor (TNF)- α , which are considered the main triggers of SIRS.

We therefore evaluated CRP, IL-6 and TNF- α kinetics in canine emergency SIRS patients hypothesizing that CRP is (1) increased in dogs with a clinical SIRS-diagnosis, (2) correlated with IL-6 and TNF- α concentrations, (3) influenced by the underlying etiology, and (4) a prognostic marker.

Canine emergencies with clinically diagnosed SIRS were prospectively included. Serum and plasma were immediately stored at -80°C after sampling at presentation, after 6 (T6), 12 (T12), 24 (T24) and 72 (T72) hours, and at a control visit (T1 m) over one month after discharge. Serum CRP was measured with a canine-specific immunoturbidimetric CRP assay. Plasma IL-6 and TNF- α were measured using a bioassay measuring biologically active cytokine concentrations. Disease categories were infection (I), neoplasia (N), trauma (T), gastric-dilation and volvulus (GDV), other gastrointestinal (GI), renal (R) and miscellaneous (M) diseases. Statistical analysis was performed with SAS. Concentrations of inflammatory cytokines were expressed logarithmically, with univariate analysis confirming normal distribution. A correlation procedure, mixed procedure on a linear model and a logistic procedure were performed (p-value < 0.05).

Sixty seven dogs (I = 12, N = 13, T = 6, GDV=11, GI=4, R = 3, M = 18) were included. Forty-three patients survived (seven died, seventeen were euthanized). Twenty patients had a control visit. CRP was elevated in 71.2% of dogs at presentation, and only remained within reference range (0-14.9 mg/L) throughout hospitalization in four dogs (6.0%). CRP concentrations were significantly higher from T0 (92.7 ± 113.7 mg/L) to T24 (95.2 ± 90.2 mg/L) decreasing at T72 (71.1 ± 76.6 mg/L), and returning within reference range at T1 m (2.4 ± 4.5 mg/L) in all but one dog (18.2 mg/L). CRP was significantly correlated with logarithmical concentrations of IL-6 and TNF- α , however, these did not change significantly over time. None of the evaluated parameters was associated with disease category, nor outcome.

CRP appears useful to diagnose SIRS in emergency patients, and tends to decrease during hospitalization. However, CRP, neither IL-6 nor TNF- α concentrations appear useful to predict the underlying disease and outcome in SIRS patients.

Conflicts of interest: No conflicts of interest reported.

ESVCP-O-5

BIOLOGICAL VARIATION OF CANINE CALPROTECTIN CONCENTRATIONS IN SERUM. R.M. Heilmann¹, C.G. Ruau², N. Grützner¹, J.S. Suchodolski¹, J.M. Steiner¹. ¹College of Veterinary Medicine, Texas A&M University, College station, United States of America, ²College of Veterinary Medicine, Oregon State University, Corvallis, United States of America

Calprotectin (S100A8/A9 complex) belongs to the S100/calgranulin family, and is primarily released from activated neutrophils and macrophages. Serum calprotectin concentrations (CP) were shown to be increased in dogs with inflammatory diseases such as inflammatory bowel disease, pancreatitis, systemic inflammatory response syndrome, and sepsis. Canine CP thus appears to be a biomarker of inflammation. Considerable day-to-day variation of fecal canine CP was found in both healthy dogs and dogs with chronic gastrointestinal disease. However, the biological variation of canine CP in serum has not been reported. The aim of this study was to determine the biological variation of serum canine CP and its minimum critical difference (MCD).

Eleven healthy dogs were used for this study. Biological variation of serum canine CP was evaluated over a 2.6-months period. Tests for outliers were carried out at 3 levels (within-run analytical variance, intra-, and inter-individual variation). A nested analysis of variance (ANOVA) model was used to calculate analytical (CV_A), intra-individual (CV_I), inter-individual (CV_G), and total variation (CV_T), and to determine the index of individuality (II), index of heterogeneity (IH), and MCD.

A total of 14 serial specimens were collected from 6 dogs, 13 serial samples from 3 dogs, and 12 serial samples from 2 dogs. Four within-subject outliers were detected and excluded from further analysis, yielding a total of 147 serum samples and slightly right-skewed data. No outlying observations (Cochrane test) or outliers among mean concentrations of subjects (Reed's criterion) were detected. CV_A was calculated as 3.0%, CV_I as 29.9%, and

CV_G as 33.2%, resulting in a CV_T of 66.1%. Index of Individuality (II) was determined to be 0.905 and IH was 4.939, yielding a one-sided MCD of 6.4 mg/L. The analytical goal of $CV_A \leq \frac{1}{2} \times CV_I$ was satisfied.

Although serum canine CP remained within a relatively narrow concentration range in healthy dogs, moderate individuality was detected. Moderate changes in serum canine CP (6.4 mg/L) between sequential measurements are needed to be considered clinically relevant, and using a population-based reference interval may or may not be appropriate for serum canine CP. Using the MCD with the previously determined median canine CP concentration (4.9 mg/L) for the reference sample group yielded a serum canine CP concentration close to the upper limit of the previously established reference interval (11.9 mg/L), showing that the reference interval for serum cCP (0.9-11.9 mg/L) is within reasonable limits.

Conflicts of interest: The assay used in the study was developed at the GI Laboratory, Texas A&M University. Most authors also work at the GI Laboratory, Texas A&M University.

ESVCP-O-6

CORRELATION OF ACUTE PHASE PROTEINS WITH CLINICAL AND LABORATORY PARAMETERS, AND CLINICAL STAGING IN 80 DOGS WITH LEISHMANIASIS CAUSED BY L. INFANTUM/CHAGASI. D. Alatza¹, C. Koutinas¹, T.A. Petanides², J.J. Ceron³, S. Martinez-Subiela³, Z. Polizopoulou¹. ¹Faculty of Veterinary Medicine, Aristotle University of Thessaloniki, Thessaloniki, Greece, ²Clinic of Medicine, Faculty of Veterinary Medicine, University of Thessaly, Karditsa, Greece, ³Interlab-UMU, Faculty of Veterinary Medicine, University of Murcia, Murcia, Spain

Canine leishmaniasis (CanL) is a multisystemic disease that is endemic in the Mediterranean region. In the past, concentrations of acute phase proteins (APPs), and specifically C-reactive protein (CRP), haptoglobin (Hp), ceruloplasmin (Cp), serum amyloid A (SAA) and albumin (Alb), have been reported to change in dogs with leishmaniasis, and revert to normal after successful treatment, highlighting the intrinsic inflammatory reaction of the host to the parasite.

Since the spectrum of clinical and laboratory derangements is broad, it is possible that APPs are increased specifically because of certain clinicopathological syndromes associated with CanL.

A total of 80 dogs with CanL, diagnosed on the basis of cytological amastigote identification and IFAT serology, were retrospectively included in the study. In all of them, CRP, SAA, Hp and Alb were measured at Interlab-UMU, Murcia, Spain, in aliquots of serum, which were stored in -22°C for 4 - 8 years (median: 4 years). Results for each of the APPs were correlated to laboratory and clinical parameters (n:80), clinical and parasitological scoring (n:40), *Ehrlichia* and *Leishmania* serology (n:40), and clinical staging according to LeishVet (n:80), using an array of linear and ordinal regression models, as well as one-way ANOVA, t-test and Fisher's LSD test.

CRP and Alb were by far the APPs most frequently correlated with clinical and laboratory abnormalities such as nutritional status, lethargy and skin ulcers ($P < 0.05$), as well as urinary protein to creatinine ratio (UPC), total serum protein, and urine specific gravity ($P < 0.02$). There were limited associations between Hp, Cp, SAA and clinicopathological parameters. A minor linear relationship was observed between CRP and clinical scoring. CRP and Alb were also correlated with parasitological scoring in bone marrow, but not lymph node cytology ($P < 0.04$). Dogs with *Ehrlichia* titers had higher CRP, Cp and lower Alb concentrations. Finally, CRP concentrations were higher in later compared to earlier stages of the infection, as defined by the Leishvet criteria.

The inflammatory component to *Leishmania* infection doesn't seem to be exemplified by the reaction of a particular tissue, with the possible exception of glomerulonephritis. The magnitude of increase in CRP and decrease in albumin is correlated with clinical staging and bone marrow parasitological scoring.

Conflicts of interest: No conflicts of interest reported.

ESVCP-O-7
DEVELOPMENT OF A REMOTE PLATELET P-SELECTIN TEST FOR DELAYED MEASUREMENT OF PLATELET FUNCTION IN DOGS AND CATS. M. Dunning¹, J. May², A. Struck², S. Fox², ¹University of Nottingham, Leicestershire, United Kingdom, ²Cardiovascular Medicine, School of Medicine, University of Nottingham, Nottingham, United Kingdom

The consequences of abnormal platelet function in dogs and cats can be devastating and the use of anti-thrombotic therapy to prevent thrombotic events is increasingly common. The ability to measure platelet function and the efficacy of anti-thrombotic therapy is difficult due to limited availability of equipment and inability to delay platelet function analysis.

The aim of this study was to adapt and validate test procedures and protocols previously developed for humans for use in dogs and cats.

Residual samples of citrate anticoagulated blood were used from dogs and cats presented to a specialist referral centre for various reasons unrelated to clotting abnormalities. Initially the blood was stimulated using specific combinations of either arachidonic acid/epinephrine (AA/EPI) or ADP/U46619, designed to assess the effects of the anti-thrombotic agents aspirin and clopidogrel respectively. After 5 minutes stimulation, the blood samples were fixed using a patented platelet fixative solution developed for human platelets, which allows the delayed analysis of P-selectin an established marker of platelet activation.

All analysis was performed by flow cytometry. In order to do this, specific antibodies were selected for the recognition of both canine and feline platelets. CD61 was used as a platelet identifier antibody while appropriate CD62P (P-selectin) antibodies for each species were chosen. Fixed samples were repeatedly analysed at time points between 0 to 35 days following fixation to establish the stability of the fixed samples. Thirteen dogs and three cats were analysed.

High P-selectin expression was detected following stimulation with AA/EPI and ADP/U46619 in both dogs and cats following fixation. This was significantly different to unstimulated blood ($p < 0.0001$). There was no significant difference in detectable P-selectin expression following storage of the fixed samples at any time-point up to 35 days. This confirmed the fixative was suitable as a preservative of canine and feline platelets.

A limited number of dogs were evaluated whilst receiving anti-thrombotic medication. There was a significant difference in the activation of platelets in the dogs treated with either aspirin ($p < 0.03$) or clopidogrel ($p < 0.004$) compared with untreated dogs following stimulation with AA/EPI (aspirin group) or ADP/U46619 (clopidogrel group).

Our results show that fixation and delayed analysis of platelet function in dogs and cats is possible for up to 35 days. This demonstrates an exciting opportunity to analyse platelet function remotely and to determine the efficacy of thromboprophylaxis in animals presenting to clinics that do not have on-site platelet analysers.

Conflicts of interest: No conflicts of interest reported.

ESVE-O-1
BEHAVIOUR IN DOGS BEFORE AND AFTER TREATMENT OF SPONTANEOUS HYPOTHYROIDISM WITH L-THYROXINE. A. Hrovat Vernik¹, T.de Keuster¹, H.S. Kooistra², L. Duchateau¹, S. Daminet¹. ¹Ghent University, Merelbeke, Belgium, ²Utrecht University, Utrecht, The Netherlands

Several authors consider thyroid hormone supplementation as a valid initial treatment option for dogs with aggression related problems. Indeed, mood and behaviour modulating properties of thyroid hormones may, in part, be mediated through the interaction of thyroid hormones with neurohormones such as serotonin and prolactin. At present, prospective trials evaluating neurohormonal status or behaviour in hypothyroid dogs before and after thyroid supplementation are lacking. Therefore, the aims of this study were to assess behaviour and measure serum serotonin and prolactin concentrations in dogs with spontaneous hypothyroidism before and after treatment. Twenty three client-owned dogs diagnosed with spontaneous primary hypothyroidism were

prospectively included in our study. After diagnosis all dogs were treated with levothyroxine (10 micrograms/kg BID). Behaviour of dogs was screened at initial presentation, at 6 weeks and 6 months after initiation of therapy. Owners had to fill in a hard copy of the standardized Canine Behavioural Assessment and Research Questionnaire (C-BARQ) consisting of 101 scored questions evaluating seven behavioural categories. The average score on all questions was calculated for each dog at each of the three time periods and a paired t-test was used for comparison. Serum serotonin and prolactin concentrations were evaluated at each time period using a commercially validated ELISA kit and heterologous RIA, respectively. Results of the C-BARQ after six weeks of thyroid hormone supplementation when compared with the time zero demonstrated a significant increase ($P < 0.05$) in excitability, activity and aggression, which most likely became unmasked owing to improved overall activity of dogs. Conversely, at six months period when compared with the time zero no significant changes in any of the behavioural symptoms were observed. Serum serotonin was measured in 18/23 dogs colorimetrically at 450 nm. At time zero, 6 weeks and 6 months serum serotonin was 0.135 (range, 0.075-0.337), 0.141 (range, 0.071-0.392) and 0.149 (range, 0.086-0.378). No significant difference was noted between 6 week and 6 month period comparing to time zero ($P = 0.84$ and $P = 0.37$). Serum prolactin concentration measured in 22/23 dogs at time zero, 6 weeks and 6 months was 3.35 ng/ml (range, 1.4-6.36), 3.57 ng/ml (range, 1.87-7.39) and 3.92 ng/ml (range, 2.01-12.92) and did not differ significantly in either time period when compared with time zero ($P = 0.99$ and $P = 0.52$). Altogether, results of this study failed to demonstrate a significant role of thyroid supplementation on the majority of evaluated behavioural symptoms as well as neurohormonal status of hypothyroid dogs during 6 months of therapy.

Conflicts of interest: No conflicts of interest reported.

ESVE-O-2
OVERT OR SUBCLINICAL IATROGENIC HYPOTHYROIDISM IN CATS: CLINICAL, LABORATORY, AND THYROID SCINTIGRAPHIC FINDINGS IN 35 CASES. M.E. Peterson, J.N. Guterl. Animal Endocrine Clinic, New York, United States of America

Iatrogenic hypothyroidism is a recognized complication of radioiodine treatment of hyperthyroidism in cats, but no prospective studies of the prevalence, clinical features, routine laboratory findings, or results of thyroid function tests have been reported in a series of hypothyroid cats. In this study, we describe the features of hypothyroidism in 35 cats treated with radioiodine over a 15-month period (October 2012-March 2014). During this same period, we treated ≈ 500 hyperthyroid cats with radioiodine, providing a prevalence rate of 7%.

Hypothyroidism was diagnosed 41-814 days (median, 120 days) after ¹³¹I treatment, with doses ranging from 65-1000 Mbq (median, 100 Mbq; median pretreatment T4, 142 nmol/L). The 35 hypothyroid cats ranged in age from 9-19 years (median, 13 years). All were DSH/DLH; 28 (80%) were female and 7 were males ($P = 0.0004$). Clinical signs in these 35 cats included overweight/obesity in 6 (17%), lethargy/dullness in 6 (17%), poor appetite in 3 (8.6%), and polyuria/polydipsia in 9 (26%). Abnormalities on physical examination included dermatologic signs (dry coat, seborrhea, matting) in 5 (14%) and bradycardia (< 150 bpm) in 2. Twenty-two cats (63%) had no noticeable clinical features of hypothyroidism. Routine laboratory abnormalities included hypercholesterolemia (> 6 mmol/L) in 7 (20%) and new or worsening azotemia (> 140 μ mol/L) in 26 (74%) and 4 (11%) cats, respectively.

Median serum concentrations of total T4 (11.6 nmol/L; reference interval [RI], 10-50 nmol/L), T3 (0.6 nmol/L; RI, 0.5-0.8 nmol/L), and FT4 (15 pmol/L; RI, 10-50 pmol/L) were all in the low end of the RI. Normal RI values for T4 and FT4 were maintained in 22 (63%) and 30 (86%) of the cats, respectively. Serum cTSH values were high in all cats (median, 3.3 ng/dl; range, 0.47-12.0 ng/dl; RI, 0.03-0.30 ng/ml). Thyroid scintigraphy showed less-than-normal amounts of residual tissue, as well as low values for thyroid-to-salivary ratio and %-uptake of per-technetate, in 30 (86%). Of those 5 cats with normal scintiscans, serum cTSH decreased into the RI without treatment when retested 1-6 months later.

In conclusion, this study confirms that ^{131}I -induced hypothyroidism is not uncommon, with an apparent female sex predilection. Serum T4 and FT4 remain normal in most cats, but high serum cTSH values and thyroid scintigraphy aid in diagnosis. Unless cats have overt, long-standing hypothyroidism, most cats with subclinical disease are relatively asymptomatic, other than worsening azotemia. Subclinical hypothyroidism will be transient in some cats, with normalization of cTSH values within a few months.

Conflicts of interest: No conflicts of interest reported.

ESVE-O-3

IATROGENIC FELINE HYPOTHYROIDISM: CHALLENGES AND COMPLEXITIES OF THYROID HORMONE REPLACEMENT IN CATS. M.E. Peterson, J.N. Guterl. Animal Endocrine Clinic, New York, United States of America

Iatrogenic hypothyroidism is a recognized complication of radioiodine treatment for hyperthyroidism in cats. At our clinic where we use a variable ^{131}I dosing protocol (based on tumor volume and severity of hyperthyroidism), the prevalence of overt or subclinical hypothyroidism is at least 7%. During the 15-month period from October 2012 to March 2014, we treated 19 cats with iatrogenic hypothyroidism, which had developed 41-323 days (median, 119 days) after treatment with radioiodine (median dose, 100 Mbq). These cats ranged in age from 9-19 years (median, 14 years); all were DSH/DLH; 14 (74%) were female and 5 were males. New or worsening azotemia ($>140 \mu\text{mol/L}$) was documented in 16 (84%) and 2 (10.5%) cats, respectively. Diagnosis of hypothyroidism was based on the following: 1) low to low-normal serum concentrations of T4, FT4, and T3; 2) high serum TSH concentration ($>0.5 \text{ ng/dl}$); and 3) less-than-normal amounts of residual tissue on thyroid scintigraphy. All cats were given thyroid hormone replacement as a liquid L-T4 preparation (Leventa; Merck Animal Health). Cats were monitored at 1-3 month intervals by repeating serum T4 and TSH concentrations 3-4 hours after the morning L-T4 dose.

Ten of the 19 cats were started on a once-daily L-T4 regimen (100 μg); of these, only 2 (20%) had suppression of high serum TSH values into the reference interval (RI). Of the 8 cats that had persistently high TSH values, 5 were switched to twice-daily administration (75-100 μg , BID), which successfully lowered high TSH concentrations in 4 cats. The remaining 9 cats were started on twice daily L-T4 (75 μg , BID); of these, normalization of TSH occurred in 5 cats. Overall, L-T4 treatment was successful in normalizing TSH concentrations in 11 (58%) cats, 2 with once-daily and 9 with twice-daily dosing. Peak serum T4 concentrations of $\geq 30 \text{ nmol/L}$ were needed in most cats to normalize TSH values. Higher serum T4 and lower TSH concentrations were achieved when L-T4 was administered on an empty stomach rather than given with food. A significant decrease ($P < 0.003$) in serum creatinine occurred after treatment with L-T4.

In conclusion, our results indicate that twice-daily administration of L-T4 is needed in most cats with iatrogenic hypothyroidism to normalize high serum TSH concentrations. Many cats appear to absorb L-T4 rather poorly, which can be enhanced by giving the drug on an empty stomach. The azotemia that commonly develops in cats with hypothyroidism improved or stabilized with adequate L-T4 supplementation.

Conflicts of interest: No conflicts of interest reported.

ESVE-O-4

CONGENITAL HYPOTHYROIDISM WITH GOITER IN CATS DUE TO A TPO MUTATION. U. Giger¹, K. Raj¹, C.V. Murrow², A. Traas², A.M. Erat², M. van Hoesen², H. Mazrier², M.E. Haskins². ¹Section of Medical Genetics, University of Pennsylvania, Philadelphia, United States of America, ²University of Pennsylvania, Philadelphia, United States of America

Congenital hypothyroidism (CH) has been reported in many species; the hereditary forms can be divided into thyroid dysmorphogenesis and dysmorphogenesis. While thyroid hypoplasia

has been described in dogs and cats, the molecular basis remains unknown. In contrast few breeds of dogs with goiterous CH were found to have deficient thyroid peroxidase (TPO) activity. The purpose of our study was to characterize a family of domestic shorthair cats with goiterous CH and disease-causing TPO gene mutations.

Clinical features included dwarfism and dullness, known as cretinism and seen with CH in all species, but also constipation and megacolon which are unique to cats with CH. Pedigree analysis documented an autosomal recessive mode of inheritance. Affected kittens developed a goiter and had low serum thyroxine (T4) and triiodothyronine (T3) when compared to controls, but high thyroid stimulating (TSH) hormone levels indicating thyroid dysmorphogenesis. Oral thyroid supplementation corrected the progression of clinical signs and prevented further constipation and reversed the megacolon.

The TPO enzyme activity was extremely low in hypothyroid cats when compared to that of normal cats. Genomic DNA and cDNA from affected, carrier, and normal cats were extracted and sequenced based upon primers developed from the feline genome database. A homozygous missense point mutation (c.1333G>A) in TPO, which results in an amino acid change (p.Ala445Thr), was discovered in affected cats and the mutant allele segregated within the family with goiterous CH.

This is the first report of a TPO deficiency in cats. Other unrelated domestic shorthair cats with goiterous CH did not have this same TPO mutation. The prevalence of this TPO mutation in the domestic cat population seems low, but CH is likely underreported in cats.

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ESVE-O-5

ASSESSMENT OF A GLUCAGON-LIKE PEPTIDE-1 ANALOGUE (EXENATIDE EXTENDED-RELEASE) IN CATS WITH NEWLY DIAGNOSED DIABETES MELLITUS. A.R. Riederer¹, F. Fracassi², E. Salesov¹, I. Padrut¹, T. Lutz¹, T. Stöckle², E. Zini¹, C. Reusch¹, K. Macha¹. ¹Clinic for Small Animal Internal Medicine, Vetsuisse Faculty, University Zurich, Zurich, Switzerland, ²Department of Veterinary Medical Sciences, University of Bologna, Ozzano dell'emilia, Italy, ³Institute of Veterinary Physiology, Vetsuisse Faculty, University of Zurich, Zurich, Switzerland

Glucagon-like peptide-1 (GLP-1) is a gastrointestinal hormone released in response to food intake that increases insulin secretion, inhibits glucagon secretion, slows gastric emptying and induces satiation. It is also assumed to stimulate beta-cell proliferation. GLP-1 agonists are successfully used in humans with type 2 diabetes mellitus usually either in combination with insulin or other anti-diabetic drugs. In healthy cats twice daily (exenatide) as well as once weekly (exenatide extended-release (ER)) application of GLP-1 agonists induced pronounced insulin secretion. Benefits of exenatide ER are the regimen of once weekly injection and less side effects. The objective of the study was to assess whether administration of exenatide ER in addition to standard treatment leads to improved glycemic control and higher remission rates in cats with newly diagnosed diabetes.

The study was designed as a prospective, placebo-controlled clinical trial. Cats were randomly assigned to two groups receiving exenatide ER (group 1: Bydureon[®], 200 mg/kg, q7d, SC) or 0.9% saline solution (group 2: q7d, SC). Both groups additionally received insulin glargine (Lantus[®], initial dose: $\leq 4 \text{ kg}$: 1.0 IU, q12 h; $> 4 \text{ kg}$ 1.5-2.0 IU, q12 h) and diet (Purina DM[®]). Exenatide ER was applied over 16 weeks or, in case of remission, for 4 additional weeks after cessation of insulin application. Cats were rechecked 1, 3, 6, 10 and 16 weeks after starting therapy. Remission of diabetes was defined as absence of clinical signs of diabetes and normal blood glucose and fructosamine concentrations for at least 4 weeks after discontinuing insulin injections.

So far 17 cats have completed the study. Mild and transient side effects in group 1 (n = 9) were reduced appetite (n = 6), nausea (n = 2), vomitus (n = 4), tiredness (n = 2) and hiding in dark

spots of the house (n = 2). In group 1 remission was achieved in 4/9 (44%) cats and good metabolic control in 4/5 (80%) non-remission cats. In group 2 remission was achieved in 2/8 (25%) cats and good metabolic control in 4/6 (66%) non-remission cats. Median insulin dose given during the study period was 0.47 IU/kg/day in group 1 and 0.56 IU/kg/day in group 2.

The preliminary results suggest that exenatide ER can be used safely in diabetic cats. A tendency for higher remission rate, better metabolic control and lower insulin requirement was seen when exenatide ER was added to the standard treatment regimen. Further cases need to be evaluated to verify the potential beneficial role of exenatide ER.

Conflicts of interest: No conflicts of interest reported.

ESVE-O-6

LASER MICRODISSECTION OF PANCREATIC ISLETS ALLOWS FOR QUANTITATIVE REAL-TIME PCR DETECTION OF ISLET-SPECIFIC GENES IN HEALTHY AND DIABETIC CATS. M.T. Öhlund¹, P. Franzén², G. Andersson³, B. Ström Holst¹, J. Lau¹. ¹Dept. of clinical sciences, Swedish university of agricultural sciences, Uppsala, Sweden, ²Dept. of Medical Cell Biology, Uppsala University, Uppsala, Sweden, ³Dept. of animal breeding and genetics, Swedish university of agricultural sciences, Uppsala, Sweden

Feline diabetes mellitus shares many similarities with human type 2 diabetes mellitus (T2DM), including clinical, physiological and pathological features of the disease. Domestic cats spontaneously develop diabetes associated with insulin resistance in their middle age or later, with residual but declining insulin secretion. Humans and cats share the same environment and risk factors for diabetes, such as obesity and physical inactivity. Moreover, amyloid formation and loss of beta cells are found in the diabetic cat pancreas, as in humans. Subsequently, studying the molecular mechanisms in the failing beta cells may contribute to a better understanding of the pathophysiology of T2DM in both cats and humans.

The aim of the present study was to develop a method to study mRNA expression of islet-specific genes in healthy and diabetic cats. Previous attempts in isolating feline islets with different collagenase-based protocols have led to damaged islets or islets coated with exocrine acinar cells, which either way compromise the results obtained from gene expression studies.

By using the laser microdissection technique, we were able to sample islets that were not contaminated with exocrine tissue, from both healthy and diabetic cats. High RNA quality was confirmed with gel electrophoresis. By quantitative real-time PCR (qRT-PCR), mRNA levels of the islet-specific genes *insulin*, *PDX-1*, *IAPP*, *CHGA* and *IA-2* were detected in both healthy and diabetic cats. We used *actin b*, *GAPDH* and *RPS7* as internal reference genes for normalizations of our qRT-PCR data.

The laser microdissection technique allows studies of islets without contamination of acinar cells, as shown in this study, and is of great advantage since it is difficult to get pure feline islets from collagenase-based isolation. Differences in gene expression in healthy and diabetic cats may reveal underlying mechanisms for beta cell dysfunction and decreased beta cell mass in human and feline type 2 diabetes.

Conflicts of interest: The study was financially supported by the Swedish Juvenile Diabetes Foundation, the Fredrik and Ingrid Thuring Foundation, the Magnus Bergvall Foundation, the Lars Hierta Memorial Foundation, and the Foundation for Research, Agria Insurance Company.

ESVE-O-7

THE FELINE AIP GENE: THE KEY TO HYPERSOMATOTROPISM TUMORIGENESIS? C. Scudder, S.J.M. Niessen, B. Catchpole, R.C. Fowkes, D.B. Church, Y. Forcada. Royal Veterinary College, North Mymms, Hertfordshire, United Kingdom

Feline acromegaly is an increasingly recognised endocrinopathy among diabetic cats, caused by chronic excessive growth

hormone secretion by a functional somatotrophinoma in the pars distalis of the anterior pituitary gland. The majority of human somatotrophinomas are sporadic, however up to 20% of familial isolated pituitary adenomas are caused by germline mutations of the aryl-hydrocarbon-receptor interacting protein (AIP). Feline acromegaly has phenotypic and biochemical similarities to human familial acromegaly with AIP mutations, such as male predominance, somatotroph macroadenoma and resistance to octreotide therapy.

The objective of this study was to identify the feline AIP gene, identify single nucleotide polymorphisms (SNPs) within this gene and compare any SNPs with reported human AIP SNPs. Stored pituitary tissue from an acromegalic cat was used to create feline AIP cDNA using feline specific AIP primers. Stored EDTA blood from 10 acromegalic cats (diagnosis of insulin resistant diabetes mellitus, serum IGF-1 > 1000 ng/ml and pituitary mass > 4 mm identified using pituitary computed tomography or necropsy) and 10 control cats (no history of diabetes mellitus and greater than 15 years of age) were selected, DNA extracted and genotyped using PCR, agarose gel electrophoresis and Sanger sequencing.

The feline AIP gene was identified, encoding a 330 amino acid protein with 98% homology to the human AIP protein. A BLAST search revealed this gene contained 6 exons and exon specific primers were created to enable sequencing. A single non-conservative SNP was identified in exon 1 (AIP:c.9G>T), encoding for an amino acid change from aspartic acid to glutamic acid in 2/10 acromegalic patients and 0/10 control cats. Two additional conservative SNPs were also identified (AIP:c.826T>C and AIP:c.481T>C).

Exon 1 encodes for a region of the AIP protein considered essential for AIP-AIP receptor interaction. Although 70 different human AIP mutations have been identified to date, a human AIP:c.9G>T mutation has not yet been identified. The AIP N-terminal is required for the stability of the AIP protein-AIP-receptor complex, and essential for the regulation of translocation into the nucleus, where it binds to aryl hydrocarbon receptor nuclear translocator leading to activation of genes thought to act as tumor suppressors. Loss of normal AIP activity is thought to promote somatotrophinoma development. It is therefore possible that the detected AIP:c.9G>T mutation predisposed to somatotrophinoma tumorigenesis in the two affected patients, and a study containing a larger number of cases is indicated.

Conflicts of interest: No conflicts of interest reported.

ESVE-O-8

EXPERIENCES OF A NEWLY ESTABLISHED HYPOPHYSECTOMY CLINIC FOR TREATMENT OF FELINE HYPERSOMATOTROPISM. P.J. Kenny, C. Scudder, S.V. Keyte, J.W. Swann, R.C. Fowkes, D.B. Church, Y. Forcada, S.J.M. Niessen. Royal Veterinary College, London, United Kingdom

Hypersomatotrophism (HS) is an important cause of feline diabetes mellitus (DM). In humans surgical removal of the somatotrophinoma is generally recommended, though hypophysectomy programs have suffered from significant initial morbidity and mortality given a documented steep learning curve in newly established programs. Hypophysectomy as treatment for feline HS has thus far only been described in a handful of cases, all having been treated by one single experienced hypophysectomy team. This study's aim was to evaluate the learning curve of a *de novo* established hypophysectomy program, through analysis of peri- and post-operative morbidity and mortality, and endocrine outcomes in the first cohort of cats with HS treated.

From 2012 owners of diabetic cats with confirmed HS (IGF-1 > 1000 ng/ml, pituitary mass) presented at the Royal Veterinary College were offered hypophysectomy. All cats undergoing surgery were operated by one neurosurgeon with previously only cadaveric experience of the procedure, through an adapted transphenoidal approach referencing bony landmarks to computed tomographic scans reconstructed on neuronavigation software. The somatotrophinoma was extirpated using fine surgical tools. All cats received intense electrolyte and blood pressure monitoring, peri- and post-operative DDAVP and intravenous insulin

and hydrocortisone infusion, transitioning to subcutaneous glargine, conjunctival DDAVP, oral hydrocortisone and levothyroxine.

Between April 2012-February 2014, 12 cats underwent hypophysectomy (median + range age: 10.2 years, 5.4-14.8; IGF-1: 1846 ng/ml, 1138->2000; pituitary height 6.2 mm, 4.0-10.6). All displayed uncontrolled DM due to HS (median fructosamine: 619 umol/l); none displayed overt neurological deficits.

Two cats (17%, cats 7 and 8; pituitary height (mm): 10.6 and 6.6) required mechanical ventilation post-operatively and both were euthanized. Post-mortem magnetic resonance imaging revealed brain herniation and cerebral ischaemia was suspected. One cat suffered cardiac arrest post-operatively at time of jugular catheter placement, though made an uneventful recovery. Four other cats developed congestive heart failure within 5 days, which was successfully treated not necessitating ongoing therapy. Temporarily diminished tear production was seen in 5 cats. Seven of the ten surviving cats went into diabetic remission within a median of 19.7 days (9-42); 3 others saw reduction of insulin needs by 91%. Serum IGF-1 normalised rapidly and significantly in all but one cat (median serum IGF-1 485 ng/ml within 9 days). Persistent neurological deficits or palatal wound breakdown were not encountered.

Starting a hypophysectomy program to treat feline HS was associated with some risk of mortality, though surviving cases benefited from the procedure with a high incidence of diabetic remission.

Conflicts of interest: No conflicts of interest reported.

ESVE-O-9

URINARY EXCRETION OF CALCIUM AND PHOSPHATE IN DOGS WITH PITUITARY DEPENDENT HYPERCORTISOLISM: CASE CONTROL STUDY IN 167 DOGS. F. Fracassi¹, M. Caldin². ¹University of Bologna, Ozzano dell'emilia, Italy, ²San Marco Veterinary Clinic, Padova, Italy

Pituitary dependent hypercortisolism (PDH) in dogs is frequently associated with high serum phosphate and parathormone concentrations. The pathogenesis of such abnormalities remains unknown and the evaluation of the urinary fractional excretion of phosphate and calcium in PDH dogs might be helpful in enhancing the knowledge regarding this issue. The aim of the present study was to evaluate the serum and urinary concentrations and the urinary fractional excretion of phosphate and calcium in dogs with PDH. Medical records from one referral center were retrospectively evaluated between 2003 and 2013. The diagnosis of PDH was confirmed using the cortisol to creatinine ratio, the LDDS test and/or ACTH stimulation test, the plasma ACTH concentration, ultrasonography of the adrenal glands and computer tomography (CT) of the pituitary and the adrenal glands in dogs with consistent clinical signs. Only newly diagnosed dogs, before treatment for PDH, were evaluated. Two control groups were included: one healthy and one sick control dog (without PDH) for each dog with PDH were included. Healthy control dogs (HCD) and sick control dogs (SCD) were matched for age (± 6 months), breed, sex and sexual status. Data were analysed using non-parametric tests and expressed as median and ranges. Significance was set at $p < 0.05$. One-hundred-sixty-seven dogs with PDH were eligible for inclusion in the study. The median age at diagnosis was 10 years (range: 3-16) and the median body weight was 14.7 kg (range: 3.0-65.5). There were 100 female (64 spayed) and 67 male (11 castrated).

Serum phosphate concentration (4.1 mg/dl, 1.3-10.7) was significantly ($p < 0.0001$) higher compared to HCD (3.4 mg/dl, 1.3-5.2) and SCD (3.8 mg/dl, 1.1-32.1). Serum calcium concentration (10.4 mg/dl, 7.0-14.5) was significantly higher compared to SCD (10.1 mg/dl, 5.5-12.9) but not different compared to HCD (10.2 mg/dl, 8.9-12.0). Urinary fractional excretion of phosphate (15.5%, 0.5-65.1) was significantly lower compared to HCD (20.4%, 0.4-67.9) and SCD (16.9%, 0-83.6). Urinary fractional excretion of calcium (0.37%, 0-5.70) was significantly higher compared to HCD (0.22%, 0.02-1.76) and SCD (0.24%, 0-18.71). Urinary calcium to creatinine ratio (4.78, 0-54.50) was significantly higher compared to HCD (2.17 0.20-16.80) and SCD (2.38, 0-16.40), while urinary phosphate to creatinine ratio were not significantly different in PDH dogs, HCD and SCD.

In conclusion PDH dogs have lower phosphaturia and higher calciuria compared to control dogs. This findings suggest that, at least in part, the high serum phosphate concentrations are related to the renal retention of phosphate.

Conflicts of interest: No conflicts of interest reported.

ESVE-O-10

COMPARISON OF FOUR MONITORING METHODS FOR TRILOSTANE TREATMENT OF CANINE HYPERADRENOCORTICISM. L.L. Cosgrove, T. Parkin, I.K. Ramsey. University of Glasgow, Glasgow, United Kingdom

Four cortisol-based methods of monitoring trilostane treatment of canine hyperadrenocorticism were compared to the results of a clinical scoring scheme based on an owner questionnaire.

Cases of canine hyperadrenocorticism that had received a consistent dose of trilostane for more than one month were recruited from first opinion and referral practice. Each dog was used only once. Owners were asked to complete a questionnaire that assessed clinical control. The dogs were then categorised as being over-controlled, well-controlled, moderately-controlled and poorly-controlled. Cortisol was measured in serum samples taken pre-trilostane (peak), 3 hours post-trilostane (trough) and 1 hour post-ACTH injection. Dogs that had an increase in cortisol after trilostane administration were excluded. A scoring system was developed for each of these 3 measurements. A fourth scoring system was developed using a novel algorithm that combined the peak and trough cortisol (peak-trough). The results of each of the 4 scoring systems categorised the dogs into those that would be expected to be over-controlled, well-controlled, moderately-controlled and poorly-controlled. Weighted Kappa was calculated to assess the agreement between the categorisation according to each of the 4 methods compared to the categorisation using the owners score. The Pearson correlation coefficient was calculated to assess relationships between the various parameters.

In total 31 tests were analysed. When compared to the results of the owner's questionnaire 16, 13, 9 and 5 dogs were correctly categorised using the peak-trough, peak alone, post-ACTH and trough alone respectively. Amongst the miscategorised results 12, 17, 16 and 15 dogs were incorrect by 1 category and 3, 1, 6 and 6 dogs by 2 categories using the peak-trough, peak alone, post-ACTH and trough alone respectively. All methods correctly recognised the over-controlled dog that had been identified by the owner's score.

The weighted kappas for post-ACTH and trough cortisol categories compared to the owner score categories were 0.09 and 0.11 respectively (defined as slight agreement). In contrast the weighted kappas for the peak and peak-trough categories were 0.36 and 0.34 respectively (defined as fair agreement). There were no significant correlations between the absolute clinical scores and cortisol concentrations. There were significant correlations between the 3 cortisol measurements.

The novel methods of peak-trough and peak cortisol better reflected the level of clinical control of hyperadrenocorticism identified by the owners' questionnaire than either post-ACTH stimulation or trough cortisol. Peak-trough and peak cortisol concentrations should be further investigated as monitoring methods for trilostane.

Conflicts of interest: Financial support from Dechra pharmaceuticals.

ESVE-O-11

SURVIVAL ANALYSIS OF DOGS WITH ADRENOCORTICAL INSUFFICIENCY. J. Hanson¹, K. Tengvall², B. Bonnett³, A. Hedhammar¹. ¹Swedish University of Agricultural Sciences, Uppsala, Sweden, ²Dept. of Medical Biochemistry and Microbiology (MBIM), Uppsala University, Uppsala, Sweden, ³Consulting Epidemiologist, B Bonnett Consulting, Georgian bluffs, Canada

The prognosis of canine adrenocortical insufficiency is generally regarded to be excellent. However, there is paucity of sur-

vival analyses in the literature. The aim of the present study was to evaluate the survival of dogs with the diagnosis adrenocortical insufficiency based on data from a cohort of 525, 028 Swedish client-owned dogs insured in one insurance company (Agria Pet Insurance, Stockholm, Sweden) during the time period 1995-2006. Dogs were identified by search for insurance claims with the register code for adrenocortical insufficiency. Dogs were excluded from analysis if they had a previous history of hypercortisolism, and if they were born before begin of the study period. Kaplan-Meier survival analysis was performed. Dogs were regarded as censored when the registered cause of death was other than adrenocortical insufficiency or hypercortisolism that was registered after the first claim for adrenocortical insufficiency. Data from 297 dogs was included. One hundred twenty-four dogs were registered to be dead. In 81 dogs the cause of death was related to the adrenocortical insufficiency. The 1-year estimated survival-rate was 81% (95% CI, 76-86%). The 3-year estimated survival-rate was 69% (95% CI, 63-76%). The 5-year estimated survival-rate was 59% (95% CI, 51-68%). Twelve dogs (4.0%) were still alive after 7 years. In conclusion, the long-term survival of dogs with adrenocortical insufficiency was reasonably good. However, the diseases-related mortality was higher than expected, and occurred mainly during the first years after diagnosis.

Conflicts of interest: This study was supported by grants from the Swedish Research Council and the Foundation for Research, Agria Insurance Company.

ESVE-O-12

MULTIPLE ENDOCRINE NEOPLASIA IN DOGS AND CATS: A RETROSPECTIVE STUDY OVER 10 YEARS (2004-2014). L. Beatrice¹, F.S. Boretto², N. Sieber-Ruckstuhl², C. Müller², C. Kümmerle-Fraune², M. Hilbe³, P. Grest³, C.E. Reusch². ¹University of Zurich, Zurich, Switzerland, ²Clinic for Small Animal Internal Medicine, University of Zurich, Zurich, Switzerland, ³Institute of Veterinary Pathology, University of Zurich, Zurich, Switzerland

The concomitant occurrence of two or more endocrine tumors and/or hyperplasias, known as multiple endocrine neoplasia (MEN) is a well-known entity in humans. Multiple gene mutations have been identified. The two major forms are MEN1 and MEN2. In MEN1, the main affected organs are parathyroid, pancreas and pituitary gland. MEN2 occurs in 3 clinical variants: MEN2A, characterized by medullary thyroid carcinoma (MTC), pheochromocytoma and primary hyperparathyroidism; MEN 2B, characterized by MTC, pheochromocytoma and additionally abnormalities; familial medullary thyroid carcinoma. In dogs and cats only a few cases have been reported and it is unknown whether hereditary MEN-like syndromes exist in these species. The aim of this study was to evaluate the prevalence of multiple endocrine tumors in dogs and cats at our institution, to identify possible breed and sex predispositions and to investigate similarities with the human MEN syndromes. Autopsy reports of dogs and cats from 2004 until 2014 were reviewed. Animals with at least two endocrine tumors/hyperplasias (ETH) were included. Autopsy reports of 951 dogs and 1155 cats were examined. 149 dogs had ETH affecting a single organ, 24 had multiple ETH; 123 cats had single ETH, 21 had multiple ETH. In dogs with multiple ETH, the most common breeds were West Highland White Terrier (WHWT, 3/24), Poodle, Golden Retriever, mixed-breed dogs (each 2/24). 14/24 were male (13 intact); 10/24 were female (10 neutered). Median age was 12 years (range 7-18). The most common combination was multiple testicle tumors of various types (4/24). The most common affected organs were the adrenals (18/24). Adrenal cortical adenomas/carcinomas/hyperplasias were mainly associated with pheochromocytomas (3/24), testicle tumors (3/24) and insulinomas (2/24). All 3 WHWTs had adrenal adenomas. Both Poodles had pheochromocytoma associated with pituitary adenoma or adrenal hyperplasia. 2 dogs showed tumor combinations similar to the

human MEN1 syndrome: pituitary adenoma and insulinoma; pituitary adenoma and parathyroid hyperplasia. 19/21 cats were domestic short/long hair, 2/21 were Persians. 11/21 were male (7 castrated); 10/21 were female (9 neutered). The median age was 15.5 years (range 10-19). The most common affected organs were thyroid glands (18/21), combined mostly with lesions of parathyroid (10/21) and adrenal glands (7/21). None of the cats had combinations similar to the human MEN syndromes. The prevalence of multiple ETH in dogs and cats was 2.5% and 1.8%. MEN-like syndromes were extremely rare in dogs and non-existing in cats. No sex predisposition was observed. Possible breed predispositions need further investigations.

Conflicts of interest: No conflicts of interest reported.

ESVIM-O-1

DETECTION OF ANGIOSTRONGYLUS VASORUM IN THE BRONCHOALVEOLAR LAVAGE FLUID OR FAECES OF COUGHING AND HEALTHY DOGS IN BELGIUM. M. Cannonne-Guibert¹, E. Roels¹, Y. Caron¹, B.L. Losson¹, D. Peeters¹, I. Peters², F. Billen¹, C. Clercx¹. ¹University Teaching Hospital, Liège, Belgium, ²TDDS Ltd., The Innovation Centre, University of Exeter, Exeter, United Kingdom

Canine angiostrongylosis is an increasingly reported disease worldwide, including many European countries, possibly due to climatic factors, presence of foxes (acting as reservoir) or more simply, to the availability of more accurate diagnostic methods. Although detection of the first-stage larvae (L1) using the Baermann technique on faecal samples (preferably collected over three consecutive days) remains the gold standard, recently developed serological and molecular tests (quantitative polymerase chain reaction, qPCR) are now available. Until now, the prevalence of canine angiostrongylosis among healthy and coughing dogs in Belgium was unknown.

The aims of the present study were (1) to describe a clinical series of recent autochthonous cases and (2) to retrospectively assess *Angiostrongylus vasorum* qPCR in bronchoalveolar lavage fluid (BALF) samples, collected over the last 7 years from a larger series of dogs, healthy or with other respiratory conditions, in order to investigate the past prevalence of the disease in Belgium.

Seven dogs, living in Southern or Eastern Belgium, were recently diagnosed as having angiostrongylosis (mean age= 2.3 y, mean body weight= 15.3 kg). They all presented with respiratory signs of variable severity. In 5 dogs, BALF was obtained and qPCR was positive in all of them, at moderate or high level (Ct from 26,7 to 29,6) while larvae were detected in the faeces of only 2 animals. In the remaining two dogs, no BALF was obtained, but coproscopy was positive. All dogs responded to medical treatment, consisting in a 3-week course of fenbendazole and/or two spot-on application of moxidectin at 1-month interval.

BALF samples were collected between 2008 and 2014 from 10 asymptomatic client-owned dogs and 55 dogs with various respiratory conditions, including 16 dogs with confirmed bordetellosis, 18 dogs with eosinophilic bronchopneumopathy (EBP), 8 dogs with chronic bronchitis and 13 dogs with bacterial bronchopneumonia, and were retrospectively assessed with a *A. vasorum* qPCR assay. Amongst those 65 dogs, only one BALF, from a dog with EBP, yielded a positive qPCR result. In this dog, faecal analysis was negative.

The present data show that, based on BALF qPCR and coproscopy, presence of angiostrongylosis in healthy and coughing dogs was negligible in Belgium until the last 12 months. It is now considered as an emerging condition and must be included in the differential diagnosis in coughing dogs. The present results also support that qPCR detection of *A. vasorum* in BALF, when available, is an adequate and reliable detection technique.

Conflicts of interest: No conflicts of interest reported.

ESVIM-O-2

LIGNEOUS MEMBRANITIS IN A FAMILY OF SCOTTISH TERRIERS. S.L. Mason¹, C. Fisher², N.X. Bommer³, L. Ressel⁴, L.M. Buckley⁵, P. McElroy⁵, S. Ainsworth⁶, J. Dawson⁶, S. Carter⁶, T.J. Nuttall³. ¹SATH, University of Liverpool, School of Veterinary Science, Neston, United Kingdom, ²Warbreck House Veterinary Centre, Liverpool, United Kingdom, ³University of Edinburgh, Royal Dick School of Veterinary Studies, Roslin, United Kingdom, ⁴Veterinary Pathology, University of Liverpool, School of Veterinary Science, Neston, United Kingdom, ⁵Eye Vet Referrals, Frodsham, United Kingdom, ⁶Infection and Immunity, University of Liverpool, School of Veterinary Science, Liverpool, United Kingdom

Ligneous membranitis is a rare chronic inflammatory disease associated with congenital plasminogen deficiency. It has only been described in six unrelated dogs. The objective of this study is to report the presentation, clinicopathological and post mortem findings in three related Scottish terrier puppies with ligneous membranitis.

Ligneous membranitis is well described in humans, where it is inherited in an autosomal recessive manner. Patients commonly present as infants. Ocular, oral and genital lesions are most common, but other organs are occasionally involved and congenital obstructive hydrocephalus is reported in some individuals. Numerous mutations and polymorphisms in the plasminogen gene have been identified in affected individuals.

The affected Scottish Terriers (two male and one female) presented at 2 months of age with severe proliferative and ulcerative conjunctivitis and gingivitis/stomatitis; biopsy confirmed ligneous membranitis. Other clinical signs included increased upper respiratory tract noise, nasal discharge and lymphadenopathy. One male was cryptorchid. Clinical pathological findings included neutrophilia, proteinuria and hypoalbuminaemia. Serum plasminogen activity was measured in two dogs, and was low in one. The dam and sire of the affected dogs had normal serum plasminogen activity and no history or clinical signs consistent with ligneous membranitis.

No significant clinical improvement was evident following treatment with antibiotics, glucocorticoids, topical ciclosporin or heparin. One dog died of cardiopulmonary arrest in the hospital and the two other dogs were euthanized due to progressive clinical signs. Post-mortem evaluation of the affected dogs revealed multiple abnormalities including severe proliferative fibrinous lesions affecting the trachea, larynx and epicardium, and multiple fibrous adhesions throughout the thoracic and abdominal cavities. The male dog had internal hydrocephalus and lacked a cerebellar vermis.

This is the first report of ligneous membranitis in related dogs and the first report in Scottish Terriers. Sequencing the plasminogen gene in the affected dogs, their parents and unrelated control dogs to identify polymorphisms or mutations that may be associated with ligneous membranitis in dogs is ongoing.

Conflicts of interest: The author received a travel scholarship from Zoetis to attend this congress.

ESVIM-O-3

SCREENING OF APPARENTLY HEALTHY ELDERLY DOGS. A. Willems¹, D. Paepe¹, S. Marynissen¹, P. Smets¹, I. van de Maele², P. Picavet², L. Duchateau¹, S. Daminet¹. ¹Ghent University, Faculty of Veterinary Medicine, Merelbeke, Belgium, ²Hill's Pet Nutrition, Belgium

Health screening of elderly dogs is often recommended, but scientific information on clinical and laboratory abnormalities in senior and geriatric dogs is scarce. This study was undertaken to describe blood pressure measurement, physical examination (PE) abnormalities and routine laboratory test results in senior and geriatric dogs that were apparently healthy for the owner.

Because life expectancy in dogs is related to body size, the inclusion of 100 dogs was based on a human/pet analogy chart to determine whether a dog was senior (n = 41) or geriatric (n = 59). To verify health status, owners were asked to complete an extensive questionnaire. Systolic blood pressure (SBP) was measured using the Doppler technique according to the ACVIM guidelines. Subsequently a thorough PE was performed, including

body and muscle condition scoring, orthopedic examination, neurologic evaluation, indirect funduscopy and bilateral Schirmer tear test. Complete blood count, serum biochemistry and urinalysis (including urinary sediment, urinary protein:creatinine ratio (UPC) and bacterial culture) were evaluated.

In 53 of 100 dogs SBP exceeded 160 mmHg, none of the dogs had fundoscopic lesions secondary to hypertension. Body condition score was abnormal in 41 animals, 39 were overweight or obese. Physical examination revealed a heart murmur in 22, submandibular lymphadenopathy in 13, moderate to severe dental plaque in 51 and one or more (sub)cutaneous masses in 56 dogs. Twenty-three dogs were leukopenic, 29 had a decreased phosphorus, 32 an increased serum creatinine and one dog a decreased total thyroxine (with concurrent increased thyroid stimulating hormone). Crystalluria was commonly detected (62/96) and mostly due to low numbers (<1/high power field) of amorphous crystals (82%). Struvite crystals were present in 18% of the crystalluric dogs. Overt and borderline proteinuria were detected in 13 and 18 of 98 dogs, respectively. Four dogs had a positive urinary culture. SBP was not significantly different between the senior and geriatric group. There was no significant effect of obesity or gender on SBP. The platelet count (p = 0.014), total thyroxine concentration (p = 0.008) and the frequency of orthopedic problems (p = 0.007) and cutaneous masses (p = 0.002) were significantly higher in the geriatric compared to the senior dogs. Hematocrit (p = 0.007) and body temperature (p = 0.044) were significantly lower in the geriatric group.

These findings indicate that physical and laboratory abnormalities are common in apparently healthy senior and geriatric dogs. This underlines the necessity for regular health screening in elderly dogs and the urgent need for reliable and maybe age specific reference intervals in veterinary medicine.

Conflicts of interest: The cost of examinations reported in this study were covered by Hill's Pet Nutrition Belgium.

ESVIM-O-4

ANTINUCLEAR ANTIBODY SPECIFICITY IN DOGS WITH IMMUNE-MEDIATED RHEUMATIC DISEASE. H. Hansson-Hamlin. The Dpt. of Small Animal Clinical Sciences, Uppsala, Sweden

Systemic Lupus Erythematosus, SLE, is a chronic autoimmune disorder with varying clinical manifestations and diagnosis is based on both clinical signs and laboratory findings. Other systemic rheumatic diseases, referred to as SLE-related diseases or immune-mediated rheumatic disease (IMRD), are also described. The most common clinical signs in dogs are stiffness and pain from varying joints. One hallmark of SLE and SLE-related diseases in both dogs and humans is high titres of circulating antinuclear antibodies (ANA), which can be demonstrated by the indirect immunofluorescence (IIF) ANA test. Earlier studies have shown that canine IIF ANA positive samples may be divided into two main subgroups: homogenous (ANA^H) and speckled (ANA^S) IIF ANA fluorescence pattern. In humans, further determination of the specificity of ANA positive sera is frequently employed to characterize the ANA reactivity. Some of these ANA specificities have been demonstrated in man to strongly associate with different systemic autoimmune diseases and also with different IIF ANA staining patterns. Presence and character of antinuclear antibodies in canine SLE-related diseases are not well described.

The aim of this work was to further characterize the ANA specificity in dogs with SLE-related disease/IMRD.

Sera from 208 ANA positive dogs, including 61 different breeds, were analyzed with ELISA and line blot techniques (ELISA and EUROLINE ANA Profile, EUROIMMUN, Germany). The five most prevalent breeds were German shepherd dog, Nova Scotia duck tolling retriever, cocker spaniel, crossbreed and golden retriever. 68 sera displayed a homogenous and 140 a speckled IIF ANA fluorescence pattern.

Several specific ANA-reactivities earlier characterized in human patients were identified.

The majority of ANA^H, n = 41, 60%, showed reactivity against nucleosomal antigens and 9 (13%) against dsDNA when conducted on line blot. These sera also reacted against nucleosomes and dsDNA on the ELISA. There were some additional

positive with the ELISA, so in total the ELISA identified 72% with nucleosomal and 24% with dsDNA reactivity. In few cases, other reactivities identified were against histones, PCNA, Jo-1 and RNP.

In the ANA^S subgroup, the Sm+RNP antigen evoked the most frequent reactivity, $n = 30$, 21% with both line blot and ELISA. In few cases, reactivity against dsDNA, PCNA, Jo-1, PMScl100kD, Scl-70, SSA and SSB were identified. In several dogs no specific antigen was identified.

Further studies are in progress in order to in more detail characterize and identify subtypes of already known and unknown antigens with clinical importance in canine autoimmunity.

Conflicts of interest: One of the authors, Erik Lattwein, is employed by EUROIMMUN where the analyses were performed.

ESVNU-O-1

BIOLOGICAL VALIDATION OF FELINE CYSTATIN C: THE EFFECT OF BREED, AGE AND GENDER AND ESTABLISHMENT OF A REFERENCE INTERVAL. L.F.E. Ghys¹, D. Paepe¹, L. Duchateau¹, E.R.L. Taffin¹, S. Marynissen¹, J. Delanghe², S. Daminet¹. ¹Faculty of Veterinary Medicine, Ghent University, Merelbeke, Belgium, ²Faculty of Health Medicine and Life Sciences, Ghent University, Ghent, Belgium

Chronic kidney disease (CKD) has a high prevalence in cats. Routine renal markers, serum creatinine (sCr) and urea are not sensitive or specific enough to detect early CKD. Serum Cystatin C (sCysC) has advantages over sCr for the detection of early kidney dysfunction, both in humans and dogs. A significant higher sCysC concentration in CKD cats has been demonstrated. The objective of this study was to determine the effect of age, gender and breed on feline sCysC and to establish a reference interval for feline sCysC.

In total, 132 healthy cats between one and 16 years were included. Serum CysC was determined with a validated particle-enhanced nephelometric immunoassay (PENIA). Serum Cr, urea, urine specific gravity (USG), urinary protein: creatinine ratio (UPC) and systolic blood pressure (SBP) were also measured. To test for difference between the groups, the *F*-test was used. The lower and upper value of the 95% reference interval were obtained as the 2.5% and 97.5% quantiles of the sCysC observations.

No significant differences in sCysC concentration were observed between young, middle-aged and old cats; between female, female neutered, male and male neutered cats; and between purebred and domestic short-or longhaired cats. The 95% reference interval for feline sCysC was determined as [0.58-1.95 mg/L]. There was a significant difference in sCr concentration between domestic short- or longhaired cats and purebred cats. The SBP was significantly influenced by gender as well as age, while urea was influenced by both age, gender and breed.

This study showed that the biological factors age, gender and breed have little or no impact on feline sCysC, in contrast to sCr and serum urea, making it an interesting marker. Therefore, further studies are warranted to evaluate the diagnostic value of sCysC as a renal marker in cats.

Conflicts of interest: This study received support from the Institute for the Promotion of Innovation by Science and technology in Flanders (IWT) through a bursary to L. Ghys.

ESVNU-O-2

A PILOT STUDY TO ASSESS THE FEASIBILITY OF TRANSCUTANEOUS GLOMERULAR FILTRATION RATE MEASUREMENT USING FLUORESCENCE-MARKED SINISTRIN IN DOGS AND CATS. S. Steinbach¹, N. Krolop¹, S. Strommer¹, Z. Herrera-Pérez², N. Gretz², R. Neiger¹. ¹Small Animal Clinic (Internal Medicine), Justus-Liebig University, Giessen, Germany, ²Medical Research Center, University of Heidelberg, Mannheim, Germany

Assessment of renal function is often needed, however existing methods including urine and plasma clearances are

invasive, cumbersome and time consuming. In this pilot study the feasibility of a transcutaneous glomerular filtration rate measurement was investigated. The transcutaneous disappearance rate (expressed as half-life) of fluorescein-isothiocyanate-labelled sinistrin (FITC-S) was measured in three healthy research dogs and three healthy research cats. Plasma clearance of sinistrin (7 data points) was performed in both species as previously described (Res Vet Sci 1998;64:151-6 and J Fel Med Surg 2003;5:175-81) and half-life was calculated using a 2-compartment model with a freely available pharmacokinetic calculator (Comput Meth Prog Bio 2010;99:306-14). Renal elimination of FITC-S was measured transcutaneously for 4 hours (7000-8000 data points) using a miniaturized device as described previously for the same purpose in rats (Kidney Int 2011 79:1254-8). The procedures were performed in awake, freely moving animals using escalating doses of FITC-S (10 mg/kg, 30 mg/kg, 50 mg/kg) with a wash-out period of at least 24 h in each animal. To find the best position for the device, multiple devices were placed on each animal. The resulting FITC-S disappearance curves were visually assessed to determine the most suitable location and the appropriate dose to reach an adequate transcutaneous peak signal for kinetic analysis. In both species 30 mg/kg were adequate for kinetic calculation. The most suitable place for the device was the lateral thoracic wall in dogs and the ventral abdominal wall in cats, respectively. Transcutaneous FITC-S clearance was then repeated using the optimal dose and location and in parallel with the plasma sinistrin clearance. Plasma sinistrin clearances [ml/kg/min] were 5.5, 5.0 and 3.8 in the three dogs, respectively. Corresponding plasma elimination half-lives [min] were 26, 31 and 35, and corresponding transcutaneous elimination half-lives [min] were 26, 34 and 55, respectively. Plasma sinistrin clearances [ml/kg/min] were 2.8, 2.2 and 1.9 in the three cats, respectively. Corresponding plasma elimination half-lives [min] were 51, 60 and 61, and corresponding transcutaneous elimination half-lives [min] were 75, 96 and 83, respectively. In conclusion, transcutaneous FITC-S clearance is a feasible method for assessment of GFR in awake dogs and cats. It is noninvasive, well tolerated and easy to perform even in a clinical setting with results being readily available. A dose of 30 mg/kg of FITC-S seems adequate for kinetic assessment. Further studies are now needed to establish reference values and evaluate transcutaneous renal clearance in various conditions.

Conflicts of interest: ZHP and SG are supported by the EC FP7 Marie-Curie programme: NephroTools. The device development was supported by the FP7 activity: PLACE-it.NG is owner of a patent covering FITC-sinistrin and the technology for its measurement.

ESVNU-O-4

INITIAL EVALUATION OF AUTOMATED URINARY ALBUMIN: CREATININE RATIO (UAC) AND URINARY CYSTATIN C: CREATININE RATIO (UCYC) AS SCREENING TESTS FOR THE DETECTION OF AZOTAEMIC CHRONIC KIDNEY DISEASE IN CATS. T.L. Williams, J. Archer. University of Cambridge, Cambridge, United Kingdom

Excretion of urinary biomarkers of renal damage should occur at an early stage of chronic kidney disease (CKD), thus facilitating earlier diagnosis of renal disease. Albumin and cystatin C in the renal ultrafiltrate are mostly reabsorbed by the proximal tubular cells, therefore increased urinary excretion of albumin and cystatin C (UAC and UCysC) would be expected to correlate with the presence of renal tubular damage and CKD. The aim of this study was to establish biological validity of two particle enhanced turbidimetric assays (PETIAs) for the measurement of albumin and cystatin C (previously validated for use in feline urine) by comparing the UAC and UCysC between non-azotaemic cats and cats with azotaemic CKD.

Blood and urine samples were obtained from cats at three UK first opinion practices as part of a geriatric screening programme. Haematology, serum biochemistry (including total thyroxine concentration (TT4)) and urinalysis (including urine protein:creati-

nine ratio (UPC)) were performed. Dental disease score (calculus and gingivitis) and body condition score (BCS) were recorded. Cats with TT4 > 40 nmol/L, evidence of pyuria or bacteruria, or significant systemic disease were excluded. UAC and UCysC were determined in non-azotaemic cats (n = 50) and cats with azotaemic CKD (n = 10, defined as a serum creatinine concentration >153 µmol/L and concurrent urine specific gravity <1.035). Comparisons between the non-azotaemic and azotaemic CKD groups were made using the Mann Whitney U test. Correlations were assessed by Spearman's correlation coefficient. Data are presented as median [25th, 75th percentile] and statistical significance was defined as P < 0.05.

UAC was significantly higher in the azotaemic group than the non-azotaemic group (31.5 [9.8, 97.6]x10⁻³ vs. 11.6 [6.4, 19.0]x10⁻³; P = 0.034), whereas UPC was not significantly different between the groups (P = 0.619). Unexpectedly, UCysC tended to be lower in azotaemic cats than non-azotaemic cats (3.2 [1.5, 9.1]x10⁻⁶ vs. 11.8 [4.4, 23.2]x10⁻⁶; P = 0.071). UAC was weakly positively correlated with serum urea concentration (r_s=0.327, P = 0.011), but was not correlated with serum creatinine concentration. UCysC was not significantly correlated with serum concentrations of urea, creatinine or TT4. UAC was also weakly negatively correlated with dental calculus score (r_s= -0.307; P = 0.019) and BCS (r_s= -0.438; P = 0.001).

UAC appears to be a more sensitive test for azotaemic CKD than UPC, however the apparent low specificity may limit the utility of UAC as a urinary screening test for CKD. Increased UCysC (determined by PETIA) would not appear to be a marker of azotaemic CKD in cats.

Conflicts of interest: No conflicts of interest reported.

ESVNU-O-5

CYSTINURIA IN KROMFOHRLÄNDER DOGS. A. Sewell¹, R. Klein², E. Müller², M. Dick³, U. Giger⁴. ¹Bioscientia GmbH, Ingelheim am rhein, Germany, ²Laboklin, Bad kissingen, Germany, ³Biocontrol, Ingelheim am rhein, Germany, ⁴University of Pennsylvania, Philadelphia, United States of America

Cystinuria is an inherited metabolic disorder that causes defective tubular reabsorption of the aminoacids cystine, ornithine, lysine and arginine (COLA). The low solubility of cystine in acidic urine promotes formation of cystine crystals and uroliths in the urinary tract resulting in the clinical signs of stranguria, urinary obstruction and renal failure in affected individuals. Cystinuria occurs in >70 breeds of dog and has been classified into types IA (Newfoundland, Landseer, Labrador), IIA (Australian Cattle dog), IB (Miniature Pinscher) and III (androgen-dependent; e.g. Mastiff, Irish Terrier).

The Kromfohländer is a medium-sized companion dog, bred initially as a cross between a Wire Fox Terrier and a Grand Griffon Vendéen, first recognised internationally in 1955. Cystinuria has been suspected in this breed but no cases have been reported in the literature to date.

We determined urinary COLA concentrations in 81 adult Kromfohländer dogs aged 1-10 years comprising 48 intact and 6 castrated males, and 26 intact and 1 spayed females. A total of 15 (31%) intact males aged 1.5 to 8.5 years had COLA values >700 µmol/g creatinine and several developed cystic calculi. Furthermore, 9 intact male dogs had increased COLA but normal cystine levels. All castrated males had normal COLA concentrations. No females had increased COLA and cystine concentrations or formed any cystine calculi.

We conclude that cystinuria with cystine calculi occurs frequently in adult intact male Kromfohländer dogs but neither is seen in females. This appears to be an androgen dependent type III cystinuria, as seen in Mastiff-type dogs and Irish Terriers. Thus, castration may resolve the increased urinary COLA excretion and risk for cystine calculi formation and obstruction. The precise mode of inheritance is still unclear. All adult intact male Kromfohländer dogs should be screened by urinary COLA testing.

Conflicts of interest: Work carried out at the author's previous place of employment (University Children's Hospital, Frankfurt, Germany). ACS, RK, EM, MD and UG provide a diagnostic service for cystinuria and other inborn errors of metabolism in companion animals.

ESVNU-O-6

URETERAL CALCULI IN CATS: RETROSPECTIVE ANALYSIS OF SIGNALMENT, CLINICAL DATA, MEDICAL MANAGEMENT AND SHORT-TERM OUTCOME IN 83 CASES (2005 - 2013). A. Baril, G. Benckroun, M. Manassero, C. Chery, A. Decambrou, C. Maurey. ENVA, Maisons alfort, France

Ureteral urolithiasis is an emerging medical concern in cats. There are few reports on epidemiology, diagnosis or medical management of ureteral calculi in cats, particularly in Europe.

Cats diagnosed with ureteral urolithiasis in the teaching hospital of the veterinary school of Alfort from 2005 to 2013 were included in this study. Diagnosis was confirmed with radiographs, ultrasound scan and/or laparotomy. Signalment, clinical signs, clinicopathologic and diagnostic imaging findings, medical treatment and outcome were recorded. Epidemiological data were compared to a reference population of 7600 cats.

Eighty three cats were included in the study. The occurrence of ureteral urolithiasis was significantly higher in Birman (OR 20.11 [11.77 - 34.35]) and Siamese cats (OR 2.78 [1.2 - 6.45]). The mean age was 7 +/- 3.5 years [1 - 14 years]. Clinical signs included dysorexia (54/69), lethargy (53/69), weight loss (40/69) and vomiting (38/69). Polyuria and polydipsia were present in 24/69, 13/69 had a dysuria and 12/69 had an abdominal pain. Renal asymmetry was detected on 38/69 cats. 76/71 were azotemic, 16/65 cats were hyperkalemic, and 8/65 cats were hypokalemic. 4/55 cats were hypercalcemic. 45/56 were anemic. Ureteral calculi (n = 80) were unilateral in 52/69 cats and bilateral in 17/69 cats. Half were located in the proximal third of ureter. Thirty per cent were located in distal ureter. Medical treatment included parenteral administration of fluids (n = 68/68), alpha blockers (n = 56/68), amytryptillin (n = 41/68), antibiotics (n = 52/68) and diuretics (frusemide 17/68, mannitol 27/68). Improvement of renal function and/or hydronephrosis was observed in 33/68 cats. Among those, spontaneous elimination of the calculi occurred in 4 cats (group A). There was no migration of the calculi but improvement of azotemia in the others (group B). No improvement was observed in 35/68 (Group C). Measurements of urolithiasis on radiographs were compared within the 3 groups. Mean length was respectively 1.4 mm [1-3], 2.5 mm [0.9-4.1], and 2.4 mm [1-4] in group A, B, and C. Mean width was respectively 1 mm [1-1], 1.6 mm [0.9-3.1] and 1.8 mm [0.8-3.6] in group A, B, and C.

To the author knowledge, it is the first time that a higher prevalence of ureteral calculi in Birman cats is reported in Europe. Spontaneous elimination of calculus is associated with a small size (<1.5 mm). If the size of calculi tends to be bigger in cats with no improvement of renal function after medical treatment, prospective studies are still needed to determine the best medical treatment.

Conflicts of interest: No conflicts of interest reported.

ESVNU-O-7

SYSTOLIC BLOOD PRESSURE AND RENAL DISEASE IN CLINICALLY ILL CATS NATURALLY INFECTED WITH FELINE IMMUNODEFICIENCY VIRUS. E.R.L. Taffin, D. Paepe, L.F.E. Ghys, I.van de Maele, K.de Roover, S. Daminet. Faculty of Veterinary Medicine, Ghent University, Merelbeke, Belgium

Feline immunodeficiency virus (FIV) infection has been associated with kidney disease, mainly characterised by an increased prevalence of proteinuria in FIV-infected cats. However, studies evaluating renal variables in FIV-positive cats are scarce. Recently, a higher systolic blood pressure (SBP) was reported in a small number of FIV-infected cats. Hypertension is an important cause of proteinuria and a frequent cause of renal disease in human immunodeficiency virus (HIV) positive patients. Therefore, our main objective was to describe SBP in clinically ill FIV-positive cats. Secondly we aimed to evaluate routine renal variables in this population.

Naturally infected clinically ill FIV-positive cats were prospectively included. The Doppler ultrasonic technique was used to measure SBP according to ACVIM guidelines. Serum creatinine (sCreat) and urea (sUrea) concentrations, urine specific gravity

(USG) and urinary protein:creatinine ratio (UPC) were determined.

The study included 99 cats, with a mean age of 8.0 ± 3.7 years and a mean body condition score of 4.6 ± 1.5 on a nine-point scale. The SBP ranged from 91 to 170 mmHg, with a mean of 116 ± 18 mmHg. Only two cats were hypertensive (SBP > 160 mmHg). Both had isosthenuric urine, were borderline proteinuric (UPC 0.2-0.4) and one of them was mildly azotemic. Mean sCreat was 101.7 ± 64.2 $\mu\text{mol/L}$ (reference interval (RI) 44.2-141.4 $\mu\text{mol/L}$) and mean sUrea concentration 8.6 ± 5.2 mmol/L (RI 6.2-10.8 mmol/L). Thirteen cats showed increased sCreat levels, with decreased USG (< 1.035) in eight, proteinuria (UPC > 0.4) in seven and increased sUrea concentrations in ten of them. Five out of ten azotemic cats were proteinuric with a decreased USG. Mean UPC was 0.26 ± 1.06 , with a wide range from 0.05 to 9.38. Borderline proteinuria was present in 40/99 (40.4%) and proteinuria in 28/99 (28.3%). Half of the proteinuric cats had a decreased USG. Mean USG was 1.040 ± 0.013 . One third of all cats had a decreased USG, with isosthenuria in seven of them.

These results demonstrate that proteinuria and poorly concentrated urine are common in naturally infected clinically ill FIV-positive cats, confirming previous reports in cats and humans. However, longitudinal studies of (borderline) proteinuric patients are needed to elucidate the clinical relevance. The low number of hypertensive patients and low mean SBP in our study indicate that hypertension is uncommon and unlikely to be the cause of renal damage in clinically ill FIV-infected cats.

Conflicts of interest: Aratana Therapeutics NV financially supports a clinical trial on the use of antivirals in FIV cats at our university. Screening examinations reported in this trial were part of the required pre-trial investigations for that study. The presenting author is also funded by a scholarship from Aratana Therapeutics AV.

ESVNU-O-8

EVALUATION OF FIRST-LINE ANTIMICROBIAL AGENTS FOR URINARY TRACT INFECTIONS IN CATS WITH AND WITHOUT DOCUMENTED COMORBIDITIES. R. Dorsch¹, C.von Vopelius-Feldt¹, R.K. Straubinger², G. Wolf², K. Hartmann¹. ¹Clinic of Small Animal Medicine, Munich, Germany, ²Institute for Infectious Diseases and Zoonoses, Department of Veterinary Science, Munich, Germany

The objective of this study was to identify the prevalence of bacterial species and the susceptibility of isolates to doxycycline, trimethoprim-sulfamethoxazole (TMS), ampicillin, amoxicillin-clavulanic acid (AMC), cephalothin, and enrofloxacin in cats with urinary tract infections (UTI) with and without predisposing comorbidities.

A retrospective analysis of case records between 2000 and 2009 was performed and resulted in inclusion 271 cats into the study: 128 cats with a systemic comorbidity, 36 cats with indwelling urinary catheters, 40 cats with other local comorbidities, and 67 cats with no comorbidity. The most commonly isolated bacteria were *Escherichia coli* (*E. coli*), *Streptococcus* species (spp.), *Staphylococcus* spp., and *Enterococcus* spp.. The proportion of Gram-negative isolates was significantly higher in the cats with systemic comorbidities than in cats with indwelling urinary catheters ($p < 0.001$) and cats with other local abnormalities ($P < 0.001$), whereas Gram-positive isolates were significantly more commonly isolated from cats with indwelling urinary catheters and other local comorbidities than in cats with systemic comorbidities ($p < 0.001$). The proportions of isolates susceptible to AMC, enrofloxacin, and TMS and the antimicrobial impact factors (IF) were higher than the proportions of isolates susceptible to doxycycline, ampicillin, and cephalothin and the respective IF.

Based on these findings, AMC and TMS would be the first-choice antimicrobial agents for empiric treatment of bacterial UTIs in cats with systemic comorbidities. Cats with local comorbidities have a greater risk for UTI with *Streptococcus* and *Enterococcus* spp. isolates, and antimicrobial IF are lower for this group of cats.

Conflicts of interest: No conflicts of interest reported.

ESVNU-O-9

EMERGENCE OF MULTIDRUG-RESISTANT BACTERIA CAUSING URINARY TRACT INFECTIONS IN COMPANION ANIMALS: A 14-YEAR RETROSPECTIVE STUDY IN PORTUGAL. C. Marques, N. Couto, A. Belas, D. Saial, M. Delgado, C. Pomba. Faculty of Veterinary Medicine, University of Lisbon, Lisbon, Portugal

The increasing rates of resistance exhibited by uropathogens represent a serious problem for the selection of an appropriate antibiotic. The aim of this study was to determine secular trends of companion animal urinary tract infection (UTI) that involve extended-spectrum β -lactamase (ESBL)- and carbapenemase-producing Gram negative bacteria (namely, *Escherichia coli*, *Klebsiella pneumoniae*, *Proteus mirabilis*, *Acinetobacter baumannii*), methicillin-resistant-staphylococci (MRS) and ampicillin and high-level-gentamicin-resistance (HLGR) enterococci. Nine hundred and twenty two uropathogenic bacteria were isolated from dogs and cats, between January 1999 and March 2014, at the Veterinary Teaching Hospital of the Faculty of Veterinary Medicine and at veterinary private practices in the Lisbon area. Isolates were identified using standard commercial systems. Susceptibility testing was performed using the disk diffusion and broth microdilution methods. CLSI breakpoints were applied. Extended-spectrum β -lactamases (ESBL) production was screened by double-disk synergy test. The ESBL, plasmid-mediated AmpC, carbapenemases, *mecA* and *aac(6')-Ie-aph(2'')-Ia* genes were detected by PCR and gene enzymes were sequenced. Among Enterobacteriaceae 0.7% were DHA-producers, 2.7% were ESBL-producers and 3.6% were CMY-producers. All isolates were also multidrug-resistant. Cephalosporinase-producing Enterobacteriaceae were detected in 2000, the first being a CMY-2-producer *E. coli*. All the ESBL-producers were *E. coli* or *K. pneumoniae* producing CTX-M-group 1 enzymes. The first CTX-M-15-producer *E. coli* was detected in 2003 and in 2004 the pandemic O25b-ST131 human virulent *E. coli* CTX-M-15-producing clone was identified. Resistance to carbapenems was detected only in *A. baumannii* causing UTI in cats in 2006 (OXA-66) and 2009 (OXA-23). MRS represented 8.5% of all isolated staphylococci. Detection of methicillin-resistant-*Staphylococcus aureus* (MRSA) occurred back in 2001 and methicillin-resistant-*Staphylococcus pseudintermedius* (MRSP) emerged only in 2007. Ampicillin-resistance in enterococci was present throughout the years (15.4%, n = 8). HLGR appeared in enterococci in 2003 and was confirmed by the detection of the bifunctional enzyme that confers high level resistance to aminoglycosides (7 out of 8 isolates). In this study we showed that in the last decade the emergence of resistance to critically important antimicrobials among uropathogens from companion animals is a concerning fact. The multidrug-resistant Enterobacteriaceae may compromise effective therapeutic options, namely third and fourth generation cephalosporins, fluoroquinolones, trimethoprim/sulpha combinations. The emergence of MRSA/MRSP and HLGR among uropathogens is also a therapeutic challenge. The detection of uropathogens with antimicrobial resistance is not only an animal health issue but also a matter of public health, since companion animals may act as reservoirs of antimicrobial resistant bacteria or resistance genes for humans.

Conflicts of interest: The author currently receives a PhD grant funded by the Portuguese Foundation for Science and Technology. In the past, the author received once research support and honoraries from portuguese Merial for a project on canine vector borne diseases.

ESVNU-O-10

INCIDENCE- AND MORTALITY RATES OF KIDNEY DISEASE IN A LARGE POPULATION OF INSURED SWEDISH DOGS. L.E. Pelander¹, I. Ljungvall¹, A. Egenvall¹, J. Elliott², H.M. Syme², J. Häggström¹. ¹Swedish University of Agricultural Sciences, Uppsala, Sweden, ²Royal Veterinary College, London, United Kingdom

Kidney disease is an important cause of morbidity and mortality in dogs. Knowledge about the epidemiology of kidney disease

in the dog population is valuable both clinically and for planning of research investigating disease mechanisms. Large-scale epidemiological studies are needed.

The aim of the present study was to use insurance data to estimate morbidity and mortality related to kidney disease in the Swedish dog population.

Insurance company data from veterinary care-insured and life-insured dogs during the years 1995-2007 were studied retrospectively. Incidence- and mortality rates were calculated for the whole group of dogs as well as divided by sex and breed. For the 15 breeds with the highest incidence- and mortality rates, respectively, the reasons for kidney disease were investigated by dividing the diagnoses into 10 etiology groups.

The total number of veterinary care-insured dogs was 665,245. The total incidence rate of kidney disease in this group of dogs was 15.7(15.3-16.2) cases/10,000 dog-years at risk. The number of dogs in the life insurance was 548,346 and in this group the total kidney-related mortality rate was 9.7(9.3-10.2) deaths/10,000 dog-years at risk. The 2 most commonly reported etiologies of kidney disease were "etiology not determined" and "infectious/inflammatory". The 3 breeds with the highest incidence rate of kidney disease were the Bernese mountain dog, Miniature schnauzer and Boxer. The 3 breeds with the highest mortality caused by kidney disease were the Bernese mountain dog, Shetland sheepdog and Flatcoated retriever.

In conclusion, the epidemiological information provided in this study concerning kidney disease in dogs can assist clinicians in establishing diagnoses, and can assist breeders in defining priorities for preventative measures. It can also provide valuable information for future research.

Conflicts of interest: The senior author has received money from the insurance company we have used data from to write our study, for another project. Jens Häggström and Ingrid Ljungvall have received financial support for research from Sante Animale, Agria Insurance Ltd, Sveland Insurance Ltd, Forsgren Research Foundation. Both of these authors have also undertaken paid consultancy work for Boehringer-Ingelheim, Ceva Sante Animale.

ESVONC-O-1
APPLICABILITY OF THE 'KIUPEL' 2-TIER GRADING SYSTEM ON CYTOLOGY SPECIMENS IN CANINE CUTANEOUS MAST CELL TUMOURS. F. Hergt¹, W. von Bomhard², M. Wergin¹, J. Hirschberger¹. ¹Clinic for Small Animal Internal Medicine LMU, München, Germany, ²Fachpraxis für Tierpathologie, München, Germany

Mast cell tumours represent the most common cutaneous tumour in the dog. Diagnosis of a mast cell tumour can be achieved through cytological examination of fine needle aspirate. However the grade of the tumour is an important prognostic marker and requires so far histologic assessment. A 2-tier histologic grading system based on number of mitoses, multinucleated cells, bizarre nuclei and karyomegaly was recently proposed by Kiupel et al.

The aim of this study was to assess if the cytomorphological criteria proposed in the 2-tier histologic grading system are applicable on cytology specimens.

Ninety-three mast cell tumour specimens of grade I or grade III according to Patnaik with both histological specimens and fine needle aspirates were retrospectively taken from a data set and histologically and cytologically re-evaluated. According to the Kiupel grading system thirty-six were diagnosed histologically as high grades and fifty-seven were considered low-grade mast cell tumours. The cytologic examination of the corresponding specimens revealed thirty-one high grade and fifty-five low-grade tumours. An agreement between histologic and cytologic diagnosis based on the Kiupel grading system was achieved in eighty-six cases (accuracy 92.4%, specificity 93.9%, sensitivity 91.6%). Five high-grade tumours (13.8%) were considered as low grade on cytology.

Cytologic grading of mast cell tumours in the dog has satisfactory accuracy, sensitivity, and specificity. Histologic grading of canine mast cell tumours still remains the gold standard, but cytology already gives reliable information.

Conflicts of interest: No conflicts of interest reported.

ESVONC-O-2
EVALUATION OF THE JAK2-STAT5 PATHWAY AS A THERAPEUTIC TARGET IN CANINE MASTOCYTOMAS. A. Keller¹, M. Willmann², P. Valent³, E. Hadzijušević³. ¹Medical University of Vienna, Ludwig Boltzmann Cluster Oncology, Vienna, Austria, ²University of Veterinary Medicine Vienna, Vienna, Austria, ³Medical University of Vienna, Department of Internal Medicine I, Vienna, Austria

In canines mastocytomas are among the most frequently diagnosed neoplasms of the skin. High grade mastocytomas (grade III, Patnaik classification) are characterized by an uncontrolled growth of neoplastic mast cells (MC) and a poor prognosis. Recently, the KIT-targeting tyrosine kinase inhibitors masitinib and toceranib have been approved for the treatment of canine MC tumors. These drugs are able to induce responses in mastocytoma patients. However, in many patients, relapses are seen. Therefore, research is focusing on new drug targets. Recently, the transcription factor STAT5 has been reported to play an important role in the proliferation and survival of human neoplastic MC. The aim of the present study was to evaluate the JAK2-STAT5 pathway in canine mastocytomas. To address this aim, the canine mastocytoma cell lines C2 and NI-1 as well as inhibitors directed against JAK2 or STAT5 were employed. As assessed by immunocytochemistry, C2 cells and NI-1 cells were found to express pSTAT5 in their cytoplasm and nuclei. Intracellular expression of pSTAT5 was confirmed by flow cytometry. Interestingly, C2 cells were found to express higher levels of pSTAT5 compared to NI-1 cells. Next, we treated C2 cells and NI-1 cells with various concentrations of the STAT5 inhibitors piceatannol and pimozone and the JAK2 inhibitors AZD1480 and TG101348. As assessed by ³H-thymidine uptake, all 4 compounds were found to inhibit the proliferation of canine MC in a dose-dependent manner. Drug effects were found to vary in different cell lines, with the following rank-order of potency (IC₅₀ values): TG101348: 0.1-0.75 µM; pimozone: 0.1-2.5 µM; AZD1480: 1-2 µM; piceatannol: 5-50 µM. To further explore the mechanism of drug-induced inhibition of proliferation, we examined cell cycle progression and apoptosis in drug-exposed cells. Whereas all 4 drugs tested induced only moderate cell cycle arrests in the G1 phase, these drugs were found to induce substantial apoptosis in C2 cells and NI-1 cells as evidenced by microscopy and Annexin-V/PI staining. Together, our data show that JAK2- and STAT5-targeting drugs exert anti-proliferative and apoptosis-inducing effects in canine mastocytoma cells suggesting that this signaling pathway may be a promising new therapeutic target in canine mastocytomas. The clinical relevance of this observation remains to be determined.

Conflicts of interest: No conflicts of interest reported.

ESVONC-O-3
RETROSPECTIVE ANALYSIS OF MASITINIB-BASED TREATMENT OF SUBCUTANEOUS MAST CELL TUMOURS IN 25 CHEMO NAÏVE DOGS. A.M. Krupa¹, J.P. de Vos¹, G. Polton². ¹De Ottenhorst, Veterinary Oncology Referral Centre, Terneuzen, The Netherlands, ²North Downs Specialist Referrals, Bletchingley, Surrey, United Kingdom

Subcutaneous mast cell tumours (SQMCT) in dogs are relatively uncommon compared to their cutaneous counterparts. The veterinary literature describes these tumours as a specific pathological entity with, in general, a low probability of aggressive progression. Surgery is considered the main treatment modality, while medical treatment has not been described. The purpose of this study was to determine Progression Free Survival (PFS) for a chemo naïve cohort of dogs presented with non-resectable and/or metastasized SQMCT, which all underwent masitinib-based therapy.

Data were collected for patients with SQMCT presented to participating centres in the Netherlands and the UK from 01/12/2008 to 01/01/2014, which received masitinib-based therapy. Treatment protocols employed, included masitinib alone (M), masitinib and prednisolone (MP), masitinib plus vinblastine and/or lomustine and prednisolone (MPC). Response to therapy was measured conforming to RECIST 1.1. Adverse events were

graded using VCOG-CTCAE 1.1. Patients were grouped according to presence or absence of metastasis, treatment protocol used, previous surgery, and remission status achieved; simple comparisons were made to evaluate possible significance.

Twenty-five cases were identified. 18/25 were female. Median age of occurrence was 9 years (2-17). Diagnosis was made by histology in 11/25; proliferation indices were defined in only 4 dogs. Fourteen cases exhibited metastasis at initiation of therapy. PFS for all cases ranged from 8-1275 days. Median/mean PFS (days) according to treatment was M:141/317d (n = 15), MP:142/389d (n = 7), MPC:49/57d (n = 3). Median/mean PFS according to metastasis status was M0:141/324d (n = 11) and M1:103/291d (n = 14). Dogs who underwent previous surgery (n = 10) had a median/mean PFS of 108/344d compared to those who had no surgery 119/280d. Looking at remission status, median PFS of patients who achieved a complete remission was not reached, with a mean PFS of 617d (n = 9). Median/mean PFS of patients with partial remission was 86/176d (n = 11), stable disease 49/57d (n = 2), and progressive disease 12/17d (n = 3). 19/25 cases experienced suspected adverse events. Three dogs, two of which ultimately died, had seven grade 3-5 adverse events (anaemia (n = 3), hepatotoxicity (n = 3), gastrointestinal toxicity (n = 1)).

Masitinib-based treatment is effective in the management of SQMCT perceived to be aggressive. Patients do not appear to benefit from prior surgery. Metastatic status did not influence outcome. Adding chemotherapy negatively influenced PFS. Complete remission is a very favourable prognostic development.

Conflicts of interest: The authors have received financial support from AB Science to help with the costs of statistical analyses in an unrelated project

ESVONC-O-4

OUTCOME OF CANINE DIFFUSE LARGE B-CELL LYMPHOMA DURING CHEMOTHERAPY WITH L-ASPARAGINASE, CYCLOPHOSPHAMIDE, VINCRISTINE AND PREDNISOLONE (LCOP): 30 CASES (2003 - 2013). D. Sayag, F. Floch, T. Marchal, C. Fournel-Fleury, F. Ponce. VetAgro-Sup - Campus Veterinaire de Lyon, Marcy l'etoile, France

Advances in distinction between morphological subtypes of canine non-Hodgkin's lymphomas (NHL) have provided a better understanding of this cancer in dogs. Diffuse large B-cell lymphomas (DLBCL) are the most frequent form of NHL in dogs including some distinguished morphological subtypes (mainly centroblastic polymorphic and immunoblastic) according to the WHO classification. Few clinical studies reported DLBCL clinical outcomes under treatment while survival times of the centroblastic polymorphic subgroup were reported. The aim of this retrospective study was to evaluate the response of DLBCL to a standardized multi-agent chemotherapy protocol.

Medical records from dogs with a diagnosis of DLBCL between 2003 and 2013 were retrospectively reviewed. Inclusion criteria were the availability of complete initial and follow-up information and the application of a standardized multi-agent L-COP chemotherapy protocol as previously described. Dogs which received corticosteroids before the initiation of treatment and dogs which died for other reasons than their related disease before the end of the induction period (35 d) were excluded. Response to chemotherapy was evaluated every week during the induction of treatment, then every 3 to 5 weeks. Statistical analysis was performed using Kaplan-Meier analysis.

Thirty cases of DLBCL meeting all inclusion criteria were included from the initial population. Seven dogs were in clinical stage III according to the WHO classification, 19 in stage IV and 4 in stage V. Nineteen dogs were in substage a, and 11 in substage b. On 26 dogs which have an adequate response evaluation, 20 dogs (76.9%) achieved a complete remission. The median and mean duration of first remission were 175d and 239d, respectively (range 68-725 d). The median and mean durations of survival time were respectively 311d and 319d (range 55-818 d). One year and 2-years survival rates were 32% and 3% respectively. According to the statistical analysis, neither clinical stage (p = 0.726) nor substage at presentation (p = 0.83) or morphological subtype (immunoblastic vs centroblastic polymorphic, p = 0.216) were considered as a significant prognostic factor

regarding the duration of the first remission and the overall survival time. A complete response was significantly associated with longer survival times (p = 0.019).

To conclude, the DLBCL displayed a good clinical response to L-COP protocol, with a median survival time of 311 days. The only significant prognostic criterion identified was a complete clinical response to the treatment. A prospective controlled study on a larger population is warranted to confirm these results.

Conflicts of interest: No conflicts of interest reported.

ESVONC-O-5

EPIRUBICIN RESCUE CHEMOTHERAPY IN CANINE HISTIOCYTIC SARCOMA. S.L. Mason, L. Blackwood. SATH, University of Liverpool, School of Veterinary Science, Neston, United Kingdom

Canine histiocytic sarcoma (HS) is an aggressive round cell neoplasm with a poor prognosis. Both lomustine and doxorubicin have been evaluated as first line chemotherapy agents with response rates of up to 46% and median survival times around 3-5 months.

The aim of this study was to evaluate the response to epirubicin in a population of dogs with HS pre-treated with lomustine. Medical records of dogs with a diagnosis of HS that were treated with lomustine and subsequently epirubicin were retrospectively evaluated.

Fifteen dogs received lomustine followed by epirubicin. There was a measurable response to lomustine in seven of 12 dogs with evident disease, 58% (3 CR & 4 PR). An additional 3 dogs achieved stable disease for an overall biological effective response of 83%. Median TTP following lomustine could be assessed in 12 dogs and was 87 days (Range 34-141).

All fifteen dogs received epirubicin as a rescue agent: nine following progressive disease and three with stable disease on lomustine. One dog received epirubicin after completing six doses of lomustine in complete remission and two dogs in partial remission changed to epirubicin due to hepatotoxicity associated with lomustine.

Response rate to epirubicin was 15% and biologic effective response was 61% (1 CR, 1 PR, 6 SD, 5 PD) in 13 dogs. One dog was euthanized due to epirubicin associated gastro-intestinal toxicity and 1 dog stopped treatment with no assessment of response. Median duration of response to epirubicin was 69 days (Range: 40-347) and 1 dog is still alive and in remission (405 days). Overall median survival time for dogs receiving epirubicin following lomustine was 218 days (Range: 27-500).

Single agent epirubicin is a valid rescue therapy after lomustine for canine histiocytic sarcoma and results in modestly improved overall survival times in responding patients.

Conflicts of interest: The author received a travel scholarship from Zoetis to attend this congress.

ESVONC-O-6

STUDY OF THE MECHANISMS BEHIND COX-2 EXPRESSION IN CANINE MELANOCYTIC TUMOURS. I. Pires, J. Gomes, J. Prada, H. Gregorio, F.L. Queiroga. University of Trás-os-Montes and Alto Douro, Vila real, Portugal

The incidence of melanocytic lesions is increasing among canine population. Canine malignant melanoma could have an aggressive behavior, metastasize early in the course of the disease and be resistant to most current therapeutic regimens leading to the need of finding markers with potential as therapeutic targets. The overexpression of COX-2 seems to play a key role in melanocytic tumours, having being described an association between high COX-2 immunoeexpression and the malignant behavior. In order to contribute to the understanding of the role of COX-2 in melanocytic tumours, three main pathways were investigated: angiogenesis, tumour cell proliferation and inflammatory microenvironment (T-lymphocytes and macrophages).

Fifty one (51) melanocytic tumours [32 cutaneous (12 malignant melanomas and 20 melanocytomas) and 19 oral malignant melanomas] were included. All the samples were submitted to immunohistochemical staining carried out by the streptavidin-biotin-peroxidase method, with a commercial detection system with or without melanin blanching, for detection of the following markers (COX-2, Ki-67, Factor VIII, VEGF, CD3 and MAC387).

In melanocytic tumours (n = 51), both COX-2 labelling extension and intensity revealed a statistically significant association with angiogenesis by Factor VIII (p < 0,01), VEGF (p < 0,01); Ki-67 (p < 0,001), CD3 + T-lymphocytes (p < 0,001) and MAC387 (p < 0,001).

Considering only malignant melanomas (n = 31 cases), COX-2 labelling extension revealed a statistically significant association with angiogenesis (p = 0,041) and CD3 + T-lymphocytes (p = 0,005). COX-2 intensity was also positively associated with angiogenesis (p = 0,008) and with MAC387 (p = 0,015).

Present study demonstrated a link between high COX-2 immunorexpression and increased angiogenesis and tumoural T-lymphocyte and macrophage infiltration in malignant melanomas. These findings reinforce the usefulness of using selective COX-2 inhibitors as a valuable therapeutic tool in malignant melanocytic tumours.

Conflicts of interest: This study received financial support from a company (Meril).

ESVONC-O-7

NEUTER STATUS AND RISK OF CANCER IN A DANISH DOG POPULATION. M.M.E. Larsen, B. Børresen, A.T. Kristensen. Faculty of Health and Medical Sciences, Copenhagen University, Frederiksberg, Denmark

Neuter status and risk of malignant neoplasia is not well evaluated in the canine population, when excluding neoplasia not normally believed to be sex-hormone dependent. Denmark and the Scandinavian countries have a high proportion of intact dogs compared to populations from other parts of the world. In the present study it was hypothesized that there would be no difference in gender and neuter status between the population of canine patients with a non-sex-hormone dependent malignant neoplasia reported to the Danish Veterinary Cancer Registry and a general population. From August 2005 to March 2014, 3801 canine neoplasms were reported to the Danish Veterinary Cancer Registry. The total number of malignant (1788) and benign (1953) were comparable (42% and 46%). Malignant neoplasms totalled 1262, when tumors from areas of distribution with known sex-hormone dependency (reproductive organs, mammary gland, perineal), and cases with unknown area of distribution were excluded. The overall distribution of malignant neoplasia was 481 (38%) intact male dogs, 157 (12.5%) neutered male dogs, 404 (32%) intact female dogs and 220 (17.5%) neutered female dogs. The distribution was even between male and female dogs (50.5% and 49.5%). Compared to a known standard population of dogs, there was an overall statistically significant association of malignant neoplasia with neuter status in both sexes. For both genders this was significant for lymphoma, mast cell tumors and osteosarcomas. For neutered females, but not males, there was increased risk of hemangiosarcoma, squamous cell carcinoma and malignant melanoma. These findings indicate that there might be an association between neuter status and development of malignant neoplasia but larger prospective studies are needed to evaluate the risk of non-sex hormone dependent cancers in neutered dogs.

Conflicts of interest: No conflicts of interest reported.

ESVONC-O-8

SERUM ACUTE PHASE PROTEINS IN FELINE MALIGNANT MAMMARY TUMOURS. H. Vilhena¹, J.J. Cerón², A.C. Figueira³, A. Tvarijonaviciute⁴, F. Teclès⁵, S. Miranda⁵, F. Gärtner⁶, J. Pastor⁴, A.C. Silvestre-Ferreira⁷. ¹Escola Universitária Vasco da Gama / Hospital Veterinário Baixo Vouga / CECAV, Portugal, ²Interdisciplinary Laboratory Clinical Analysis INTERLAB-UMU, University Murcia, Murcia, Spain, ³Escola Universitária Vasco da Gama / ICBAS-UP / IPATIMUP, Coimbra, Portugal, ⁴Departament de Medicina i Cirurgia Animals, Universitat Autònoma de Barcelona, Barcelona, Spain, ⁵Escola Universitária Vasco da Gama / CECAV / HVBV, Coimbra, Portugal, ⁶Instituto de Ciências Biomédicas Abel Salazar / IPAT-IMUP, Porto, Portugal, ⁷Departamento de Ciências Veterinárias, UTAD / CECAV, Vila real, Portugal

Serum acute phase proteins (APPs) are considered biomarkers of the acute phase reaction, and are being increasingly used in human and veterinary medicine in diagnosis and monitoring of neoplastic diseases. In the cat, serum amyloid A (SAA) is considered a positive major APP, haptoglobin (Hp) a moderate APP, and albumin and insulin-like growth factor-1(IGF-1) negative APPs.

The aim of the present study was to characterize the APPs response in cats with mammary tumours. For that purpose, SAA, Hp, IGF-1 and albumin serum concentrations were determined in 20 female cats with malignant mammary tumours. Cats with history of previous tumours or with concomitant tumours or other diseases were excluded. Information on cats age, gender, breed, tumour type, histological grade, tumour size and location, skin ulceration, vascular neoplastic infiltration, necrosis, metastasis to regional lymph nodes, thoracic or abdominal organs, and survival time from diagnosis was assessed. Blood samples were collected before surgery in all cats, and whenever possible, serial samples collected on control visits. Owners gave informed consent.

Studied population included 20 domestic short-haired cats with ages ranging from eight to 17 years (11,5 + / - 2,7). All had carcinomas, including solid carcinomas (n = 10), tubulopapillary carcinomas (n = 8), one cribriform carcinoma and one carcinosarcoma.

At the time of diagnosis, 70% of cats had an increase in serum concentration of Hp and 20% of SAA, and 25% had a decrease in concentration of albumin. Mean and standard deviation values were of 2,76 + / - 0,48 g/dl for albumin (reference range 2,5-3,6 g/dl), 319,12 + / - 162,89 µg/dl for IGF-1, 12,95 + / - 29,76 µg/ml for SAA (reference value 5 µg/ml), and 5,12 + / - 3,20 g/l for Hp (reference value 3 g/l).

A positive correlation (r = 0,60) was detected between increases in serum concentrations of SAA and Hp. The increase in the size of tumour was significantly associated with the concentration of SAA (p 0,05). Serum Hp concentrations were significantly increased in tubulopapillary carcinomas (p 0,05), and IGF-1 decreased in solid carcinomas (p 0,05). In the cats where serial determinations were performed, development of thoracic metastasis was significantly associated with a decrease of serum concentration of albumin (p 0,05), and with an increase of SAA (p 0,05).

This study suggests that feline mammary tumours are associated with an acute phase response. According with the results obtained, SAA, Hp, albumin and IGF-1 might be important serum biomarkers in diagnosis and monitoring of the evolution of feline malignant mammary neoplasias.

Conflicts of interest: No conflicts of interest reported.

ESVONC-O-9

ESTABLISHMENT AND CHARACTERISATION OF A NOVEL CANINE HISTIOCYTIC SARCOMA CELL LINE. P.D. Martin¹, A. Gow^{1,2}, B. Beraio², T. Raposo², P.M. Beard¹, J. Lawrence¹, D.J. Argyle^{1,2}. ¹University of Edinburgh, Roslin, United Kingdom, ²The Roslin Institute, Roslin, United Kingdom

Histiocytic sarcoma (HS) is a neoplastic proliferation of interstitial dendritic cells or tissue macrophages. Dogs with HS can present with local disease or with multifocal (disseminated) involvement. Disseminated HS is poorly responsive to therapy and almost always fatal. Little is established regarding the aetio-

pathology of histiocytic sarcoma in dogs. The purpose of this study was to establish and characterise a HS cell line from fresh tumour samples obtained from a dog with disseminated HS in order to further clarify disease pathogenesis and behaviour.

With owner consent, treatment-naïve tumour sections were collected from a dog with disseminated HS that was euthanased. Tumour tissue was assessed with immunohistochemistry (IHC) using antibodies against canine CD18, CD3, and PAX-5 to support the diagnosis of histiocytic sarcoma. Primary cell cultures (HSCs), established from the tumour were cultured and maintained in modified Eagle's medium with 10% fetal bovine serum, L-glutamine, penicillin and streptomycin, in standard conditions. HSCs were characterised by alpha naphthyl acetate esterase (ANAE) and lysozyme staining while PCR was used to detect cell markers CD1a, CD11c, MHC II, CD204, CCR2, E-cadherin, and CD4. Cell surface markers were compared to an established canine HS cell line (DH82). Phagocytic activity of HSC cells was assessed using cellular uptake of carboxylated fluorescent beads and documented using flow cytometry and fluorescent microscopy.

Tumour tissue was strongly CD18 positive and negative for CD3 and PAX-5. Cultured cells exhibited morphological characteristics consist with dendritic cells, such as projections and pleomorphism. HSC cells stained positively for non-specific esterase (ANAE) and lysozyme, and PCR indicated cells were positive for CD1a, CD11c, MHC II and CD204 and negative for CD90 and E-cadherin. HSC cells were positive for MHC II and CCR2 while DH82 cells were negative. Phagocytic activity was evident.

A novel HS cell line (HSC) was established and characterized from primary tumour tissue collected from a dog with disseminated disease. HSC cells were most consistent with interstitial dendritic cell origin based on CD1a, CD11c, and MHC II staining as well as demonstrable phagocytic activity. HSC cells also displayed expression of CCR2, unlike the established DH82 line, supporting a notion that HS consists of a variety of subtypes. CCR2 has been linked to HS growth and metastasis, suggesting it may represent a possible therapeutic target. Further studies establishing and characterising canine HS cells may contribute to the elucidation of mechanisms of tumourigenesis.

Conflicts of interest: No conflicts of interest reported.

ISCAID-O-1

CLINICAL AND GENETIC CHARACTERIZATION AND VIRUS NEUTRALIZATION PATTERNS OF FELINE CALCIVIRUS ISOLATES FROM FOUR VIRULENT-SYSTEMIC DISEASE OUTBREAKS IN CATS IN SWITZERLAND AND LIECHTENSTEIN. B. Willi¹, A.M. Spiri², A. Berger², M.L. Meli³, B. Riond², F. Graf⁴, K. Diserens⁵, I. Padru⁶, A. Samman⁷, R. Hofmann-Lehmann³. ¹Clinical Laboratory and Clinic for Small Animal Internal Medicine, Zurich, Switzerland, ²Clinical Laboratory, University of Zurich, Zurich, Switzerland, ³Clinical Laboratory and Center for Clinical Studies, University of Zurich, Zurich, Switzerland, ⁴Tierärztliche Praxis Dr. Felix Graf AG, Buchs, Switzerland, ⁵Cabinet Vétérinaire du Dr. Gmür, Lausanne, Switzerland, ⁶Clinic for Small Animal Internal Medicine, University of Zurich, Zurich, Switzerland, ⁷Medical Research Council, University of Glasgow Centre for Virus Research, Glasgow, United Kingdom

Feline calicivirus (FCV) is a RNA virus that causes upper respiratory tract disease, oral ulcerations and limping syndrome. Virulent-systemic (VS)-FCV that induce cutaneous edema, ulcerations of the head and feet, and occasionally jaundice have been described in the USA and Europe. Here we characterize for the first time VS-FCV outbreaks in cats in Switzerland and Liechtenstein.

The four outbreaks occurred in three geographically separated locations: Schaan (Liechtenstein, shelter 1), Zurich (Switzerland) and Lausanne (Switzerland, shelter 2) between November 2011 and January 2013. PCR (FCV and Feline herpesvirus-1, FHV-1), virus isolation and FeLV/FIV testing were performed on saliva and blood samples collected from clinically affected cats. Furthermore, saliva for PCR was collected from 31 additional cats in shelter 1. Phylogenetic analyses were performed based on the capsid (VP1) gene sequence of FCV. VS-FCV isolates were tested for

virus neutralization with sera raised against common FCV vaccine strains.

Outbreak 1 occurred in a cattery in Liechtenstein and involved five non-vaccinated, 3-months old siblings with fever, edema, skin and tongue ulcerations. Outbreak 2 occurred in a small animal clinic in Zurich. A 10-year old cat presented with severe paw edema, fever, tongue and skin ulcerations, progressive hypoproteinemia and hyperbilirubinemia. Outbreaks 3 and 4 happened in a cattery in Lausanne five months apart and involved two litters of non-vaccinated, 2- to 3- months old kittens. The cats presented with fever, nasal discharge, edema and skin and oral ulcerations.

All affected cats tested FCV-positive but negative for FHV-1, FeLV and FIV, except for one kitten from outbreak 4 (FIV-positive). All cats in outbreaks 1 and 3 recovered, whereas all cats in outbreaks 2 and 4 died or were euthanized because of clinical deterioration. Each outbreak was caused by a phylogenetically distinct VS-FCV strain. In shelter 1, the queen and three in contact cats remained asymptomatic although infected with the same VS-FCV strain. Furthermore, 7/27 other cats of shelter 1 were infected with closely related, but distinct FCV strains. The VS-FCV isolates from the two outbreaks in shelter 2 were distinct but phylogenetically related. All VS-FCV isolates from cats from the same outbreak showed a similar virus neutralization pattern, but neutralization differed between different outbreaks.

In conclusion, all VS-FCV outbreaks involved multi-cat environments. The same VS-FCV strains with similar virus neutralization patterns were isolated from cats from the same outbreak. Not all cats infected with a VS-FCV strain developed disease and mortality varied between the outbreaks.

Conflicts of interest: The sera for virus neutralization were provided by Merial, France.

ISCAID-O-2

CASE CONTROL STUDY ON FELINE CALCIVIRUS TO INVESTIGATE RISK AND PROTECTIVE FACTORS FOR INFECTION. A. Berger¹, B. Willi², M.L. Meli³, F.S. Boretto⁴, S. Hartnack⁵, H. Lutz¹, R. Hofmann-Lehmann³. ¹Clinical Laboratory, University of Zurich, Zurich, Switzerland, ²Clinical Laboratory and Clinic for Small Animal Internal Medicine, Zurich, Switzerland, ³Clinical Laboratory and Center for Clinical Studies, University of Zurich, Zurich, Switzerland, ⁴Clinic for Small Animal Internal Medicine, University of Zurich, Zurich, Switzerland, ⁵Section of Epidemiology, University of Zurich, Zurich, Switzerland

Feline calicivirus (FCV) is a RNA virus that occurs worldwide. Infected cats show upper respiratory tract disease (URTD; defined herein as presence of sneezing, nasal- and/or ocular discharge, conjunctivitis and/or keratitis), but also oral cavity lesions, chronic stomatitis, limping syndrome and, rarely, virulent systemic disease. The aims of the present study were to compare cats suspected of FCV (FCV-SC) based on clinical symptoms and healthy controls (controls) and to investigate potential risk and protective factors, such as co-infection with feline herpesvirus-1 (FHV-1), *Mycoplasma felis*, *Chlamydomydia felis*, *Bordetella bronchiseptica* and feline retroviruses, vaccination, gender, age, breed, housing and corticosteroid and antibiotic treatment. Oropharyngeal, nasal and conjunctival swabs from 200 FCV-SC and 100 controls were collected into transport medium, processed within 96 hours after collection and analyzed for FCV by virus isolation and for all tested pathogens using molecular assays. The samples were collected by randomly selected veterinary practices in 20 different areas of Switzerland (10 FCV-SC and 5 controls/area). To record clinical data, retroviral status and vaccination history of the cats, a questionnaire was filled out by the private veterinarian. The seven tested pathogens were found in the investigated population. The prevalence (FCV-SC vs. controls) was: FCV 45% vs. 8%; FHV-1 20% vs. 9%, *C. felis* 8% vs. 1%, *B. bronchiseptica* 4% vs. 2%, *M. felis* 48% vs. 31%, Feline leukemia virus 2% vs. 1% and Feline immunodeficiency virus 2% vs. 1%. FCV-SC were positive for FCV significantly more often compared with controls (OR 9.2) and shed more FCV. Co-infections with up to four pathogens were detected; FCV-SC were significantly more frequently co-infected (40%) compared with controls

(14%). Gingivostomatitis and oral ulceration but not URTD were highly associated with FCV infection. In contrast, *C. felis* was associated with URTD; FHV-1 was associated with nasal and ocular discharge and *M. felis* with conjunctivitis and ocular discharge. Risk factors for FCV infection were housing in groups (especially ≥ 4 cats), an intact gender, Maine Coon breed and corticosteroid therapy. FCV-positive cats with gingivostomatitis were older and more commonly vaccinated than FCV-positive cats without gingivostomatitis. Moreover they shed more FCV than cats with URTD. Vaccination and primary immunization defined as two vaccinations 2-6 weeks apart with the same vaccine brand were protective factors against FCV but not FHV-1 infection. Vaccination was associated with a decreased incidence of URTD in FCV-infected cats (OR 0.3). Further analyses will investigate cross-neutralization patterns of the prevailing FCV isolates.

Conflicts of interest: The study was partially funded by Merial, France, and Biokema, Switzerland.

ISCAID-O-3
EFFICACY OF PASSIVELY TRANSFERRED ANTIBODIES IN CATS WITH ACUTE VIRAL UPPER RESPIRATORY TRACT INFECTION. Y. Friedl¹, B. Schulz¹, A. Knebl¹, C. Helps², U. Truyen³, K. Hartmann¹. ¹Clinic for Small Animal Medicine, Munich, Germany, ²Molecular Diagnostic Unit, Langford Veterinary Services, University of Bristol, Bristol BS40 5DU, United Kingdom, ³Institute of Animal Hygiene and Public Veterinary Services, University of Leipzig, 04103 Leipzig, Germany

Antibody preparations are commonly used for the treatment of feline upper respiratory tract disease (FURTD), although their efficacy has not been proven. The aim of this study was to evaluate efficacy of a commercial serum containing antibodies against feline herpesvirus-1 (FHV-1) and feline calicivirus (FCV) in cats with acute viral FURTD.

This prospective, randomized, placebo-controlled, double-blind study included 42 cats with acute (<7 days) clinical signs of FHV-1 and/or FCV infection (confirmed by quantitative PCR). All cats received symptomatic treatment and either hyperimmune serum ($n = 22$) (≤ 12 weeks 2 ml, >12 weeks 4 ml, subcutaneously q24 h, topically into eyes, nostrils, and mouth q8 h) or saline ($n = 20$) for three days. Clinical signs, including a 'FURTD score' and general health status, were recorded daily (day 0 to 7 and on day 21). FCV shedding was determined on day 0 and 21. Statistical analyses included one-way analysis of variance, Mann-Whitney *U*, and Student's *t*-test (improvement of clinical signs), Fisher's exact test (FCV shedding), and Spearman analysis (correlation clinical signs with virus load).

Clinical signs and general health status improved significantly in both groups. However, while placebo-treated cats had only improved significantly by day 7, cats receiving antibodies already significantly improved in their 'FURTD score' ($P = 0.046$) and general health status ($P = 0.032$) by day 3. There was no significant difference in the number of cats shedding FCV and no correlation between viral load and clinical manifestation.

Administration of antibodies lead to faster improvement of clinical signs in cats with acute viral FURTD, but did not influence FCV shedding.

Conflicts of interest: No conflicts of interest reported.

ISCAID-O-4
SAMPLING SITES FOR DETECTION OF FELINE HERPESVIRUS-1, FELINE CALICIVIRUS, AND CHLAMYDOPHILA FELIS IN CATS WITH FELINE UPPER RESPIRATORY TRACT DISEASE. B. Schulz¹, C. Schulz¹, R.S. Mueller¹, C. Helps², K. Hartmann¹. ¹Medizinische Kleintierklinik, Muenchen, Germany, ²University of Bristol, Bristol, United Kingdom

Different viral and bacterial pathogens can be involved in feline upper respiratory tract disease (FURTD). Although some clinical signs have been associated with certain pathogens, clinical

signs can be variable and non-specific. Aim of the study was to compare detection rates of feline herpesvirus-1 (FHV-1), feline calicivirus (FCV), and *Chlamydomphila felis* (*C. felis*) in cats with FURTD on 4 different sampling sites, and to correlate test results and clinical signs.

Swabs of nose, oropharynx, tongue, and conjunctiva were taken from 104 cats with signs of FURTD. On all samples, reverse transcription polymerase chain reaction (RT-PCR) was performed for detection of FCV, and polymerase chain reaction (PCR) for detection of FHV-1 and *C. felis*. Fisher's exact test was used for all comparisons. The level of significance was $p < 0.05$.

Pathogens were detected in 89.4% of cats. Of these, 55.8% were positive for FHV-1, 50.0% for FCV, and 35.6% for *C. felis*. FCV was isolated significantly more often from oropharynx (92.3% of FCV-positive cats) and tongue (90.4%) compared to conjunctiva (38.5%) ($p < 0.001$). There was no significant difference between the 4 sampling sites for detection of FHV-1 and *C. felis*. In addition, there was no preferred sampling site in cats with respective clinical signs, including oral ulceration, conjunctivitis, and keratitis.

In cats with FURTD, the oropharynx can be recommended as the preferred sampling site for detection of FCV, FHV-1, and *C. felis*. Based upon clinical signs it cannot be determined which sampling site should be selected for detection of the pathogens.

Conflicts of interest: No conflicts of interest reported.

ISCAID-O-5
IMMUNOHISTOCHEMICAL DETECTION OF IGG AND IGM IN LUNG TISSUE OF DOGS WITH LEPTOSPIRAL PULMONARY HAEMORRHAGE SYNDROME (LPHS). S. Schuller¹, S. Callanan², S. Worrall², T. Francey¹, A. Schweighauser¹, J.E. Nally². ¹Bern University, Bern, Switzerland, ²University College Dublin, Dublin, Ireland

Leptospirosis Pulmonary Haemorrhage Syndrome (LPHS) is a severe form of leptospirosis, which has been increasingly recognised in humans and many animal species in the past 20 years. Patients with LPHS may develop rapidly progressive intra-alveolar haemorrhage, leading to high mortality. The pathogenic mechanisms of LPHS are poorly understood hampering the application of effective treatment strategies. Studies in humans and experimentally infected guinea pigs have demonstrated deposition of immunoglobulin and complement C3 in LPHS lung tissue in the absence of significant numbers of leptospire, suggesting that LPHS is, in part, caused by autoimmunity. The aim of this project was to describe the histopathologic features of LPHS in dogs and to investigate whether IgG and IgM deposition is present in affected canine lung tissue.

Single-step immunohistochemistry (IHC) for dog IgG, IgM and leptospiral outer membrane vesicles was performed on lung tissues from 11 dogs with LPHS, 4 dogs with pulmonary haemorrhage due to other causes and 4 healthy dog lungs.

Acute intra-alveolar haemorrhage and oedema in the absence of significant inflammatory infiltrates were present in all LPHS lung tissues. Three IHC staining patterns were observed in LPHS lung tissue: alveolar septal wall staining with (IgG $n = 8$ /IgM $n = 6$) and without intra-alveolar staining (IgG $n = 2$ /IgM $n = 0$) and staining of intra-alveolar fluid only (IgG $n = 1$ /IgM $n = 5$). Intra-alveolar staining appeared to favour alveolar surfaces in some cases (IgG $n = 5$ /IgM $n = 5$). Healthy control lungs showed no staining, whereas haemorrhagic lung showed staining of intra-alveolar fluid (IgG/IgM $n = 3$) and occasional, mild and discontinuous staining of alveolar septa ($n = 1$). Leptospirosis antigens were not detected in any of the tissues.

Results indicate that histopathologic features of canine LPHS are similar to what has been described in other species. IHC demonstrated that alveolar septal deposition of IgG/IgM is present in most dogs with naturally occurring LPHS. While these findings support a role of the humoral immune response in the development of LPHS, our findings do not indicate whether autoimmunity is a primary or secondary event in the pathogenesis of LPHS.

Conflicts of interest: No conflicts of interest reported.

ISCAID-O-6

CLINICAL, LABORATORY, RADIOLOGICAL FEATURES AND OUTCOME OF 99 DOGS WITH LEPTOSPIROSIS (2006-2013). S.V. Knoepfler¹, A. Mayer-Scholl², E. Luge², K. Noeckler², R. Klopffleisch³, A. Gruber³, B. Kohn¹. ¹Clinic of Small Animals, Faculty of Veterinary Medicine, FU Berlin, Berlin, Germany, ²Federal Institute for Risk Assessment, Berlin, Germany, ³Department of Veterinary Pathology, Freie Universität Berlin, Berlin, Germany

Leptospirosis, a zoonotic bacterial disease with a worldwide distribution, is a re-emerging disease in humans and dogs. Acute renal and hepatic failure are the most frequently reported clinical manifestations of canine leptospirosis.

The aim of this study was to describe clinical, laboratory and radiological features, the outcome as well as the distribution of *Leptospira* serogroups in dogs with leptospirosis (2006-2013).

Medical records of dogs diagnosed with leptospirosis were evaluated retrospectively. Diagnoses were based on microscopic agglutination testing (MAT), blood/urine PCR, and histopathology (Levaditi staining). MAT-titers \geq 1:800 against non-vaccine and \geq 1:3200 against vaccine serovars or a 4-fold rise of titers within 2-3 weeks were considered diagnostic.

99 dogs met the inclusion criteria. In 72 dogs diagnostic MAT-titers were present (mainly against serogroups Grippotyphosa (65%), Australis (61%), and Pomona (60%).

At initial presentation, the most common clinical signs were lethargy (96%), anorexia (88%), vomitus (85%), a painful abdomen (39%), diarrhea (38%), oliguria (27%), tachypnea (26%), delayed capillary refill time (18%), pale mucous membranes (17%), fever (15%), hypothermia (15%), and icteric mucous membranes (10%). Abnormal findings of the CBC included anemia (63%), thrombocytopenia (62%) and leukocytosis (57%). Biochemistry abnormalities included increased creatinine concentrations (82%), increased liver enzyme activities (80%), hyperbilirubinemia (70%), hyperphosphatemia (67%), hyponatremia (63%), and hypoalbuminemia (55%). Urinalysis often revealed glucosuria (77%) and an elevated urine-protein/creatinine-ratio (75%). Radiological pulmonary changes were detected in 57% of the dogs initially or during the course of disease. 32 dogs died or were euthanized, 24 of them due to "leptospirosis pulmonary hemorrhage syndrome".

In this study, non-vaccine serogroups were the most common serogroups detected by MAT. In the majority of patients renal (95%) and/or hepatic (93%) disease was detected. A pulmonary form of leptospirosis was present in 57% of the dogs. Lung involvement represented a severe complication causing increased mortality depending on the severity of respiratory signs.

Conflicts of interest: No conflicts of interest reported.

ISCAID-O-7

CARDIAC AND EXOCRINE PANCREATIC INVOLVEMENT IN DOGS WITH LEPTOSPIROSIS. A. Buono¹, S. Knöpfler², A. Mayer-Scholl³, K. Nöckler³, J.S. Suchodolski¹, J.M. Steiner¹, B. Kohn². ¹Gastrointestinal Laboratory, Texas A&M University, College station, United States of America, ²Small Animal Clinic, Faculty of Veterinary Medicine, Freie Universität Berlin, Berlin, Germany, ³Federal Institute for Risk Assessment, Berlin, Germany

Leptospirosis is a zoonotic disease that can affect multiple organs with renal and hepatic involvement being considered to be the most common. The aim of this study was to evaluate a large number of dogs with leptospirosis for cardiac and/or exocrine pancreatic involvement.

A total of 59 dogs were diagnosed with leptospirosis based on clinical signs and either microscopic agglutination test, blood/urine polymerase chain reaction, and/or histopathology. At the time of admission and, in most patients, after an average of two weeks canine pancreatic lipase immunoreactivity (cPLI, as measured by Spec cPLI[®]), ultrasensitive cardiac Troponin I (cTnI), and C-Reactive Protein (CRP) were analyzed. Data were analyzed with non-parametric statistics. The level of significance was set at $p < 0.05$.

Upon admission, common clinical signs reported included lethargy (n = 57), vomiting (n = 50), abdominal pain (n = 20), dyspnea (n = 16), pale mucous membranes (n = 13), oliguria (n = 11), hypothermia (n = 11), and fever (n = 10). Anemia (n = 39), thrombocytopenia (n = 41), leukocytosis (n = 38), were frequently reported hematology findings. Increased concentrations of creatinine (n = 48/59), phosphorus (n = 43/57), ALT (n = 31/58), SAP (n = 43/57) and bilirubin (n = 41/58) were also frequently recorded.

CRP (median: 48.7 mg/L; range: 0.1-60.1 mg/L, reference interval (RI): 0.1-7.6 mg/L), cTnI (median: 0.137 ng/L; range: 0.005-24.063 ng/L, RI: 0-0.059 ng/L), and cPLI (median: 217 μ g/L; range: 29-1001 μ g/L, RI: 0-200 μ g/L) concentrations were above the upper limit of the reference intervals in 52/59 (88%), 42/59 (71%), and 30/59 (51%) dogs, respectively and serum cPLI concentration was above the suggested cut-off value for a diagnosis of pancreatitis in 15/59 (25%) dogs. CRP and cTnI, but not cPLI were higher upon admission compared to the re-check measurement ($p = 0.0001$ and 0.0056 , respectively). Dogs with increased serum cPLI concentrations also showed a higher proportion of dogs with increased serum cTnI concentrations ($p = 0.001$). There was no statistically significant correlation of cPLI concentrations with a history of abdominal pain and/or vomiting. Biochemical results were compatible with multiple organ impairment with involvement of kidneys, liver, heart, and exocrine pancreas where at least two organs were affected in 36/59 (61%) dogs. Forty (68%) of 59 dogs recovered, 10 (17%) died, and 9 (15%) were euthanized. cTnI and cPLI were higher in non-survivors, but these differences did not reach statistical significance. However, the number of organs affected and outcome were significantly correlated ($p = 0.012$).

Our data suggest that infection with *Leptospira* is characterized by a systemic inflammation with variable multiple organ involvement and damage, often including the heart and also the exocrine pancreas.

Conflicts of interest: The study was funded by Texas A&M University. The primary author and two co-authors work at the GI Laboratory, Texas A&M University.

ISCAID-O-8

MOLECULAR IDENTIFICATION OF BARTONELLA IN DOGS WITH LEISHMANIOSIS (LEISHMANIA INFANTUM) WITH OR WITHOUT ARTHRITIS. M. Mylonakis¹, N. Soubasis¹, N. Balakrishnan², K. Theodorou¹, D. Kasabalas³, M. Saridomichelakis³, C. Koutinas¹, A. Koutinas¹, E. Breitschwerdt². ¹School of Veterinary Medicine, Aristotle University of Thessaloniki, Thessaloniki, Greece, ²North Carolina State University (NCSU-CVM), Raleigh, NC, USA, Greece, ³School of Veterinary Science, University of Thessaly, Karditsa, Greece, Greece

Canine bartonellosis is increasingly recognized worldwide and may be associated with diverse clinical manifestations. Recent evidence suggests that bartonellosis also causes lameness and polyarthritis in dogs. However, PCR amplification of *Bartonella* DNA and isolation of *Bartonella* species from canine synovial fluid (SF) samples have rarely been reported. Canine leishmaniosis (CanL) due to *Leishmania infantum* is a multisystemic disease commonly associated with polyarthritis. Based on the hypothesis that concurrent *Bartonella* infection may be a contributing factor for the development of arthritis in dogs with CanL, the main objective of this study was to investigate the microbiological and molecular prevalence of *Bartonella* spp. in dogs with naturally-occurring CanL, with or without cytologically documented arthritis. From a previous study, 38 dogs with CanL were retrospectively studied for *Bartonella* spp. infection. Diagnosis of CanL was based on compatible clinical and clinicopathological abnormalities, positive serology, and lymph node or bone marrow (BM) cytology. Dogs with serological evidence of other vector-borne infections (anaplasmosis, borreliosis, dirofilariosis and ehrlichiosis) and dogs recently vaccinated or medicated were excluded from the study. Arthritis defined as a neutrophil percentage in excess of 10% of nucleated cells in SF cytology was documented in 31/

38 (81.6%) of dogs. A total of 74 archived specimens from 38 dogs, including 33 EDTA-anticoagulated blood samples, 19 BM and 22 SF aspirates were tested for *Bartonella* spp. DNA using a *Bartonella* alpha proteobacteria growth medium (BAP-GM) diagnostic platform. Eight (21.1%) dogs were infected with one or two *Bartonella* species, including *Candidatus Bartonella merieuxii* (n = 5), *B. henselae* SA2 (n = 3) and *B. rochalimae* (n = 1). *Bartonella* spp. DNA was amplified from BM in 4 dogs and from blood in 3 dogs but was not amplified from any SF sample. Overall, 6 (19.4%) dogs with and 2 (28.6%) dogs without arthritis were infected with a *Bartonella* species. The prevalence of *Bartonella* spp. DNA in the dogs with or without arthritis did not differ (χ^2 test for independence, $P = 0.589$). These results indicate that a subset of dogs with CanL is co-infected with various *Bartonella* species, including *B. henselae*, *B. rochalimae* and *Candidatus Bartonella merieuxii*. Future studies are warranted to determine if *Bartonella* spp. infections contribute to the occurrence of *L. infantum*-associated arthritis or other clinical manifestations.

Conflicts of interest: No conflicts of interest reported.

ISCAID-O-9

RISK FACTORS OF GIARDIA INFECTION AND PATHOGENICITY IN WEANING PUPPIES. A. Grellet¹, H. Mila², M.A. Debouchaud², A. Feugier¹, S. Chastant-Maillard². ¹Royal Canin, Aimargues, France, ²Ecole Nationale Vétérinaire de Toulouse, Toulouse, France

The prevalence of *Giardia* in dogs ranges between 5.4% and 55.2%, with a higher prevalence in puppies. However, the risk factors for *Giardia* infection around weaning have been poorly described. The aim of the study was to evaluate risk factors for *Giardia* infection in puppies during the first weeks of life and to determine an impact of this parasite on feces quality.

192 puppies from 58 litters living in a breeding kennel were followed between 4 and 9 weeks of age. Each puppy was treated with fenbendazole (Panacur[®], MSD, France, 50 mg/kg, per os, q 24 h) for 3 consecutive days at 2, 4, 6 and 8 weeks of age. For each puppy, fecal consistency was evaluated using a 13-point scale. Excretion of enteropathogens was evaluated by qPCR for canine parvovirus type 2 (CPV2), qRT-PCR for canine coronavirus (CCV), coproantigens quantification for *Giardia* (ProSpecT-Giardia, Remel), and McMaster flotation technique for any eggs and oocysts. A generalized linear mixed model (proc GLIMMIX) with *Giardia* infection as a binary outcome was used to assess the following effects: breed size, age, and CPV2, CCV and *Iso spor a ohioensis* infections. A linear mixed model (proc MIXED) with fecal score as outcome was used to determine the following effects: breed size, age, and *Giardia*, CPV2, CCV and *I. ohioensis* infections.

A total of 277 fecal samples were collected; CPV2, *Giardia*, *I. ohioensis* and CCV were detected in respectively 13.4%, 17%, 30.3% and 67.9% of the samples. The risk of *Giardia* infection increased with age (Odd Ratio= 1.5; 95%CI=1.1-2.1; $p = 0.015$). Neither breed, nor CPV2, CCV and *I. ohioensis* infections influenced risk of *Giardia* infection ($p = 0.299$; $p = 0.213$; $p = 0.892$; $p = 0.282$ respectively). *Giardia* infection did not impact feces quality ($p = 0.11$), whereas a significant influence of CPV2 ($p < 0.001$), CCV infection ($p = 0.030$) and breed size ($p < 0.001$) was evidenced.

This study underlines that even with an adapted deworming program the eradication of *Giardia* is difficult to obtain in large dog packs. The higher prevalence of *Giardia* in puppies of 7 weeks and older could be linked with the immunity gap during this period. *Giardia* was not associated in our study with an increased risk of diarrhea. The lack of pathogenicity of the parasite per se could be hypothesized, but also an efficacy of the treatment for the prevention of the clinical signs or a local and systemic immunity limiting clinical signs.

Conflicts of interest: Financial support from Royal canin.

SCH-O-1

MAGNETIC RESONANCE SPECTROSCOPY (MRS) IN CANINE HEPATIC ENCEPHALOPATHY DIAGNOSIS AND MONITORING. M. Dolera, L. Malfassi, S. Marcarini. La Cittadina Fondazione Studi e Ricerche Veterinarie, Romanengo, cr, Italy

Hepatic Encephalopathy (HE) can occur in dogs as a complication of primary liver disease or as consequence of congenital portosystemic shunts (PSS). The aim of this study was to assess Magnetic Resonance Spectroscopy (MRS) usefulness in HE diagnosis.

Twenty dogs with a presumptive diagnosis of HE were enrolled. Inclusion criteria were: a clinical examination, a blood cell count and biochemical panel, at least one plasmatic ammonia determination higher than 100 $\mu\text{mol/L}$, optional histopathology of the liver, requirement of Magnetic Resonance Imaging (MRI) investigation by the clinician. Even though MRS represents a non-invasive procedure, informed consent was obtained from dogs' owners. HE diagnosis was confirmed in 20/20: 16/20 had a PSS, 3/20 showed hepatic microvascular dysplasia and 1/20 had hepatic cirrhosis histologically confirmed. The control-group was made of 23 patients retrieved from our database that underwent MRI without showing any abnormalities on brain scans. The MR protocol included T2-weighted fast spin-echo, T1-weighted sequences, Proton Density, and Radio-frequency pulses Gradient-Echo. On MRI examination cerebral atrophy was evident in all the patients (20/20), ranged from mild to severe. In 17/20 patients symmetrical bilateral hyperintensities in the globus pallidus on T1-weighted images were evident. MRS was performed using a short TE (30 ms) and it was defined a volume of interest in the parieto-occipital region by 10-mm side cube (1 cc of volume). Glutamine-glutamate (*Gln*), *N*-acetylaspartate and *N*-acetyl aspartyl glutamate (*NAA*), Choline derivatives (*Cho*), and Myo-Inositol (*Ins*) were analyzed.

Comparing spectra from the HE affected dogs with those from the control-group decreased values of *Ins*, *Cho*, *NAA* as well as increased values of *Gln* were found in the HE dogs. Mean values and connected standard deviations are reported: *Gln/Cr* 0.40 std. 0.13; *Cho/Cr* 1.14 std. 0.34; *Ins/Cr* 0.15 std. 0.08; *NAA/Cr* 1.02 std. 0.35. From the cohort of 20 dogs with confirmed HE, 15 patients that underwent medical therapy for HE had a complete MRI and MRS follow-up examination 3 months later. At the follow-up MRS, the *Gln* peak was found decreased in 15/15 patients and values of *Ins* and *Cho* increased in 14/15.

High field MRI combined with brain MRS provide accurate and non-invasive diagnosis of canine HE. In accordance with human medicine publications, it could be state that MRS has a role in HE diagnosis and follow-up with particular mention monitoring.

Conflicts of interest: No conflicts of interest reported.

SCH-O-2

DIAGNOSTIC COMPARISON OF NEEDLE AND WEDGE LAPAROSCOPIC BIOPSIES IN DOGS: 22 CASES. A. Lecoindre¹, J.L. Cadore¹, M. Chevallier², P. Lecoindre³. ¹Vet Agro Sup, Marcy l'etoile, France, ²Biomnis, Lyon, France, ³CVC veterinary clinic, Lyon, France

Diagnosis of hepatobiliary diseases often requires hepatic tissue sampling for histologic evaluation. The laparoscopic technique is a safe method and allows acquisition of tissue by wedge and needle biopsies from different liver lobes in a minimally invasive way. Specimens obtained with an 18-gauge biopsy needle must be interpreted with caution due to considerable variability in tissue involvement with certain disease processes. We hypothesized that needle biopsy specimens would produce findings divergent from those produced by wedge biopsy specimens.

The goal of the study was to compare histological findings from two laparoscopic biopsy methods (wedge and needle) and assess which sampling technic can represent the overall disease process.

Procedure: All dogs included in this prospective study were suspected diffuse hepatic disease and underwent laparo-

scopic hepatic biopsy (wedge and needle 14-16G) between 2012 and 2014. All biopsy specimens were examined on the basis of morphologic criteria and a comparison was made between the two types of biopsies procedures according to WSAVA Liver Standardization Group morphologic criteria.

Results: Twenty-two dogs were included. No complications were reported during the laparoscopic procedure. The median number of portal triads per needle biopsy specimen was 6 (range, 4 to 8) compared to 20 (range 14;25) with wedge biopsy specimen. The median length of needle biopsy specimens was 10 mm; (range, 6 to 16 mm) and 6 mm for all wedge biopsies. On the basis of biopsy interpretation, the diagnosis was overall similar with the two methods: 8 dogs had vacuolar hepatopathy, 4 acute cholangitis, non-specific acute form in 3 dogs. Chronic hepatitis with cirrhosis was found in 2 cases, 3 dogs had diffuse neoplasia and 2 miscellaneous hepatic disorders. The fibrosis was considered to be severe in 3 dogs, moderate in 3 dogs and mild in 11 dogs. No quantitative and qualitative difference was observed between the two types of biopsies specimen.

This study demonstrates that the biopsies with a needle length of at least 10 mm brings satisfactory information for the evaluation of most of the inflammatory, vacuolar hepatopathies, fibrosis and diffuse tumoral infiltrations. Wedge biopsies allow to examine the largest number of portal triad, more contributory for certain forms of cholangitis affecting larger canals and for a single case, images of peri-hepatitis were counted at the level of the capsule. Fibrosis does not seem to be more important in the sub-capsular zone contrary to what is observed in human pathology.

Conflicts of interest: No conflicts of interest reported.

SCH-O-3

A PILOT STUDY TO ASSESS THE FEASIBILITY OF TRANSCUTANEOUS INDOCYANINE GREEN CLEARANCE AS LIVER FUNCTION TEST IN CATS AND DOGS. S. Strommer¹, S. Steinbach¹, N. Krollop¹, Z. Herrera-Pérez², N. Gretz², R. Neiger¹. ¹Small Animal Clinic (Internal Medicine), Justus-Liebig University, Giessen, Germany, ²Medical Research Center, University of Heidelberg, Mannheim, Germany

Indocyanine green (ICG), a fluorescence dye, is excreted solely by the liver without enterohepatic re-circulation. Hence it has been used for decades as an ideal albeit invasive marker of hepatic function and blood flow in cats and dogs. Here we evaluated the feasibility of a minimally invasive transcutaneous ICG clearance to assess hepatic function instantaneously.

Transcutaneous ICG clearance was performed in 3 healthy research cats and 3 healthy research dogs with normal liver function (bile acid stimulation test, ammonia tolerance test) using a modified device (Kidney Int 2011 79:1254-8) with an excitation wave length of 760 nm and an emission wave length of 820 nm. The devices were placed on different locations (lateral thoracic wall, ventrolateral abdomen, metatarsus and antebrachium) and fixed with a light bandage. To find a suitable dose to reach adequate transcutaneous peak concentrations escalating doses of ICG (0.1, 0.2 and 0.3 mg/kg) were injected intravenously with a wash out period of at least 24 h in-between. Measurement was continued for 1 hour after injection in awake animals moving freely. The resulting ICG disappearance curves were visually inspected to find best location (minimal artifacts, acceptable background noise) and dose.

In all animals a dose of 0.2 mg/kg was deemed ideal and the thoracic and abdominal wall gave consistent results. Half-life of ICG clearance was calculated using a one-compartment model. Half-lives in cats were 4.99, 5.23 and 7.19 minutes, respectively; in dogs: 9.66, 12.51 and 15.58 minutes, respectively.

In conclusion, the transcutaneous assessment of ICG clearance is feasible in a clinical setting. Results are obtained within one hour and can be assessed instantaneously. The procedures are minimally invasive and well tolerated by the animals. Given that most patients with a presumed liver problem undergo abdominal ultrasound no further clipping of hair is necessary as the device might be placed in this area. Further studies are necessary to obtain reference values in healthy pets and those with various conditions leading to impaired hepatic function.

Conflicts of interest: No conflicts of interest reported.

SCH-O-4

EVALUATION OF PREPRANDIAL AND POSTPRANDIAL GALLBLADDER VOLUME USING THREE-DIMENSIONAL ULTRASONOGRAPHY IN HEALTHY DOGS. V. Rahmani¹, S. Jamshidi¹, M. Molazem¹, Y. Vali¹, M. Hanifeh². ¹Faculty of Veterinary Medicine, University of Tehran, Tehran, Iran, ²Faculty of Veterinary Medicine, University of Tabriz, Tabriz, Iran

Gallbladder diseases like gallbladder mucocele and cholecystitis can reduce gallbladder motility and may lead to cholestasis. Since impaired gallbladder emptying contributes to sludge and gallstone formation, the evaluation of gallbladder motility requires accurate and appropriate methodology. Three-dimensional (3D) ultrasonography has been shown to be accurate and appropriate tool for measurement of gallbladder volume in humans. Therefore, we applied this novel technique for the first time to study preprandial and postprandial gallbladder volume in 10 healthy mixed-breed dogs and compared the results to two-dimensional (2D) ultrasonography. The dogs were placed in dorsal recumbency to obtain ultrasonographic measurements of the gallbladder. Measurements by both 2D and 3D ultrasonography were recorded in preprandial state and after ingestion of full-fat milk. The preprandial and postprandial gallbladder volumes determined by 3D ultrasonography were significantly higher than corresponding volumes by 2D ultrasonography (1.11 ± 0.07 vs 0.77 ± 0.06 and 0.81 vs 0.61 ml/kg, respectively, $P < 0.05$). In 2D ultrasonography, most dogs (8/10 [80%]) had a preprandial gallbladder volume ≤ 1.00 ml/kg. However, in 3D ultrasonography, 6/10 (60%) of dogs had a preprandial gallbladder volume ≥ 1.00 ml/kg. Gallbladder contraction index was higher in 3D ultrasonography than 2D ultrasonography, however, it did not reach statistical significance ($P = 0.25$).

In conclusion, 3D ultrasonography showed larger gallbladder volumes than 2D ultrasonography in healthy dogs. It seems that 3D ultrasonography is appropriate adjunct device to 2D ultrasonography to estimate gallbladder volume when 2D ultrasonography could not detect whole gallbladder volume. More research is needed to determine clinical value of 3D ultrasonography in canine gallbladder imaging.

Conflicts of interest: No conflicts of interest reported.

VBPS-O-1

COMPARISON OF DIRECT AND INDIRECT BLOOD PRESSURE MEASUREMENTS IN CONSCIOUS BEAGLES. F. Manczur¹, N. Kubik¹, I. Novák². ¹SZIE, Faculty of Vet Science, Budapest, Hungary, ²ATRC Aurigon Toxi-Coop Ltd., Budapest, Hungary

The aim of our study was to compare high-definition oscillometry (HDO) and Doppler ultrasonographic measurements with direct blood pressure measurements in conscious dogs.

The Doppler study was performed by three investigators and by using 3 different sphygmomanometers with 3 different sized cuffs. Devices and measurement sites were changed randomly among the investigators. Cuffs were wrapped around the antebrachium in the forelimb or around the mid-metatarsus in the hind limb. In addition to the limb sites, cuffs were also placed around the base of the tail in case of the HDO method. Cuff sizes were 45-78% of the measured limb circumferences during the Doppler measurement. For the HDO method all measurements were performed by the same investigator and the cuffs provided by the manufacturer were used: the smallest cuff was used for the hind limb and tail, and the medium cuff was used for the forelimb measurements. Dogs were gently held in lateral recumbent position, measurement were performed on the nondependent limbs. Radio-telemetry transducers were implanted to the right femoral artery some months to a year preceding the blood pressure measurements for reasons unrelated to our study. Direct blood pressures varied 110-214 mmHg and 139-192 mmHg during the Doppler and HDO measurements, respectively.

Two-hundred paired simultaneous Doppler and direct measurements from 11 dogs and 100 paired simultaneous HDO and direct telemetric measurements from 7 dogs were obtained. At least 3 successful consecutive measurements could be obtained by

the same investigator at the same site during the Doppler or HDO measurement in 53 and 22 cases, respectively. Thus, the mean of these measurements could be calculated similarly to the established everyday clinical practice.

Bias (mean difference), precision (standard deviation) and limits of agreements were calculated both from the individual paired measurements and from the means of the consecutive measurements using Bland-Altman spot analysis.

Systolic measurement performed on the tail with the HDO-method yielded the smallest bias and deviation and the best limits of agreement during this study. Both Doppler and HDO-measurements performed on the forelimb overestimated, while hind limb measurement underestimated the direct telemetric pressures. Results of all three measurement sites by HDO performed better than forelimb or hind limb Doppler-measurements, however HDO-measurements were more difficult to obtain and more often resulted with measurement failure compared to the Doppler technique. Cuff size above 55% of the measured limb circumference showed better results than smaller cuff sizes during the Doppler measurements.

Conflicts of interest: No conflicts of interest reported.

VBPS-O-2

SYSTOLIC ARTERIAL PRESSURE MEASURED SIMULTANEOUSLY BY DOPPLER TECHNIQUE, USING FORELIMB AND HINDLIMB, IN DOGS. S. Crosara¹, A. Borrelli², F. Rondato², C. Quintavalla¹, G. Faranda², A. Tarducci², R. Zanatta². ¹Dep. Veterinary Science, Parma, Italy, ²Dep. Veterinary Science, University of Turin, Italy

The Doppler technique is considered the most repeatable indirect method to measure systolic arterial pressure (SAP) in dogs. However, recent studies emphasized the effect of body position and used limb on SAP measurement. The aim of this study was to determine whether a difference existed in SAP measured simultaneously in dogs using different limbs, with two Doppler units by two different operators. Sixty client-owned dogs, admitted to the veterinary hospital for different reason, were enrolled. They were divided in 3 groups based on body size: 20 small breed dogs (<15 kg); 20 medium breed (15-30 kg); 20 large breed (> 30 kg). For each dog the anxiety status was recorded. SAP was measured via Doppler technique when dogs were in right lateral recumbency in a quite environment. Right and left forelimb SAP and left forelimb and left hindlimb SAP were recorded simultaneously, with two identical Doppler units equipped with headphones, by two operators. Measurement was performed based on the ACVIM guidelines. Five measurements were recorded, the higher and lower values were discarded from the analysis. The relationship of mean SAP for each limb with body weight, sex, anxiety status and SAP value was evaluated. Mean \pm SD SAP was significantly higher for the right forelimb (175.81 ± 37.15) compare to the left forelimb (165.14 ± 33.35) on overall population. The difference was significant for large breed dogs, males and dogs with SAP ≥ 180 mmHg. SAP was higher for the left forelimb (163.67 ± 32.11) compare to the left hindlimb (151.34 ± 34.31) on overall population. The difference was significant for medium and large breed dogs, females, calm animals and dogs with SAP ≥ 180 mmHg. The mean SAP from the left forelimb recorded by two different operators at two different moments, were compared and no difference was evident. In conclusion, SAP measurement from different limbs, in dogs in right lateral recumbency, is poorly correlated. Measurement of SAP from the left forelimb is more repeatable during time and between different operators. SAP trend monitoring should be done using the same measurement site for any animal.

Conflicts of interest: No conflicts of interest reported.

VBPS-O-3

EFFICACY AND CLINICAL SAFETY OF A NEW PALATABLE FORMULATION OF AMLODIPINE IN THE TREATMENT OF HYPERTENSIVE CATS. M. Huhtinen¹, G. Derré², H.J. Renoldi³, M. Rinkinen⁴, K. Adler⁵, J. Aspegren¹, J. Elliott⁶. ¹Orion Corporation, Orion Pharma, Turku, Finland, ²Clinique Vétérinaire de la Plage, Marseille, France, ³Tierarztpraxis Renoldi, Neuwied, Germany, ⁴MEVET Eläinlääkäriasema, Helsinki, Finland, ⁵Klifovet AG, Munich, Germany, ⁶The Royal Veterinary College, London, United Kingdom

Amlodipine has been considered the treatment of choice for hypertension in cats for more than a decade. There is, however, an unmet need for a cat-specific formulation. The aim of the study was to assess the efficacy of chewable amlodipine tablets in reducing systolic blood pressure (SBP) in cats diagnosed with hypertension.

Seventy-seven client-owned cats were included in the study (mean age 14 years). The study was randomised, double-blind, placebo controlled, and consisted of two phases. In the blinded phase, 42 cats received 0.125 mg/kg amlodipine once daily for 14 days. If they responded the dose remained the same to day 28. For non-responders, the dose was increased to 0.25 mg/kg. Thirty-five cats received placebo following the same protocol. Arterial blood pressure was measured using a high definition oscillometry method. At day 28 a responder was defined as a cat showing a decrease of SBP to ≤ 150 mmHg or a decrease from baseline of at least 15%. After 28 days all cats continued with amlodipine for 2-3 months in an open phase with the placebo cats repeating the same dose escalation protocol as in the blinded phase.

The responder rate was 63% in the amlodipine group and 18% in the placebo group following the dose escalation from day 14 being applied to 54% and 80% of cats receiving amlodipine and placebo respectively. Cats receiving amlodipine were 7.9 (95% CI 2.6 to 24.1) times more likely to be classified as responders when compared to those receiving placebo (logistic regression model, $p = 0.0003$). From a baseline value of 181.6 ± 12.5 and 179.3 ± 10.8 mmHg the mean SBP decreased to 153.6 ± 16.9 mmHg with amlodipine and to 167.7 ± 20.5 mmHg with placebo (repeated measures analysis of covariance model, $p < 0.001$) by day 28. The responder rate was not influenced by factors other than amlodipine treatment (e.g. baseline blood pressure, concomitant ACE inhibitor therapy, renal disease). There were no differences between the amlodipine and placebo groups in the frequency of adverse events reported during the 28-day blinded phase. Likewise, there were very few changes in the laboratory values over time in either group.

The present study is the first large clinical trial to show that amlodipine is clearly superior to placebo in the treatment of cats with hypertension. The chewable amlodipine formulation effectively reduced SBP, had a good palatability and was well tolerated. It can be used concomitantly with ACE inhibitors and in cats with renal disease.

Conflicts of interest: M. Huhtinen and J. Aspegren are employees of the sponsor

J. Elliott has the following information to disclose: Consultancy: Pfizer Animal Health / Zoetis, CEVA Animal Health, Boehringer Ingelheim, Vetoquinol Ltd, Orion Corp., Elanco Ltd, Idexx Ltd, Niche Generics Ltd., Triveristas Ltd., Virbac Ltd. Advisory board membership: International Renal Interest Society (supported by Novartis), European Emesis Council (sponsored by Pfizer Animal Health - now Zoetis), Cardiorenal Board - Vetoquinol Ltd., Idexx Renal Advisory Board. Research Grants or contracts: Vetoquinol Ltd, Novartis Ltd, Pfizer Animal Health Ltd (now Zoetis), Royal Canin Ltd, Boehringer Ingelheim Ltd, Waltham Centre for Pet Nutrition, CEVA Animal Health, Orion Corp.

VBPS-O-4

PHARMACODYNAMIC AND PHARMACOKINETIC MODELING OF AMLODIPINE IN FELINE HYPERTENSIVE PATIENTS. J. Elliott¹, L. Pelligand¹, M. Huhtinen². ¹Royal Veterinary College, London, United Kingdom, ²Orion Corporation, Orion Pharma, Turku, Finland

Amlodipine is the treatment of choice for feline hypertension. Limited published data exist on serum concentrations achieved in

hypertensive cats. The aim of the study was to assess serum amlodipine concentrations in cats treated with a new formulation of amlodipine and relate these to the blood pressure reduction achieved.

Seventy-seven client-owned hypertensive cats were enrolled into a randomized, double-blind and placebo controlled study consisting of two phases. In phase one, 42 cats (Group A) received 0.125 mg/kg amlodipine once daily for 14 days. If they were deemed to have responded (see below) the dose remained the same to day 28. For non-responders, the dose was increased to 0.25 mg/kg. Thirty-five cats (Group B) received placebo following the same protocol. Blood pressure was measured using high definition oscillometry. A responder was defined as a cat showing a decrease of systolic blood pressure (SBP) to ≤ 150 mmHg or a decrease from baseline of $\geq 15\%$. Following day 28 (phase 2), Group A continued on amlodipine and Group B switched to amlodipine and the dose was adjusted as per phase 1. Both groups were followed for 90 days on amlodipine. Blood was collected at days 28 (Group A) and 90 (both Groups) and serum [amlodipine] measured by liquid chromatography mass spectrometry. The SBP measured on treatment was calculated as percentage of the baseline SBP and plotted against serum [amlodipine] using a sigmoidal Emax model (WinNonLin software). Data are expressed as mean \pm SE.

The serum concentrations of Group A cats that remained on 0.125 mg/kg were 29.8 ± 2.5 ng/ml whereas those switched to 0.25 mg/kg were 51.2 ± 7.8 ng/ml. When data from groups A and B were pooled, a sigmoidal relationship between percentage baseline SBP and serum [amlodipine] was found. Estimated values of lowest percentage baseline blood pressure on treatment (Emax) was $83.1 \pm 1.7\%$, with an EC_{50} value of 10.4 ± 2.6 ng/ml and a slope function of 2.5 ± 1.2 . The serum concentration required to reduce blood pressure by 15% was estimated to be 20 ng/ml.

The present study related blood pressure reduction to serum [amlodipine] in feline clinical hypertension. The limitations of this study were the limited number of blood samples collected and lack of information relating to the exact timing of blood sampling relative to dosing in some cats. However, these data could be used to define appropriate therapeutic serum [amlodipine] in hypertensive cats.

Conflicts of interest: M Huhtinen is an employee of the sponsor; J. Elliott has the following information to disclose: Consultancy: Pfizer Animal Health / Zoetis, CEVA Animal Health, Boehringer Ingelheim, Vetoquinol Ltd, Orion Ltd., Elanco Ltd, Idexx Ltd, Niche Generics Ltd, Triveristas Ltd., Virbac Ltd., Advisory board membership: International Renal Interest Society (supported by Novartis) European Emesis Council (sponsored by Pfizer Animal Health - now Zoetis) Cardiorenal Board - Vetoquinol Ltd, Idexx Renal Advisory Board Research Grants or contracts: Vetoquinol Ltd, Novartis Ltd, Pfizer Animal Health Ltd (now Zoetis), Royal Canin Ltd, Boehringer Ingelheim Ltd, Waltham Centre for Pet Nutrition, CEVA Animal Health Orion Ltd.; L Pelligand has the following information to disclose:

In receipt of research grant / contract funding from Orion Ltd., Novartis Animal Health, Transpharmation Ltd, DeltaDOT Ltd; Acted as a consultant for: Triveritas Ltd. and Novartis Animal Health.

POSTERS

ESCG-P-1

ANALYTICAL COMPARISON OF NEWLY INTRODUCED COMMERCIAL ASSAYS FOR THE MEASUREMENT OF CANINE AND FELINE PANCREATIC LIPASE IMMUNOREACTIVITY (cPLI AND fPLI) TO ESTABLISHED ASSAYS. E. Höfel¹, T. Rieker¹, J.S.Suchodolski², J.M. Steiner². ¹Klinik am Hochberg, Ravensburg, Germany, ²Texas A&M University, College station, United States of America

Commercial assays for the measurement of canine and feline pancreatic lipase immunoreactivity (Spec cPL[®] and Spec fPL[®], respectively; Idexx Laboratories, Westbrook, ME, USA) have been available for a few years and have previously been analytically and clinically validated. Recently, new commercial assays for the measurement of these parameters have become available, though neither one of these assays have been analytically or clinically validated in the literature. Thus, the goal of this

study was to compare these newly available assays to the established assays.

Leftover serum samples from diagnostic submissions to the GI Laboratory were collected based on certain parameters (e.g., results throughout the working range of the assay, good quality sample or hemolytic, lipemic, or icteric sample) and were assigned random sample ID numbers. The samples were evaluated by Spec cPL[®] or Spec fPL[®], sent on dry ice to the Klinik am Hochberg, and one aliquot of each sample was blindly submitted to Laboklin for measurement of cPLI and fPLI by their newly released in-house assay and also to the GI Lab at Texas A&M University for repeated analysis by Spec cPL[®] and Spec fPL[®] to exclude any effect of shipping.

There was no significant difference between serum cPLI or fPLI concentrations before or after shipping at the GI Lab (p-values for Wilcoxon matched-pairs signed rank tests: 0.593 and 0.672, respectively). In contrast, there was a significant difference between serum cPLI or fPLI concentrations between the newly released assays and the previously established assays (p-values < 0.0001 and 0.0241, respectively). While there was a significant correlation between the newly released and the previously released assays (Spearman r: 0.775 and 0.7386, respectively), this correlation was very poor for assays that supposedly measure the same analyte. Also, the interpretation for serum cPLI and fPLI results between the previously developed assays and the new assays did not agree for many of the samples. Finally, both newly developed assays showed some erratic results.

In conclusion, the newly released assays for the measurement of cPLI and fPLI do not agree with previously established and validated assays, provide different interpretations, and show erratic results. Thus, further research is needed before these newly released assays could be recommended for clinical use.

Conflicts of interest: Dr. Steiner serves as Director and Dr. Suchodolski serves as Associate Director of the Gastrointestinal Laboratory at Texas A&M University. Dr. Steiner also serves as a paid consultant to Idexx Laboratories, Westbrook, ME, USA. Both the Gastrointestinal Laboratory and Idexx Laboratories offer cPLI and fPLI testing on a fee-for-service basis.

ESCG-P-2

EFFECT OF AGE, BREED SIZE AND ENTEROPATHOGEN INFECTION ON FECAL IMMUNOGLOBULIN A CONCENTRATIONS IN WEANING PUPPIES. A. Grellet¹, H. Mila², A. Feugier¹, R. Lopes³, N. Gruetzner³, J.S. Suchodolski³, J.M. Steiner², S. Chastant-Maillard². ¹Royal Canin, Aimargues, France, ²Ecole Nationale Vétérinaire de Toulouse, Toulouse, France, ³Gastrointestinal Laboratory, Texas A&M University, College station, United States of America

Digestive health is a main concern for growth, morbidity and mortality in weaning puppies. Fecal immunoglobulin A (IgA) has been suggested as a useful noninvasive biomarker for mucosal immunity. The purpose of this study was to evaluate the effect of infection with enteropathogens on fecal IgA concentrations in puppies and that of physiological factors such as age and breed size.

282 puppies from 33 breeding kennels were included in the study. Puppies were between 5 and 14 weeks of age (mean \pm Standard Deviation (SD): 7.8 ± 1.5 weeks). Depending on the mean adult body weight of their respective breed, the puppies were divided into small (if mean adult body weight < 25 kg) or large (>25 kg) breed puppies. For each puppy, fecal consistency was evaluated using a 13-point scale and feces were collected for the evaluation of presence of fecal enteropathogens and fecal IgA concentrations. The presence of enteropathogens in fecal samples was evaluated by qPCR for canine parvovirus type 2 (CPV2), qRT-PCR for canine coronavirus (CCV), coproantigen quantification for Giardia (ProSpecT-Giardia, Remel), and McMaster flotation technique for other parasite eggs and oocysts. Fecal IgA concentrations were measured by an ELISA test. Statistical analyses were performed using SAS software. A linear mixed model (proc MIXED) with fecal IgA concentration as outcome was used to determine the following effects: enteropathogen infection, breed size, age, and fecal score. The respective influence of litter and breeding kennel as random effects was also determined. Data is presented as mean \pm SD.

Small breed dogs represented 27.3% (77/282) of the total number of dogs included. At least one enteropathogen was identified in 76.2% of puppies (214/281). Fecal IgA concentration was significantly influenced by fecal enteropathogens ($p = 0.037$). Puppies infected with at least one enteropathogen had significantly lower fecal IgA concentrations than puppies without any enteropathogens ($5.0 \pm 4.4 \mu\text{g/g}$ vs. $6.9 \pm 5.5 \mu\text{g/g}$). Breed ($p = 0.029$), but not age ($p = 0.082$), influenced IgA concentration. Small breed puppies had significantly higher fecal IgA concentrations than large breed puppies ($6.8 \pm 4.8 \mu\text{g/g}$ vs. $5.0 \pm 4.7 \mu\text{g/g}$). No significant relationship between fecal IgA concentration and feces quality was evidenced ($p = 0.165$).

This study suggests that fecal IgA concentration is a promising marker for subclinical infection by at least one enteropathogen and confirms that digestive physiology varies with the breed size. A link between lower digestive immunity and higher susceptibility to enteropathogen infection needs further investigation.

Conflicts of interest: Financial support of Royal Canin.

ESCG-P-3

A VERY LOW MOLECULAR WEIGHT POULTRY FEATHER HYDROLYZED-BASED EXTRUDED DIET ALLOWS IDIOPATHIC CANINE IBD CLINICAL MANAGEMENT WITHOUT IMMUNOSUPPRESSIVE TREATMENT: A 13 CASE PROSPECTIVE PILOT STUDY. V. Freiche¹, A. Leclerc², O. Dossin³, J. Dahan³, O. Toulza⁴, I. Mougeot⁵, V. Biourge⁶. ¹Ecole Nationale Vétérinaire d'Alfort, Maisons alfort, France, ²Hôpital Vétérinaire Daubigny, Québec, Canada, ³Ecole Nationale Vétérinaire de Toulouse, Toulouse, France, ⁴Clinique Aquivet, Eysines, France, ⁵Royal Canin Canada, Guelph, Canada, ⁶Royal Canin SAS, Aimargues, France

Canine chronic enteropathies (CCE) include diet-responsive, antibiotic-responsive, and immunosuppressive-responsive enteropathies (IRE). This prospective study was designed to evaluate a commercial hypoallergenic dry diet^a containing oligopeptides as the only protein source for the management of dogs with IRE and as an alternative to immunosuppressive therapy over a 10 week period.

Nineteen dogs across France and Quebec entered the study. Dogs with food or antibiotic-responsive chronic enteropathy, hypoproteinemia, or treated with immunomodulating drugs were excluded from the study. Dogs were included in the study after complete clinical, ultrasonographic, endoscopic evaluation and histopathological evaluation of intestinal biopsies showing signs of intestinal inflammation. The owners were instructed to feed exclusively the study diet^a.

Canine Inflammatory Bowel Disease Activity Index (CIBDAI) scores, fecal scores as observed by the dog-owners, and body weight were evaluated at baseline, 2, 5 and 10 weeks after inclusion. Dietary treatment was regarded successful if the CIBDAI score was reduced by at least 75%. The protocol has been reviewed and accepted by Royal Canin ethics committee and owners completed an informed consent. Results are presented as mean \pm SD (range). Statistical comparisons were performed with a Wilcoxon test.

Thirteen dogs (7 intact males, 4 neutered and 2 intact females) completed the trial. Seven dogs were excluded (2 diagnosed with *Giardia*, 4 s with no histological evidence of inflammation, 1 with hypoadrenocorticism). Mean age was 4.4 years \pm 3.01 (1.1 - 11.2), mean body weight 16.2 kg \pm 13.36 (2.5 - 43.0). CIBDAI score was 9.4 \pm 3.43 (4-16) at inclusion, was 2.7 \pm 1.84 (0-6) after 5 weeks and was 1.5 \pm 1.31(0-3) after 10 weeks ($p = 0.0015$ and $p = 0.0022$ vs inclusion, respectively). Fecal scores after 5 [4.0 \pm 0.58 (3-5)] and 10 weeks [4.0 \pm 0.60 (3-5)] were improved compared to inclusion scores 1.9 \pm 1.12 (1-4) ($p = 0.0033$ and $p = 0.0051$, respectively).

The low molecular weight poultry feather hydrolyzed protein-based dry extruded diet^a appears to be effective in the management of idiopathic IBD without any concurrent immunosuppressive drug over the 10 week period of this pilot study. These preliminary findings should be confirmed by a prospective, randomized double blind study.

^a ANALLERGENIC[TRADEMARK] Canine Formula, Royal Canin Veterinary Diets[TRADEMARK], Aimargues, France

Conflicts of interest: Drs I.Mougeot and V.Biourge are Royal Canin SAS associates.

ESCG-P-4

ESTABLISHMENT OF SEVERITY SCORING SYSTEM FOR OUTCOME PREDICTION IN CATS WITH PANCREATITIS. S.Y. Wang, Y.J. Lee, B.L. Su. Institute of Veterinary Clinical Sciences, National Taiwan University, Taipei, Taiwan

Feline pancreatitis is the most common exocrine pancreatic disorder with varied mortality. However, there is no available and reliable method to evaluate the severity and prognosis of the disease. Ninety-two cats diagnosed as pancreatitis with acute onset of compatible clinical signs and a positive SNAP[®] fPL[™] Test between October 2011 and September 2013 were enrolled in this study. All Cats were divided into survival ($n = 48$) and non-survival ($n = 44$) groups. Fifty-two parameters including signalments, clinical signs, physical examinations, clinicopathological examinations, diagnostic images, complications and concurrent diseases were analyzed and compared between the two groups. Parameters with $P \leq 0.05$ were considered for further analyses. The mortality in this study was 47.8%. Hematocrit, albumin, BUN, creatinine, total bilirubin, calcium, phosphorous, body temperature, systolic blood pressure, the body cavity fluids, complications, e.g. systemic inflammatory response syndrome (SIRS) and acute renal failure (ARF) were found to be significantly associated with disease severity and prognosis, and were selected for constructing the scores. Continuous variables outside the reference interval were separated into quartiles to yield quartile-specific odds ratios (ORs) for survival. Based on the integer value of the OR, the scoring system was then developed by incorporating weighting factors assigned to each quartile. A predictive total score was calculated for each cat by summing all weighting factors. The total scores of each cat ranged from 12 to 83. The severity scores in this study achieved an area under receiver operating characteristic (AUROC) of 0.88. The optimal cut-off point for discriminating outcome was 32.5 with the sensitivity of 89.6% and specificity of 77.3%, respectively. The mortality was 87.2% with a score ≥ 33 , whereas 18.9% with a score ≤ 32 . There was significant difference ($P < 0.001$) between the two groups of the cut-off point. Furthermore, the mortality reached to 100% when the score more than 56. The severity scoring system of this study provides a reliable and clinical applicable method to predict clinical outcome in cats with pancreatitis.

Conflicts of interest: No conflicts of interest reported.

ESCG-P-5

GENOTYPING OF CLOSTRIDIUM PERFRINGENS ISOLATES FROM EIGHT DOGS WITH ACUTE HAEMORRHAGIC DIARRHOEA SYNDROME. S. Unterer¹, K. Busch¹, J. Verspoh², G. Wolf³, R.K. Straubinger³, K. Hartmann¹. ¹LMU, Munich, Germany, ²Institute for Microbiology, Department of Infectious Diseases, Hannover, Germany, ³Institute for Infectious Diseases and Zoonoses, Munich, Germany

Convincing evidence for the role of *Clostridium (C.) perfringens* as a primary pathogen in acute haemorrhagic diarrhoea syndrome (AHDS) in dogs was recently found. It is suspected that clostridial toxins, especially *C. perfringens* enterotoxin, play a relevant role in the disease process. However, to date enterotoxigenic *C. perfringens* strains have only been described in single case reports. Thus, the aim of this study was to identify the specific *C. perfringens* genotype involved in AHDS.

Small intestinal biopsies were collected with a sterile single-use biopsy forceps from ten dogs with AHDS and immediately cultured. In 8/10 dogs, clostridial strains were isolated and identified as *C. perfringens* by mass spectrometry using MALDI-TOF MS. *C. perfringens* colonies from each dog were submitted for specific detection of the four major toxin genes (alpha, beta, epsilon, and iota), the enterotoxin gene, and the beta2 toxin by multiplex PCR.

Every clostridial isolate was typed as *C. perfringens* type A based on the detection of the alpha toxin encoding gene. In 5/8 isolates, additionally the beta2 toxin gene was identified, however, none of clostridial strains encoded for the *C. perfringens* enterotoxin gene.

The results of this study suggest that *C. perfringens* type A is the most important *C. perfringens* genotype involved in the disease process of dogs with AHDS. Although *C. perfringens* enterotoxin has been associated with intestinal diseases in humans, dogs, horses, pigs, and other animal species, this enterotoxin is most likely not responsible for the intestinal lesions in dogs with AHDS.

Conflicts of interest: No conflicts of interest reported.

ESCG-P-6

CHRONIC DIARRHEA IN DOGS: A COMPARISON OF DOGS WITH INTESTINAL LYMPHOMA AND INFLAMMATORY ENTEROPATHIES. M. Volkman^{1,5}, G.T. Fosgate², J.M. Steiner³, J. Zentek⁴, S. Hartmann², B. Kohn¹. ¹Clinic for Small Animals, Faculty of Veterinary Medicine, Freie Universität Berlin, Germany, ²Department of Production Animal Studies, Faculty of Veterinary Science, University of Pretoria, South Africa, ³Gastrointestinal Laboratory, Texas A&M University, College Station, United States of America, ⁴Institute of Animal Nutrition, Faculty of Veterinary Medicine, Freie Universität Berlin, Germany, ⁵Institute of Immunology, Faculty of Veterinary Medicine, Freie Universität Berlin, Germany

Chronic diarrhea occurs frequently in dogs and common causes include intestinal lymphoma (IL) or inflammatory enteropathies, including food-responsive (FRE), antibiotic-responsive (ARE), or steroid-responsive enteropathy (SRE). The objective of this study was to compare characteristics and blood parameters of dogs with these conditions. Medical records of 102 dogs with chronic diarrhea and a diagnosis of FRE (64), ARE (11), SRE (22), or IL (5) were retrospectively reviewed (Small Animal Clinic, Freie Universität Berlin, 2009 to 2011). *P* values less than 0.05 were considered statistically significant and were based on Kruskal Wallis tests for continuous variables and Pearson chi-square or Fisher exact tests for categorical data.

Dogs with IL and SRE were older (median [range in years]: IL: 9.0 [7.0-13.0], SRE: 8.0 [1.0-12.5], FRE: 5.3 [0.5-13.5], ARE: 4.0 [1.0-12.0]) and large breed dogs were overrepresented in the IL and SRE groups (IL: 4/5, SRE: 14/22, FRE: 24/64, ARE: 6/11), but there were no significant differences in age and breed size (*p* = 0.076 and *p* = 0.061, respectively). Underweight to thin body condition was significantly associated with IL (IL: 5/5, SRE: 17/22, FRE: 33/64, ARE: 7/11; *p* = 0.044). The canine inflammatory bowel disease activity index was significantly increased in dogs with IL or SRE (median [range in points]: IL: 11 [5-15], SRE: 11 [4-16]; FRE: 7 [2-14], ARE: 7 [4-13]; *p* < 0.000). Lack of treatment response was significantly associated with IL (IL: 3/4, SRE: 6/20, FRE: 0/63, ARE: 0/11; *p* < 0.000). Anemia, thrombocytopenia, and increased plasma urea were significantly associated with IL (anemia: IL: 4/5, SRE: 13/22, FRE: 11/64, ARE: 5/11, *p* < 0.000; thrombocytopenia: IL: 3/5, SRE: 0/22, FRE: 4/64, ARE: 0/11, *p* = 0.004; increased urea: IL: 3/5, SRE: 1/22, FRE: 2/62, ARE: 1/11, *p* = 0.003). Hypoalbuminemia (<20 g/L) and hypocalcemia (<200 pg/mL) occurred significantly more frequently in dogs with SRE (hypoalbuminemia: IL: 0/5, SRE: 10/22, FRE: 1/62, ARE: 1/11, *p* < 0.000; hypocalcemia: IL: 0/3, SRE: 9/20, FRE: 8/63, ARE: 1/11, *p* = 0.015).

Results of this study show that elderly and large breed dogs were more frequently affected with IL and SRE compared to other etiologies and both IL and SRE were associated with greater disease severity and/or a negative outcome. In comparison, anemia, thrombocytopenia, and increased plasma urea were most frequently detected in IL whereas severe hypoalbuminemia and hypocalcemia were significantly associated with SRE.

Conflicts of interest: No conflicts of interest reported.

ESCG-P-7

EFFECT OF HYDROCORTISONE ON SERUM ALPHA1-PROTEINASE INHIBITOR CONCENTRATIONS IN HEALTHY DOGS. N. Grützner¹, R.M. Heilmann¹, S. Schellenberg², J.S. Suchodolski¹, J.M. Steiner¹. ¹Gastrointestinal Laboratory, College station, United States of America, ²Tierklinik Aarau West, Aarau, Switzerland

Alpha1-proteinase inhibitor (α_1 -PI) is a proteinase-resistant protein that can be quantified in fecal, urine, and serum samples from dogs. Recently, increased fecal and urinary canine α_1 -PI (α_1 -PI) concentrations have been described in dogs with gastrointestinal diseases (e.g., inflammatory bowel disease [IBD], but also in dogs with exocrine pancreatic insufficiency) and in dogs with chronic hepatitis or chronic kidney disease, respectively. Decreased serum α_1 -PI concentrations have been reported in dogs with IBD, protein-losing enteropathy (PLE), and hypocalcemia. Treatment protocols for dogs with IBD and/or PLE commonly include corticosteroids, but the effect of corticosteroid therapy on serum α_1 -PI concentrations have not yet been reported. The aim of this study was to evaluate the effect of hydrocortisone on serum α_1 -PI concentrations in healthy dogs.

Twelve healthy Beagle dogs were randomly allocated to a placebo-group (n = 6) and to a treatment group (n = 6; hydrocortisone-group). The placebo-group received an empty gelatin capsule PO q12 h, whereas the hydrocortisone-group was treated with hydrocortisone at a dose of 8.5 mg/kg PO q12 h. Serum samples were obtained at baseline and on day 1, 5, 28, 56, and 84 during treatment as well as day 1, 5, 28, 56, and 84 post-treatment for all dogs. Serum α_1 -PI concentrations were measured at all time points using an in-house radioimmunoassay. A Mann-Whitney *U* test was used to compare the baseline measurements of both groups. The effect of hydrocortisone-treatment on serum α_1 -PI concentrations was evaluated by comparing α_1 -PI at baseline and during treatment and between baseline and post-treatment period using a MANOVA.

Baseline serum α_1 -PI concentrations did not differ between the hydrocortisone- and the placebo-group (*p* > 0.05). Serum α_1 -PI concentrations increased significantly (*p* = 0.0004) during the treatment period in the hydrocortisone-group (baseline [median in mg/L: 1,583], day 1 [1,886], 5 [2,254], 28 [3,101], 56 [3,169], and 84 [3,004]), but not in the placebo-group (baseline [1,511], day 1 [1,688], 5 [1,838], 28 [1,976], 56 [1,834], and 84 [1,796]). In contrast, no difference was observed between both groups when comparing serum α_1 -PI concentrations at baseline and during the post-treatment period (*p* > 0.05).

This study showed that hydrocortisone-treatment over 12 weeks did affect serum α_1 -PI concentrations in healthy dogs. Whether corticosteroid therapy has any effects on fecal or urine α_1 -PI concentrations in healthy dogs remains to be determined.

Conflicts of interest: The author works at Texas A&M University, whose GI Lab currently offer a commercial assay for faecal Alpha1-Proteinase Inhibitor.

ESCG-P-8

SERUM CITRULLINE AS A NOVEL MARKER OF CHRONIC ENTEROPATHY IN DOGS. M. Gerou-Ferriani¹, R. Allen², P.J.M. Noble², A.J. German², M. Caldin³, D.J. Batchelor². ¹Clinica Veterinaria Malpensa, Samarate, Italy, ²University of Liverpool, Liverpool, United Kingdom, ³Clinica Veterinaria San Marco, Padova, Italy

Canine chronic enteropathy (CE) is a common, but poorly understood syndrome, with variable response to therapy and prognosis. There is a need for novel biomarkers that are specific for intestinal disease and that provide objective measures of disease severity, progression, and prognosis. Serum citrulline is a useful biomarker in human intestinal disease as it is specific to the small intestine and indicates globally reduced enterocyte mass and absorptive function in various disease states. It is used to determine, quantitatively, intestinal integrity at the enterocyte level and is not influenced by nutritional or

inflammatory status. The aim of this study was to determine whether serum citrulline can be used as a biomarker for CE in dogs.

In this retrospective study, computer records from the University of Liverpool Small Animal Teaching Hospital were used to identify dogs with CE. Disease severity was quantified by CIBDAI. Controls were age- and breed-matched dogs without gastrointestinal disease. Serum citrulline was measured by ultra-high performance liquid chromatography with tandem mass spectrometry. In dogs with CE, serum citrulline concentration was measured at presentation and at various time points after starting treatment.

Serum citrulline was measured in 49 dogs with CE and 69 controls. 17 dogs responded to dietary manipulation (food-responsive enteropathy, FRE) and 3 responded to antibacterials (antibiotic-responsive diarrhoea, ARD), with a further 2 having invasive mucosal bacteria, of which one responded to antibacterials and one was refractory. 27 dogs were diagnosed with idiopathic IBD (on the basis of exclusion of known causes and failure to respond to therapeutic dietary and antibiotic trials), of which 12 responded to immunosuppressive therapy, 13 were refractory, and 2 were lost to follow-up. Serum citrulline concentration did not differ between dogs with CE (median 8.5 µg/mL, range 1.4-20.6) and controls (median 8.2 µg/mL, range, 1.2-34.9, $P = 0.96$). There was also no difference in serum citrulline concentration amongst dogs with FRE, ARD, IBD, and controls ($P = 0.48$). Serum citrulline did not differ between dogs that responded well, or were refractory to treatment ($P = 0.39$), between 23 dogs with and 26 without protein-losing enteropathy ($P = 0.67$), or between 35 dogs that survived and 8 that were euthanased because of CE ($P = 0.65$). Serum citrulline did not correlate with CIBDAI ($r^2 = 0.10$).

These findings do not support the use of serum citrulline as a biomarker in determining diagnosis, prognosis, or quantifying severity in dogs with CE.

Conflicts of interest: One of the co-authors (Marco Caldin) has a diagnostic laboratory offering citrulline assays.

ESCG-P-9

DIFFERENTIAL EXPRESSION OF CALPROTECTIN AND CD163 IN CANINE INTESTINE. J. Dandrieux¹, B. Bacci², C.S. Mansfield¹. ¹Translational Research and Animal Clinical Trial Study (TRACTS) Group, Melbourne, Victoria, Australia, ²Faculty of Veterinary Science, The University of Melbourne, Melbourne, Victoria, Australia

Chronic enteropathy (CE) is a multi-factorial disease, which involves aberrant immune responses to commensal bacteria or dietary antigens. Macrophages have an important role in human disease but little information is available in canine intestine. Data to date have relied solely on macrophage identification using MAC387, an antibody directed against calprotectin, which recognizes both macrophages and neutrophils. In this study an alternative antibody for macrophages, AM-3K, directed against a scavenger receptor (CD163) was used and distribution of both markers was compared. This antigen is of interest as positive cells accumulate in intestine of humans with CE.

Endoscopic duodenal biopsies were obtained from seven cross-breed dogs. Serial histologic sections were stained with MAC387 or AM-3K. Positively-stained cells were counted from 5 random areas from both villous and crypt regions. Stained cell localisation was subjectively evaluated and the percentage of positively stained cells from the total nucleated cells per 10,000 µm² in the villus or crypt was compared between both antibodies using a Wilcoxon signed-rank test.

MAC387 and AM-3K did not co-localize on serial sections. There were significantly more AM-3K positive cells than MAC387 in the crypts (3.6% [0-7.0] versus 0.8% [0-7.5], $P = 0.005$). In contrast there was no difference in expression of either markers in the villi (3.2% [0-8.16] versus 1.8% [0-11.2], $P = 0.27$).

This study reports for the first time the existence of two populations of macrophages in canine intestine. These results in normal dogs will be used to explore further the distribution and function of macrophages in dogs with CE.

Conflicts of interest: No conflicts of interest reported.

ESCG-P-10

FINAL DIAGNOSES IN 155 DOGS WITH CHRONIC VOMITING AND/OR DIARRHEA. K. Baumgart¹, M. Volkmann¹, J.M. Steiner², B. Kohn¹. ¹Clinic For Small Animals, Faculty of Veterinary Medicine, Freie Universität, Berlin, Germany, ²Gastrointestinal Laboratory, Texas A&M University, College station, United States of America

Chronic diarrhea and vomiting are common clinical signs in dogs. Primary (e.g., inflammatory, infectious, neoplastic, mechanical, or other) and secondary gastrointestinal diseases (e.g., exocrine pancreatic, hepatic, renal, or endocrine disease) are possible underlying causes.

The aim of this study was to evaluate the final diagnoses in dogs with chronic diarrhea and/or vomiting and to determine the prevalence of various primary and secondary gastrointestinal diseases in dogs with these gastrointestinal signs.

Medical records of 209 dogs presented between July 2011 and August 2013 with chronic diarrhea (D), vomiting (V) or both (diarrhea and vomiting [VD]) were retrospectively reviewed. Dogs were included if a minimum work-up (hematology, plasma biochemistry profile, and fecal parasitology) had been performed and if a final diagnosis was recorded (155/209).

A primary gastrointestinal disease was recorded in 83% of the cases (129/155) and included inflammatory diseases (90/129: food responsive enteropathy [55], antibiotic responsive enteropathy [18], idiopathic inflammatory bowel disease [10], steroid responsive enteropathy [4], protein-losing enteropathy of unknown etiology [3]), infectious diseases (23/129: giardiasis [18], leishmaniasis [2], ascariasis [2], protothecosis [1]), neoplastic diseases (13/129: intestinal lymphoma [8], adenocarcinoma [3], leiomyoma [1], histiocytic sarcoma [1]) and, in one dog each, drug related enteropathy, mechanical obstruction, and diaphragmatic rupture. A secondary gastrointestinal disease was less frequently diagnosed (17%, 26/155: chronic pancreatitis [12], portosystemic shunt [5], hepatopathy [2], exocrine pancreatic insufficiency, hypoadrenocorticism, polyendocrinopathy, dilated cardiomyopathy, and leukemia in one dog each). In total, 44% of the dogs were presented with D (69/155) followed by 33% with VD (51/155), and 23% with V (35/155). D and VD were significantly more frequent in dogs with primary gastrointestinal disease (D: 61/129, VD: 46/129), compared to dogs with secondary gastrointestinal disease (D: 8/26; VD: 5/26; $p = 0.001$, Chi square test). V was significantly more common in dogs with secondary gastrointestinal disease (13/26) as compared to dogs with primary gastrointestinal disease (22/129; $p = 0.001$).

In this study, food responsive enteropathy (36%) was the most commonly diagnosed cause of chronic gastrointestinal signs. Chronic pancreatitis was the most frequent cause of secondary gastrointestinal disease (46%). Diarrhea was significantly associated with primary and vomiting with secondary gastrointestinal disease.

Conflicts of interest: No conflicts of interest reported.

ESCG-P-11

CHARACTERIZATION OF MATRIX METALLOPROTEINASE-2 AND -9 IN INTESTINAL MUCOSA OF CLINICALLY HEALTHY DOGS. M. Hanifeh^{1,3}, M.M. Rajamäki¹, L. Mäkitalo², P. Syrjä¹, S. Sankari¹, S. Kilpinen¹, T. Spillmann¹. ¹Faculty of Veterinary Medicine, University of Helsinki, Helsinki, Finland, ²Helsinki University Central Hospital, University of Helsinki, Helsinki, Finland, ³Faculty of Veterinary Medicine, University of Tabriz, Tabriz, Iran

Matrix metalloproteinases (MMPs) 2 and 9 are zinc-dependent endopeptidases that contribute to the control of breakdown and reconstitution of extracellular matrix under both normal and pathological conditions. Intestinal mucosal levels of MMP-2 and -9 have been shown to be increased in animal models and human IBD. To our knowledge, the presence of MMP-2 and -9 has not been studied in the intestinal mucosal samples of healthy dogs as well as in canine spontaneous IBD. Thus, the main aim of this study was to identify the presence of MMP-2 and -9 in the mucosa of the small and large intestines of clinically healthy

beagle dogs using gelatin zymography technique. For the study, historical intestinal tissue samples from four different parts of the intestine (duodenum, jejunum, ileum and colon) were used. The samples were taken and snap frozen in liquid nitrogen during necropsy from 12 healthy laboratory beagle dogs after being euthanized when finishing unrelated long-term trials studying canine intestinal microbiota.

Based on WSAVA histology standards, recorded findings of all samples were considered insignificant. Pro-MMP-2 and -9 activities were found in 17/48 (35%) and 25/48 (52%) of the samples, respectively. Among four different parts of the intestine of 12 dogs, the ileum had the highest positivity rates of 7/12 (58.3%) and 8/12 (66.7%) for pro-MMP-2 and -9 activities, respectively. However, statistical analysis showed no significant difference of pro-MMP-2 and -9 activities between the separate parts of the intestine ($P > 0.05$). The enzyme activities ranged for pro-MMP-2 between 0.015 and 6.449 arbitrary units (AU) and for pro-MMP-9 between 0.018 and 5.680 AU. None of the intestinal samples showed gelatinolytic activity corresponding to the control bands of active MMP-2 and MMP-9.

This study showed that pro-MMP-2 and -9 could be detected in the intestinal mucosa of healthy dogs using zymography, which seems to be a useful tool to evaluate the role of MMP-2 and -9 in the pathogenesis of canine chronic enteropathies, including inflammatory bowel diseases.

Conflicts of interest: No conflicts of interest reported.

ESCG-P-12

SPONTANEOUS GASTROINTESTINAL PERFORATION IN 13 CATS. F. Bernardin, L. Martinez Rivera, J.H. Hernandez. CHV Frégis, Arcueil, France

Digestive perforation is called spontaneous when it arises in the absence of foreign body ingestion, gastric dilatation and volvulus, external trauma, or previous digestive surgery. In dogs many predisposing factors have been identified, including anti-inflammatory administration, severe hepatic or renal disease, stress, shock, gastric hyperacidity, neoplasia and idiopathic inflammatory bowel disease. Spontaneous digestive perforation has been uncommonly reported in cats.

The objectives of this study were to describe the clinical characteristics of spontaneous digestive perforation in cats, and to compare the frequency of malignant versus non-malignant causes for these perforations.

To be included in this study, the perforation had to be spontaneous and confirmed by exploratory surgery.

The medical records of 13 cats diagnosed as having spontaneous digestive perforation between 2010 and 2013 were reviewed. The mean age of cats was 7.6 years (9 months to 17 years). Five cats had concurrent illnesses including viral upper respiratory tract disease, pancreatitis and chronic kidney disease. The most frequently reported signs included anorexia (85%), vomiting (61%), and lethargy (54%). Histological examination was performed in 11 cats and diagnosed alimentary lymphoma in 54% and inflammatory lesions in the other 46% of them. Six cats had received anti-inflammatory within the previous 3 months. Half of them were finally diagnosed with lymphoma. Five cats with lymphoma received chemotherapy. Three cats died early in the postoperative recovery period, 5 cats were euthanized 12 to 146 days after surgery, and 2 cats were still alive at the end of this study.

In the absence of pneumoperitoneum, clinical signs and clinicopathological abnormalities are not specific enough to allow differentiation between cats with gastrointestinal ulceration and those with perforation. In most cases, there is no diagnostic test that individually determine whether perforation has occurred or is impending, and clinicians should use of multimodality diagnostic procedures such as radiography, ultrasonography, endoscopy, and abdominal fluid cytopathology to avoid delay in diagnosis of digestive perforation. Histological examination of ulceration is essential as lymphoma should be suspected in all cats presented with spontaneous perforation. The link between anti-inflammatory administration and spontaneous perforation in cats is not established.

Conflicts of interest: No conflicts of interest reported.

ESCG-P-13

WHOLE-GENOME ANALYSES OF CAMPYLOBACTER UPSALIENSIS AND C. HELVETICUS ISOLATED FROM DOGS AND CATS AND IN SILICO INVESTIGATION OF THEIR PATHOGENIC POTENTIALS. K. Bojanic, A.C. Midwinter, P.J. Biggs, N.P. French, E. Acke. Massey University, Palmerston north, New Zealand

Campylobacter species are commonly isolated from faeces of dogs and cats with *C. upsaliensis* (*CU*) and *C. helveticus* (*CH*) being the most frequently isolated. These two species are usually not considered pathogenic in dogs and cats and are closely related to each other and to *C. jejuni*, the most common cause of bacterial gastroenteritis in humans in the developed world. Interestingly, despite their close genetic relationship, in humans *CU* is considered a pathogen while *CH* is not. This study aimed to describe whole genomes of *CU* and *CH* isolated from dogs and cats and to *in silico* investigate their pathogenic potential with comparison to several published genomes of *C. jejuni* and *C. coli*.

Genomic DNA was extracted from three isolates of each of *CU* and *CH* recovered from the faeces of healthy dogs and cats. Sequencing was performed using an Illumina MiSeq to generate 250 base paired reads. Reads were trimmed for both length and quality. Contigs were assembled using the Velvet assembler. The concatenated contigs generated for each assembly were quality ranked (by number, size, maximum length and N50) and the three top ranked assemblies were annotated using the Prokka annotation tool. Ribosomal MLST nucleotide sequences were used as a proxy for the core genome to compare the phylogeny of *CU* and *CH* with other species in the *Campylobacter* genus and visualised as a NeighborNet using SplitsTree. Annotated draft genomes were clustered using OrthoMCL and pathogenic traits were investigated *in silico* using PathogenFinder and VirulentPred software.

The *CU* and *CH* draft genomes were ~1.732 Mb and ~1.844 Mb in size, and comprised on average 110 and 151 contigs, and on average 1782 and 1942 predicted genes, respectively. Of these *CU* had on average 497 and *CH* 622 hypothetical proteins. Using OrthoMCL, a core genome of 1459 and 1751 genes resulted for *CU* and *CH*, respectively. NeighborNet trees based on ribosomal MLST nucleotide sequences and the core genome confirmed the close phylogenetic relationship of *CH* and *CU* within the *Campylobacter* genus. PathogenFinder predicted all isolates as human pathogens with probabilities of 88.3-91.5%. Both PathogenFinder and VirulentPred identified many pathogenic proteins in *CU* and *CH* of different functions (e.g. chemotaxis, transporter and motility systems) but considerably fewer than in *C. jejuni* and *C. coli*.

This study provides many insights into the pathogenic potential of pet-associated emerging *Campylobacter* pathogens and is to our knowledge, the first to report a draft genome of *CH*.

Conflicts of interest: No conflicts of interest reported.

ESCG-P-14

DETERMINATION OF S100A12 AND MYELOPEROXIDASE LEVELS WITHIN INTESTINAL MUCOSA OF CLINICALLY HEALTHY BEAGLE DOGS. M. Hanifeh¹, R.M. Heilmann², S. Sankari¹, M.M. Rajamäki¹, L. Mäkitalo², P. Syrjä¹, S. Kilpinen¹, J.S. Suchodolski², J.M. Steiner², T. Spillmann¹. ¹Faculty of Veterinary Medicine, University of Helsinki, Helsinki, Finland, ²Gastrointestinal Laboratory, Texas A&M University, COLLEGE STATION, United States of America, ³Helsinki University Central Hospital, University of Helsinki, Helsinki, Finland

There are only few laboratory markers being evaluated for diagnosing and/or monitoring canine chronic enteropathies, including inflammatory bowel disease (IBD). S100A12 belongs to the S100/calgranulin-protein family and has been proposed to play a central role in both innate and acquired immune responses. It has been reported to be increased in stool samples, serum and/or intestinal mucosa in human patients with IBD. Myeloperoxidase (MPO) is an enzyme found mostly in granulocytes. Intestinal mucosal levels of MPO have been shown to be increased in animal models and human IBD. To

date, S100A12 and MPO levels in intestinal mucosal samples have been reported neither from healthy dogs nor from dogs suffering from IBD. To start investigating this aspect in dogs, the objective of this study was to evaluate mucosal S100A12 and MPO levels in the small and large intestines by using enzyme-linked immunoassay (ELISA) and spectrophotometric methods, respectively. For the study, historical intestinal tissue samples from four different parts of the intestine (duodenum, jejunum, ileum and colon) were used. The samples were taken and snap frozen in liquid nitrogen during necropsy from 12 healthy laboratory beagle dogs after being euthanized when finishing unrelated long-term trials studying canine intestinal microbiota.

Based on WSAVA standards the histologic findings of all samples were considered insignificant. S100A12 concentrations were from the highest to the lowest: ileum, 71.5 (38.9-141.9) $\mu\text{g/L}$; colon, 23.2 (6.7-75.6) $\mu\text{g/L}$; duodenum, 11.4 (6.9-28.5) $\mu\text{g/L}$; and jejunum, 8.5 (5.1-19.3) $\mu\text{g/L}$. The concentration in the ileum was significantly higher than in all other segments ($P < 0.05$), and the colonic mucosal concentration was higher than the jejunal ($P < 0.05$). The highest MPO activity was found in the ileum (0.49 [0.19-1.05] $\Delta\text{A}/\text{min}$), followed by jejunum (0.36 [0.28-1.70] $\Delta\text{A}/\text{min}$), duodenum (0.26 [0.09-0.59] $\Delta\text{A}/\text{min}$), and colon (0.09 [0.04-0.14] $\Delta\text{A}/\text{min}$). MPO activity was significantly higher in ileal and duodenal than in colonic mucosal samples ($P < 0.05$). The jejunal MPO activity was higher than the colonic and duodenal activity ($P < 0.05$).

This study showed that using ELISA and spectrophotometry allow the detection of canine intestinal mucosal S100A12 and MPO, respectively. The levels on S100A12 and MPO seem to differ between certain parts of the intestinal mucosa of healthy dogs. Both assays appear to be useful to further evaluate the role of S100A12 and MPO in the pathogenesis of canine chronic enteropathies, including IBD.

Conflicts of interest: Dr. Heilmann, Dr. Suchodolski, and Dr. Steiner have a patent pending that includes the canine S100A12 assay used in this study. The authors declare that they have no further conflicts of interest.

ESVC-P-1
PUPPIES WITH AN INNOCENT CARDIAC MURMUR HAVE LOWER HEMATOCRIT. V. Szatmári, M.W. van Leeuwen, E. Teske. Faculty of Veterinary Medicine, Utrecht University, Utrecht, The Netherlands

The aim of the present study was to establish the incidence of innocent cardiac murmurs in a fairly large number of clinically healthy puppies. A second aim was to evaluate a possible correlation between the presence of an innocent cardiac murmur and a lower hematocrit value.

Puppies of certain breeds are routinely screened for the presence of congenital porto-systemic shunts in the Netherlands. Breeders bring their nests to our clinic for individual measurement of blood ammonia concentration.

In one year time (from February 2013 until January 2014) 389 dogs of 11 different breeds were examined, with 295 of them being cairn terriers. The age of the dogs varied from 20 to 108 days (mean 53 days). While the breeders were waiting for the blood results, the cardiac auscultation was performed by a single board-certified cardiologist (VSz). Hematocrit was measured with an automatized hematology analyzer system from the surplus blood sample.

Cardiac murmur was found in 59 dogs (15%). In all cases this was a soft (1-2 out of 6) systolic murmur, most of the time with a musical character and with the point of maximal intensity on the left hemithorax, compatible with the description of an innocent cardiac murmur. No murmurs were found that could be compatible with a congenital cardiac anomaly. The hematocrit was significantly ($P = 0.003$) lower in the group of dogs with a murmur (mean 30.9%, standard deviation 2.8%) compared to the group without a murmur (mean 32.2%, standard deviation 3.1%). Multivariate analysis shows that the presence of murmur is correlated with the hematocrit, but not with the age of the dogs.

Physiologic anemia has been long suspected to be one of the possible causes of innocent cardiac murmurs in young animals

and children. However, according to the authors knowledge, no published reports exist that looked for a possible correlation. Which other factors contribute to the presence of an innocent murmur is largely unknown.

Because of a large overlap between the hematocrit values of dogs with and without a cardiac murmur, measuring hematocrit in a particular pup would not help a less experienced first line veterinary practitioner to decide whether a murmur is innocent or the result of a congenital cardiac anomaly.

Limitation of this study is that no echocardiography was performed to rule out congenital cardiac anomalies as the cause of the murmurs. Neither were the dogs re-checked for spontaneously disappearance of the murmur.

Conflicts of interest: No conflicts of interest reported.

ESVC-P-2
CARDIAC MURMURS IN NORMAL SIBERIAN HUSKY DOGS. C. Kwart. University of Agricultural Science, Uppsala, Sweden

Low intensity systolic murmurs, with point of maximal intensity over the outflow tract on the left side of thorax, are not uncommonly heard during cardiac auscultation of apparently healthy Siberian husky dogs, often with excellent exercise tolerance. The origin of these murmurs in athletic dogs such as huskies is unlikely to be due to heart disease but more likely due to turbulent blood flow in the outflow tract caused by a large stroke volume and forceful cardiac contractility in early systole. The differentiation of these murmurs from "pathological" significant murmurs can however be problematic in general practice. The aim of the present study was to investigate the prevalence of murmurs in a sample of successfully racing Siberian husky dogs and furthermore to study the phonocardiographic characteristics of these murmurs.

Phonocardiograms and ECGs were recorded in 37 actively racing Siberian husky dogs, with normal or excellent exercise tolerance. Normal stamina was confirmed by successful racing. Phonocardiograms were easy and rapid to record on a PC laptop connected to the Meditron stethoscope in ambulant "field" practice. Systole was measured as the duration measured from the onset of the first heart sound to the onset of the second heart sound and the murmur duration from the onset the first heart sound to the end of the murmur. The duration of the first heart sound plus the murmur was measured and calculated as a percentage of the duration of systole.

Cardiac murmurs of grade 1-2 were heard in 22% of dogs examined. Phonocardiogram from these dogs revealed early systolic crescendo-decrescendo or decrescendo murmurs with a duration of maximally 75% into systole. All dogs with murmurs had a silent pause at the end of systole. ECG was normal in all dogs.

Murmurs in adult athletic dogs should not be regarded as a definite sign of heart disease. Physiological flow murmurs of up to 75% of systole is a common finding in active Siberian husky dogs (prevalence 22% in the examined sample). Phonocardiography is a rapid and practical method for differential diagnoses between pathological murmurs and physiological flow murmurs.

Conflicts of interest: No conflicts of interest reported.

ESVC-P-3
INVESTIGATION OF NT-PROBNP POINT-OF-CARE ELISA ASSAY IN CLINICALLY NORMAL FELINE PATIENTS. A. Harris¹, A.H. Estrada¹, B. Mincey¹, K.E. Lamb², M. Bohannon¹, M. Powell¹, S. Swift¹, A. Gallagher¹, K. Cooke¹, J. Hanscom³, C. Mainville³. ¹Small Animal Clinical Sciences, University of Florida, Gainesville, United States of America, ²Lamb Consulting, West Saint Paul, United States of America, ³IDEXX Laboratories Inc, Westbrook, United States of America

NT-proBNP has a degree of overlap with clinically normal animals, particularly those with mild or subclinical heart disease.

Prior studies have evaluated the sensitivity and specificity of a point-of-care second generation ELISA that utilizes SNAP technology. The SNAP Feline proBNP Test uses the same biological reagents as the Cardiopet proBNP Test but provides results in 10 minutes. We sought to prospectively validate the assay in a population of clinically normal cats. Cats were recruited based upon the absence of a heart murmur, gallop, and/or arrhythmia. All cats received physical examination, non-invasive blood pressure measurement, complete biochemical analysis including a T4, urinalysis and echocardiogram. Only cats considered free of underlying cardiac or systemic disease were enrolled. Sixteen adult cats were enrolled and blood samples were obtained for NT-proBNP concentrations at 0, 2 hr, 4 hr, 6 hr, 8 hr, 10 hr. Samples were placed in EDTA tubes and centrifuged within one hour and split into two tubes for duplicate samples at each time point and stored at -80°C. Once all samples were collected, they were shipped on dry ice overnight and run in one batch (IDEXX Laboratories) for measurement of NT-proBNP concentrations. SNAP tests were visually evaluated by one blinded reader. Comparison of SNAP assay vs. quantitative ELISA revealed a 1.0 (AUC) degree of correlation between assays, and that a positive SNAP test result was associated with a NT-proBNP concentration of 126.4pmol/L or greater. The average BNP concentration of abnormal cats (191.1 ± 5.8) determined by the SNAP assay was significantly greater than the normal (26.4 ± 1.2).

Conflicts of interest: This study was funded through IDEXX and the University of Florida College of veterinary medicine resident grant competition.

ESVC-P-4

LEFT ATRIAL FUNCTION DETERMINED BY 2-DIMENSIONAL SPECKLE TRACKING ECHOCARDIOGRAPHY IDENTIFIES DOGS WITH CONGESTIVE HEART FAILURE SECONDARY TO MITRAL VALVE DISEASE. D. Caivano¹, V. Patata¹, F. Biretoni¹, M.E. Giorgi¹, M. Rishniw², F. Porcillo¹. ¹University of Perugia, Perugia, Italy, ²Veterinary Information Network, Davis, United States of America

Left atrial (LA) function (consisting of 3 phases: reservoir, conduit and booster pump) can be evaluated by speckle tracking echocardiography (STE) which measures myofiber deformation during these phases. Recent studies in humans have evaluated the utility of STE in assessing left atrial deformation/function. We evaluated the deformation of the LA by STE in healthy dogs and in dogs with myxomatous mitral valve disease (MMVD).

We acquired 2D echocardiographic cineloops from the left apical 4-chamber view optimized for the LA, and analyzed atrial longitudinal strain (St) and strain rate (SR) in 27 dogs (10 healthy dogs and 17 dogs with MMVD - 5 ACVIM Stage B1, 5 Stage B2 and 7 Stage C). Endocardial LA STE curves were obtained and peak atrial longitudinal strain (PALS), peak atrial contraction strain (PACS), conduit atrial longitudinal strain (CALS); PALS-PACS) and contraction strain index (CSI - PACS/PALS*100) were calculated. LA SR curves were similarly obtained to determine the peak positive strain rate (SRs) during left ventricle systole, the first negative peak strain rate (SRe) during early diastole and the second negative peak strain rate (SRa) during atrial contraction. For all variables, a mean of 3 measures was used for the statistical analysis. We compared each of these variables between each ACVIM stage by Kruskal-Wallis tests and post-hoc pairwise comparisons, with comparison-wise $\alpha=0.05$.

Normal dogs had higher PALS and CALS than dogs with MMVD ($p < 0.0001$ and $p = 0.0005$); Stage C dogs had lower PALS, PACS and CALS than all other dogs ($p < 0.0001$, $p = 0.0022$ and $p = 0.0005$), but CSI did not differ between groups ($p = 0.1$). Stage C dogs had lower SRs ($p = 0.0005$), higher SRe ($p = 0.0029$) and SRa ($p = 0.0004$) than other dogs. Normal dogs had lower SRe and SRa than dogs with MMVD ($p < 0.001$).

Our data suggest that STE might be useful in assessing LA function in dogs with MMVD, and might potentially differentiate dogs with severe subclinical disease from dogs with congestive heart failure.

Conflicts of interest: No conflicts of interest reported.

ESVC-P-5

BREED-SPECIFIC REFERENCE RANGES FOR ECHOCARDIOGRAPHY IN SALUKIS. S.M. Lehtinen¹, M.E. Wiberg¹, J. Häggström², H. Lohi³. ¹Faculty of Veterinary Medicine, University of Helsinki, Helsinki, Finland, ²Swedish University of Agricultural Sciences, Uppsala, Sweden, ³Molecular Neurology, University of Helsinki, Helsinki, Finland

Sighthounds are athletic dogs and they have been claimed to have larger hearts compared to similar sized breeds. The left ventricle (LV) may enlarge in response to cardiac disease, but also in response to training, so called athlete's heart syndrome, which is a benign condition. To distinguish abnormal echocardiographic measurements from normal, breed-specific reference values are needed. The aim of this study is to establish normal reference ranges for echocardiographic measurements in the Saluki breed.

The study comprised 78 clinically healthy Salukis (41 males and 37 females), mean age 72 months (\pm SD 28 months), body-weight (BW) 24,7 kg (\pm 3,7 kg). Case history was ascertained and dogs underwent physical examination, complete blood count, serum biochemistry profile, thyroid profile, blood pressure measurement and 3-min ECG. Standard M-mode and 2D echocardiographic measurements were obtained. Dogs with systolic murmur 1/6, and dogs with mitral valve regurgitation (MR) <15% (MR color flow jet area/left atrium area \times 100% in apical view) were considered normal. Linear regression models were used to establish reference ranges.

Heart rate (HR) varied from 44 to 120 bpm (81 ± 17 bpm). BW was a significant predictor for LV dimensions, i.e. M-mode LV diameter and 2D volume in diastole (LVIDD and LVEDV) and systole (LVIDS and LVESV), and mitral valve end point septal separation (EPSS). HR was a significant predictor for FS % (fractional shortening). Predicted values (95% prediction intervals) were calculated from regression models where mean BW (24,7 kg) and age (72 months), and median HR (80 bpm) were used. Normal reference ranges were: LVIDD 46,0 mm (40,0-52,0), LVIDS 33,4 mm (27,3-39,5), LVEDV 86,3 ml (64,6-108,0), LVESV 44,2 ml (29,2-59,2), FS%: 27,5% (20,3-34,6), ejection fraction EF%: 48,9% (38,6-59,1), EPSS 7,3 mm (4,4-10,2), sphericity index 1,6 (1,4-1,9), interventricular septum in diastole 10,9 mm (8,6-13,3) and systole 13,8 mm (10,5-17,1), LV free wall in diastole 10,4 mm (8,3-12,4) and systole 13,1 mm (10,0-16,1), left atrial (LA) diameter 28,5 mm (23,8-33,3), aortic (Ao) diameter 23,9 mm (20,1-27,8), LA/Ao 1,2 (1,0-1,4), and aortic and pulmonary flow velocity 1,4 m/s (0,9-1,9) and 1,2 m/s (0,8-1,6), respectively.

This study provides echocardiographic values for normal Salukis which can be used as a reference values.

Conflicts of interest: No conflicts of interest reported.

ESVC-P-6

ANALYSIS OF THE P WAVE DURATION IN ATRIAL REMODELING VS. AUTONOMIC NERVOUS SYSTEM DYSFUNCTION IN DOGS. D. Mocanu¹, I. Necluea², G. Solcan¹, M. Musteata¹. ¹Faculty of Veterinary Medicine Iasi, Iasi, Romania, ²Small Animals Private Practice, Romania

In mitral valve disease, atrial remodeling is an indicator of evolution and prognosis, the duration of the P wave being considered suggestive of the dilatation of the left atrium. In humans' studies, neurological conditions have a significant impact on cardiac electrophysiology by altering the electrical impulse conductivity.

The aim of this study is to examine the duration of the P wave in dogs suffering from mitral valve disease in comparison to dogs diagnosed with different neuropathies without cardiac abnormalities.

We analyzed standard electrocardiograms (5 min of ECG, on 6 peripheral leads) performed on three polymorphic groups of dogs (different age, weight and breed): group 1 (n = 14) healthy dogs, group 2 (n = 30) dogs diagnosed with mitral valve disease and group 3 (n = 27) dogs suffering from different neuropathies (without any associated or previously diagnosed cardiovascular disease). The duration of the P wave was measured for all dogs

(five consecutive P waves without anomalies or artifacts) and reported to the degree of atrial remodeling, assessed by left atrium/aorta ratio on echocardiography. The interpretation of the ECG and echocardiography was made by the same examiner (MD). The results were statistically evaluated in a specialized program (IBM SPSS vs. 21).

The P wave recorded average values was 0.061 ± 0.028 seconds for the MVD group with significant differences between the stages of heart failure ($p = 0.008$). No correlations were found between its increase and the dilation of the left atrium ($R^2 = 0.012$). There was no statistically significant difference regarding P wave duration when compared dogs of the neuropathy group and those of the mitral valve disease group (the P wave recorded average values = 0.068 ± 0.018 sec.).

Both, atrial tissue lesions (as in mitral valve disease) and autonomic nervous system anomalies (secondary to a neurological condition), may change the conductivity of the electrical impulse in the left atrium. The conductivity of the electrical impulse at this level does not seem to be influenced by its actual dilation, but by the impairment of the intra-atrial and inter-atrial conduction pathways. Caution must be given when P wave is analyzed in dogs with concurrent cardiologic and neurologic condition.

Conflicts of interest: No conflicts of interest reported.

ESVC-P-7

TRANSTHORACIC ECHOCARDIOGRAPHY IN CLINICALLY HEALTHY ADULT NEWFOUNDLAND DOGS: REFERENCE VALUES FOR THE BREED. C. Quintavalla, E. Martinelli, S. Crosara. University of Parma, Parma, Italy

Specific echocardiographic reference ranges have been published for several canine breeds. In 1996, one paper stated M-mode values for Newfoundland dogs. The aim of this study was to report reference M-mode, 2D and Doppler echocardiographic values for Newfoundland dogs and to compare M-mode measurements with allometric scaling reference values according to Cornell and colleagues.

Newfoundland dogs were prospectively recruited among those undergoing screening for congenital and acquired heart disease. Screening includes patient history, physical examination, and systemic arterial pressure measurement by Doppler flow meter and transthoracic echocardiography (M-mode, 2D and echo-Doppler). Screening is performed on conscious dogs of at least 1 year of age. Dogs without historical, clinical, electrocardiographic and echocardiographic signs of cardiovascular disease were included in the study.

Unpaired, two-tailed Student's t-test and linear regression were performed to evaluate the influence of gender, age and body weight (BW) on echocardiographic parameters. Echocardiographic measurements were compared to previously reported reference values. The reference limits of echocardiographic parameters in the Newfoundland dogs were calculated. Forty-six healthy adult Newfoundland dogs of both genders (20 males and 26 females), 1 to 6 years of age (mean 2.6 ± 1.6 years), 40 to 72 kg (mean 54.7 ± 8.84 kg) fulfilled the inclusion criteria. Significant but weak correlations were detected between aortic diameter (Ao) and age ($p = 0.012$, $r^2 = 0.186$), left atrial to aortic ratio (LA/Ao) and age ($p = 0.047$, $r^2 = 0.192$), E-point to septum separation (EPSS) and BW ($p = 0.038$, $r^2 = 0.117$), M-mode left ventricular internal diameter (LVID) in diastole (d) and systole (s) and BW (respectively $p = 0.002$, $r^2 = 0.201$ and $p = 0.006$, $r^2 = 0.158$), and between Ao and BW ($p = 0.008$, $r^2 = 0.203$). None of the echocardiographic measurements was statistically different between males and females.

Left ventricular internal diameter in diastole, LVIDs, Ao, EPSS increased with BW, as expected. The aorta appears to become wider with advancing age. A proportion of the studied population had M-mode parameters below the allometric scaling reference range, suggesting that this method can over-estimate M-mode parameters in this breed. These findings stress the importance to report Newfoundland breed specific normal ranges for echocardiographic parameters.

Conflicts of interest: No conflicts of interest reported.

ESVC-P-8

CORRELATION BETWEEN THORACIC CT-SCAN ANGIOGRAPHY FINDINGS AND ECHOCARDIOGRAPHIC RIGHT PULMONARY VEIN TO PULMONARY ARTERY RATIO IN WEST HIGHLAND WHITE TERRIERS WITH IDIOPATHIC PULMONARY FIBROSIS. E. Roels¹, A.C. Merveille¹, T. Couvreur¹, G. Bolen¹, E. Krafft¹, C. Clercx¹, K. Mc Entee². ¹ULG, Liege, Belgium, ²ULB, Brussels, Belgium

Canine idiopathic pulmonary fibrosis (CIPF) is a progressive interstitial lung disease usually diagnosed by thoracic CT-scan that mainly affects West Highland white terriers (WHWT). Pulmonary hypertension (PH), a severe co-morbid condition with a challenging diagnosis, may develop in CIPF dogs. The ratio between the right pulmonary vein and pulmonary artery (PV/PA) has been described as an echocardiographic indicator of PH in CIPF dogs. This study was intended to investigate whether CT-scan angiography cardiac findings are 1) altered in dogs with CIPF compared to healthy control dogs and 2) correlated with PV/PA measured by echocardiography (PV/PA^{US}). Thoracic CTA images from 6 WHWT with CIPF (Group A) and 9 healthy controls from various breeds (Group B) were retrospectively reviewed by one observer. All measurements were obtained in transverse post-contrast images displayed in a soft tissue window. PV and PA were measured dorsal to the right atrium, perpendicular to the long axis of these vessels. In addition, pulmonary trunk (PT) was assessed just ventral to the division of pulmonary arteries, perpendicular to its long axis. Ascending aorta (Ao) was also measured perpendicular to its long axis. Transverse reformatted images were obtained to have a view equivalent to the standard 4 chambers-echocardiographic view where right ventricle (RV) and left ventricle (LV) were measured. Three ratios were calculated PV/PA^{CT}, PT/Ao and RV/LV, compared between groups and correlated with PV/PA^{US} in both bi-dimensional (BD) and M-modes (MM). Statistical analyses were performed with XLStat[®] software. Values are given as mean \pm sd. Statistical significance was set at a $P \leq 0.05$. PV/PA^{CT} was lower in group A (0.64 ± 0.18) in comparison to group B (1.00 ± 0.23 , $P = 0.008$) and correlated with PV/PA^{US} (BD: $r = 0.796$, $P = 0.001$; MM: $r = 0.730$, $P = 0.003$). PT/Ao was higher in group A (1.18 ± 0.08) compared to group B (0.92 ± 0.14 , $P = 0.001$) and correlated only with PV/PA^{US} measured in BD mode ($r = -0.623$, $P = 0.02$). The RV/LV ratio was increased in group A (0.91 ± 0.11) in comparison to group B (0.70 ± 0.08 , $P = 0.001$) and a correlation between RV/LV and PV/PA^{US} was found (BD: $r = -0.726$, $P = 0.005$; MM: $r = -0.657$, $P = 0.01$). In conclusion, in WHWT with CIPF, PV/PA^{CT}, PT/Ao and RV/LV ratios measured on thoracic CTA images are correlated with PV/PA^{US} and may serve in the assessment of PH.

Conflicts of interest: No conflicts of interest reported.

ESVC-P-9

EFFECT OF A BASIC TRAINING PROGRAM ON EMERGENCY CLINICIAN ACCURACY TO SEMI-QUANTITATIVELY ASSESS THORACIC AND CARDIAC STRUCTURES USING FOCUSED CARDIAC ULTRASOUND (FOCUS). L. Wiley¹, M.A. Oyama¹, C. Ostroski¹, E. Reineke¹, E.A. Rozanski², J.E. Rush². ¹University of Pennsylvania, Philadelphia, United States of America, ²Tufts University School of Veterinary Medicine, North Grafton, United States of America

Companion animals presenting to the emergency room in distress need to be assessed rapidly and accurately to implement life-saving therapies. Focused cardiac ultrasound (FOCUS) can be a useful adjunct to the physical examination in assessing dyspneic animals in the emergency room. Rapid bedside ultrasound evaluations performed by EC are commonly used in human medicine, however feasibility and utility of FOCUS by EC in veterinary medicine has not been fully evaluated. The purpose of this study is to determine the baseline accuracy of FOCUS performed by EC and whether or not a basic training session could improve accuracy compared to evaluation by a cardiology specialist. Fifteen EC including 6 boarded emergency-critical care specialists and 9 emergency res-

idents performed FOCUS on four animals; a normal cat and dog, and a cat and dog with severe valvular and myocardial heart disease, respectively. EC semi-quantitatively assessed 6 thoracic and echocardiographic parameters including left atrial dimension, left ventricular systolic function and wall thickness, right heart dimension, and presence or absence of pleural or pericardial effusion before and after a structured didactic lecture and hands-on practical session. Primary outcome was the level of agreement with examination performed by a cardiologist. Level of agreement regarding

EC assessment of all parameters improved from 0.70 to 0.78 after training ($P < 0.01$). Level of agreement concerning left atrial diameter improved from 0.52 to 0.75 ($P < 0.01$). EC confidence in their overall FOCUS evaluation and findings improved from 51% to 70% ($P < 0.0001$). In summary, EC accuracy and confidence in semi-quantitatively assessing basic cardiac parameters using FOCUS were improved following a simple structured training session. FOCUS might be a valuable tool to rapidly assess simple thoracic and cardiac parameters in the emergency setting.

Conflicts of interest: No conflicts of interest reported.

ESVCN-P-1

EFFECTIVENESS OF A NEW DIETETIC WEIGHT MANAGEMENT FOOD TO ACHIEVE WEIGHT LOSS IN CLIENT-OWNED OBESE DOGS. U. Christmann¹, I. Becvarova², S. Werre¹, H. Meyer². ¹Virginia-Maryland Regional College of Veterinary Medicine, Blacksburg, Virginia, United States of America, ²Hill's Pet Nutrition - Europe, Middle East & Africa, Prague, Czech Republic

Obesity is an increasing health problem in dogs. Success of weight-loss programs is often limited by compliance issues. The purpose of this study was to determine the effectiveness of a new dietetic weight management food (NDWMF)* in achieving weight loss in overweight/obese, client-owned dogs, under typical household conditions. The objectives were 1) to evaluate weight loss parameters in dogs fed a NDWMF* and 2) to assess the owner's perception of the dog's quality of life. Overweight/obese, otherwise healthy, client-owned dogs (>3/5 body condition score - BCS) were enrolled in the study ($n = 162$). Initial veterinary evaluation included physical examination, nutritional assessment, determination of ideal body weight (IBW), and development of weight-loss feeding guidelines. Daily energy requirement (DER) for weight loss was calculated as $DER = 70 \times IBW_{kg}^{0.75}$. Initial and follow-up evaluations (monthly for 6 months) encompassed determination of body weight, BCS, body fat index (BFI), muscle condition score (MCS), and feeding practices. Quality of life assessment by owners included dog's level of energy, happiness, appetite, begging behavior, flatulence, stool volume, and fecal score. Statistical analysis comprised scatterplots, regression analysis, summary statistics as appropriate for the type of analyses performed (continuous or categorical variables, distribution), and a mixed model ANOVA to assess changes over time (with statistical significance at $p < 0.05$). Ninety four percent of the dogs lost weight ($n = 153$) with an average weight loss of 14.5% (SEM, 1.1%) over 6 months and an average weekly weight-loss rate of 0.73% (SEM, 0.04%). The mean duration of weight loss was 127 days (SEM, 4.3 days) with an average of 33 days (SEM, 0.7 days) between rechecks. Thirty nine percent of dogs achieved IBW (0.39, CI: 0.31-0.48). Fifty five percent of dogs ate more calories from NDWMF* than the recommended DER for weight loss (median fed above DER=8%) and 94% of these dogs (0.94, CI: 0.87-0.98) still lost weight. Thirty six percent of dogs received treats. BCS and BFI decreased significantly over time compared to baseline. Owners perceived a significant increase in energy and happiness in the dogs that lost weight without changes in appetite or begging behavior. In conclusion, this clinical study confirmed the effectiveness of the NDWMF* in achieving weight loss in overweight/obese client-owned dogs in spite of higher than recommended caloric intake. Owners reported significant improvements in dog's quality of life without negative side effects.

* Hill's[TRADEMARK] Prescription Diet[TRADEMARK] Canine Metabolic Advanced Weight Solution, dry (caloric distribution: protein=29%, fat=30%, carbohydrate=41%)

Conflicts of interest: Becvarova and Meyer are employees of Hill's Pet Nutrition Manufacturing s.r.o.

The study was sponsored by Hill's Pet Nutrition.

ESVCP-P-1

VALIDATION OF A NEW SANDWICH-ELISA TO MEASURE FELINE HAPTOGLOBIN. J. Stiller¹, A.K. Jasensky², M. Hennies³, C. Wiene³, R. Einspamer², B. Kohn¹. ¹Clinic of Small Animals, Faculty of Veterinary Medicine, Freie universität berlin, berlin, Germany, ²Institute of Veterinary Biochemistry, Faculty of Veterinary Medicine, Freie universität berlin, berlin, Germany, ³TECOMedical Group, Rheinbach, Germany

Haptoglobin is a moderate acute phase protein in cats. As a part of the innate immune system its concentration rises within 24-48 hours after tissue damage.

Aim of the study was to validate an ELISA which was recently developed for the measurement of feline haptoglobin and to compare it with a commonly used spectrophotometric assay.

The concentration of haptoglobin was measured in 38 healthy and sick cats using a sandwich-ELISA (TECOMedical Group, Rheinbach, Germany). The validation included the detection of intra-assay and inter-assay variation, dilution linearity, spike recovery and lower detection limit. A spectrophotometric assay (Tridelta Development Ltd, Maynooth, Ireland) was used as a reference method. All samples were measured in duplicate. Statistical analysis was performed using IBM® SPSS® Statistics 20 (IBM Corporation®) and included descriptive statistics, Spearman correlation (rs) and coefficients of variation (CV).

The coefficients of variation were 2.3%, 2.9% and 4.6% for intra-assay variability and 5.1%, 8.8% and 11.2% for inter-assay variability. The ratio of observed to expected dilutional parallelism of 4 serum samples diluted 3 times ranged from 108 to 118%. The ratio of observed to expected spike recovery of 4 serum samples ranged from 91% to 94%. The lower detection limit was 0.19 mg/ml. The correlation between the 2 assays was significantly strong ($rs = 0.94$, $P < 0.001$).

The recently available sandwich-ELISA provides a high accuracy and precision and can therefore be used for the measurement of feline haptoglobin.

Conflicts of interest: The 3rd and 4th author (M. Hennies and C. Wiene) work for the company TECOMedical Group that developed the ELISA which was evaluated in the study. They provided the kits and they helped with performing the tests, but they did not have any influence on the results and the interpretation of the data.

ESVCP-P-2

VASCULAR ENDOTHELIAL GROWTH FACTOR: A BLOOD BIOMARKER IN CANINE IDIOPATHIC PULMONARY FIBROSIS? E. Roels¹, E. Krafft¹, H.P. Laurila², M.M. Rajamäki², C. Clercx¹. ¹ULG, Liege, Belgium, ²University of Helsinki, Helsinki, Finland

Canine idiopathic pulmonary fibrosis (CIPF) is a progressive interstitial lung disease that mainly occurs in the West Highland white terrier (WHWT) breed. The CIPF diagnosis commonly relies on thoracic high-resolution computed tomography (HRCT) findings and ultimately on histopathology. As those tests are not easily performed in practice, identification of measurable markers of fibrosis, that might help to diagnose and/or monitor the course of CIPF, is helpful. VEGF is an angiogenic regulator involved in a variety of physiological and pathological processes. In human IPF, serum VEGF concentration has been shown to be higher in IPF patients compared to healthy volunteers and may reflect the severity of the lung disease. The aims of the present study were (1) to investigate the potential role of VEGF as a peripheral blood biomarker in CIPF; and (2) to investigate possible breed-related differences in basal VEGF concentration, that might explain the high predisposition of the WHWT breed for CIPF. Therefore, VEGF

was determined by ELISA (Canine VEGF Quantikine ELISA Kit, R&D systems) in the serum of 14 WHWT with CIPF confirmed by HRCT and/or histopathology (median age 11 years, range 5-14), 18 healthy WHWT (9, 3-17), and 85 healthy dogs of other breeds, including: 14 Scottish terrier (ST) (5, 1-10), 16 Jack Russell terrier (JRT) (7, 1-12), 15 Maltese (6, 1-13), 14 King Charles Spaniel (KCS) (6, 1-10), 12 Labrador Retriever (LR) (6, 2-12) and 14 Malinois Belgian Shepherd (6, 2-8). Health status was based on clinical examination, serum biochemistry and haematology in all healthy dogs and a thoracic HRCT was performed in 9/18 healthy WHWT. The χ^2 test with the threshold 5% was used for the statistical analysis (XLStat[®] software). Eight CIPF WHWT (57%) have serum VEGF concentrations above the kit detection limit (39.1 pg/ml) compared to 1 WHWT (0.05%) in the group of healthy dogs ($P = 0.001$). Concerning inter-breed differences in healthy dogs, most values obtained were below the kit detection limit with only 3 KCS (21%), 3 JRT (19%), 3 LR (25%) and 1 ST (7%) having VEGF serum levels above 39.1 pg/ml ($P = 0.147$). Results of the present study show that (1) VEGF might be an interesting blood biomarker for CIPF; (2) canine VEGF Quantikine Elisa kit is not appropriate for measurement of serum VEGF levels in healthy canine populations.

Conflicts of interest: No conflicts of interest reported.

ESVCP-P-3

COMPARISON OF TOTAL PROTEIN CONCENTRATIONS IN PLEURAL AND ABDOMINAL FLUID IN DOGS ANALYSED WITH A REFRACTOMETER AND USING PENTRA 400[®]. A. Rose¹, D. Funk², R. Neiger¹. ¹Justus-Liebig University Giessen, Germany, ²Kleintierklinik, Ludwigsburg-ossweil, Germany

The total protein (TP) concentration and cell count of pleural and abdominal fluid is used to differentiate a transudate from an exudate. TP can be measured by automated wet chemistry analyser or more easily using a refractometer. The aim of this study was to assess if refractometer values of TP are useful for this purpose. Retrospectively samples from canine pleural and abdominal effusions in which TP concentration was measured both with a refractometer as well using Pentra 400 (ABX Horiba, Montpellier) were included. Samples were collected into heparinized tubes and analysed within 12 hours. Bland-Altman diagrams were created and correlation between both measurements was calculated by Spearman's non-parametric correlation.

Over a 48-months period, 93 pleural and 147 abdominal effusion samples were analysed with both techniques. Median (range) TP concentrations in pleural effusion measured by refractometer or by Pentra was 33 (2-57) g/l and 29 (7-68) g/l, respectively. Median (range) TP concentrations in abdominal effusions measured by refractometer or Pentra was 42 (2-70) g/l and 34 (2-57) g/l, respectively. TP measurement between refractometer and Pentra values were significantly correlated in pleural ($r = 0.919$, $P < 0.0001$) and abdominal ($r = 0.907$, $P < 0.0001$) effusion. The Bland-Altman graph showed a bias in the thorax and abdomen of 2.8 and 5.7.

The refractometer is an acceptable, rapid and efficient method for determination of total protein concentration in pleural and abdominal effusions in dogs to differentiate transudates from exudates.

Conflicts of interest: No conflicts of interest reported.

ESVCP-P-4

WELSH SPRINGER SPANIELS WITH COAGULATION FACTOR VII DEFICIENCY FROM FINLAND. U. Giger¹, J. Donner², K. Raj³, H. Lohi⁴. ¹Section of Medical Genetics, University of Pennsylvania, Philadelphia, United States of America, ²Genoscooper Laboratories, Helsinki, Finland, ³University of Pennsylvania, Philadelphia, United States of America, ⁴Department of Veterinary Biosciences and Research Programs Unit, Helsinki, Finland

Coagulation factor VII (FVII) deficiency has been reported in Beagles since the 1960's. Deficient dogs show a mild hemorrhagic tendency, but often remain asymptomatic and are incidentally

discovered by an isolated prolonged prothrombin time due to <10% plasma FVII activity. Factor VII deficiency occurs commonly in Beagles, Alaskan Klee Kais and Scottish Deerhounds. In these 3 breeds it is caused by a single missense mutation (c.407G>A, p.Gly136Glu) in the second epidermal growth factor-like domain of FVII, which drastically reduces the secretion and activation of FVII. Research beagles were also commonly affected which may have pharmacotoxicologically affected studies but specific DNA screening programs have been established.

We report here on the discovery of FVII deficiency in Welsh Springer Spaniels (WSS) in Finland based upon a novel screening panel for ~100 known mutations underlying inherited disorders in different canine breeds (www.mydogdna.com). Among 31 WSS initially tested, 12 were heterozygously (39%), and 1 homozygously affected for the same FVII mutation, which was confirmed by sequencing in all dogs. In order to determine whether the mutation causes FVII deficiency also in this breed, we recruited 6 littermates and their mother. None of these WSS had shown an increased hemorrhagic tendency, but affecteds bled excessively following blood collection. We found that the 3 homozygous affected dogs of the litter exhibited markedly prolonged prothrombin time but normal partial thromboplastin time. They also had drastically reduced FVII activities but normal to high FVIII and FIX activities compared to their littermate controls. The 3 heterozygous carriers tested did not show any prolongations in their prothrombin time, but had half normal FVII activity.

In conclusion, we document here the presence of FVII deficiency in WSS based upon DNA and coagulation activity testing. The common Gly136Glu mutation must have arisen prior to the separation of the very different FVII deficient breeds. There is no knowledge of an advantage of the heterozygote state. While there is only a mild hemorrhagic tendency, bleeding dogs could be treated with fresh frozen or cryo-poor plasma or human recombinant FVIIa. This preliminary study indicates a high carrier frequency in WSS. Screening by new platform DNA methods for this and other ancestral defects is helpful to detect additional hereditary diseases and genetic predispositions in different breeds, while other mutations are new and restricted to one or related breeds.

Conflicts of interest: Authors are affiliated with genetic disease screening test laboratory.

ESVCP-P-5

BREED-SPECIFIC HAEMATOLOGICAL AND SERUM BIOCHEMICAL PHENOTYPES IN THE DOMESTIC DOG. R. Chang¹, J. Lawrence¹, E.B. Erin¹, L.J. Davison², B. Szladovits¹, O.A. Garden¹. ¹Royal Veterinary College, London, United Kingdom, ²University of Oxford, Oxford, United Kingdom

Remarkably little has been published on haematological and serum biochemical phenotypes of the domestic dog. Information on the signalment and complete blood cell count of all dogs with normal red and white blood cell parameters judged by existing reference intervals was extracted from a veterinary database; similar information was collected from all dogs with normal serum biochemical profiles, considering all parameters other than glucose as inclusion criteria. Normal haematological profiles were available for 6046 dogs, 5447 of which also had machine platelet concentrations within the reference interval; normal serum biochemical profiles were available from 3045 dogs, 1495 of which also had accompanying normal serum glucose concentrations. For the haematological data, 75 pure breeds plus a mixed breed control group were represented by 10 or more dogs, while for the serum biochemical data, 60 pure breeds plus a mixed breed control group were represented by 10 or more individuals. All measured haematological parameters except mean corpuscular haemoglobin concentration (MCHC), and all serum biochemical analytes except sodium, chloride and glucose, varied with age. Concentrations of white blood cells (WBCs), neutrophils, monocytes, lymphocytes, eosinophils and platelets, but not red blood cell parameters, all varied with sex, as did total protein, globulin, potassium, chloride, creatinine, cholesterol, total bilirubin, and activities of alanine aminotransferase (ALT), creatine kinase (CK), amylase and lipase. Neutering status had an impact on haemoglobin

concentration, mean corpuscular haemoglobin (MCH), MCHC, and concentrations of WBCs, neutrophils, monocytes, lymphocytes and platelets, as well as all serum biochemical analytes except albumin, sodium, calcium, urea and glucose. Principal component analysis (PCA) of haematological data revealed 37 pure breeds with distinctive phenotypes, while PCA of serum biochemical data revealed over 50 pure breeds with distinctive phenotypes. Furthermore, all haematological parameters except MCHC and all serum biochemical analytes except urea and glucose showed significant differences between specific individual breeds and the mixed breed group. Twenty-nine breeds had distinctive haematological phenotypes and 21 breeds had distinctive serum biochemical phenotypes when assessed in this way. Tentative breed-specific reference intervals were generated for breeds with a distinctive phenotype identified by comparative analysis. This study represents the first large-scale analysis of haematological and serum biochemical phenotypes in the dog and underlines the important potential of this species in the elucidation of genetic determinants of haematological and biochemical traits, triangulating phenotype, breed and genetic predisposition, as well as the urgent need for breed-specific reference intervals in clinical practice.

Conflicts of interest: The author has received funding from BBSRC, Petplan Charitable Trust, and CRUK), but none of these grants were for this study.

ESVE-P-1 PREVALENCE OF HYPERTHYROIDISM IN PORTUGUESE CATS. R. Dias Neves, L.J.I. Horspool. MSD Animal Health, Paço de arcos, Portugal

Hyperthyroidism is a common feline endocrinopathy. Anecdotal reports suggest this disorder is rare within the Portuguese feline population. A descriptive, cross-sectional study was set-up to describe the point prevalence of hyperthyroidism in Portugal from mid-March 2013 to the end of May 2014.

For each cat aged 10 years or older, irrespective of their suspected thyroid status, presented to eight veterinary practices in Portugal, the veterinarian and the pet owner had to complete a questionnaire and the veterinarian had to take a venous blood sample (into a plain tube) from the cat, after obtaining signed owner consent. The veterinary questionnaire included history, attitude, activity, heart rate and thyroid palpation. Cats aged <10 years and those diagnosed previously with hyperthyroidism were excluded. Blood samples were centrifuged and the serum harvested and stored frozen until collection by the laboratory within 2 days of sampling. Total T4 was measured using a chemiluminescent method (Immulite 1000, Siemens). Cats were classified as hyperthyroid, equivocal or euthyroid based on a total T4 concentration of >51 nmol/L, 30-51 nmol/L or <30 nmol/L, respectively. Repeat measurement of total T4 after 4-6 weeks was recommended for all equivocal cases. The individual cat was the statistical unit. Descriptive statistics was used to summarise the data and associations between different clinical signs analysed using Chi-square, Fisher's exact test or the Mann-Whitney U test. The level of significance was set at 0.05.

Thirty cats were excluded from the prevalence analysis because they were aged <10 years (between 4 and 9 years, n = 11) or their age was not stated (n = 19, four of these cats were hyperthyroid). By the end of February 2014, samples had been submitted from 197 cats that met the inclusion criteria. Based on the thyroid hormone analysis, there were 14/197 (9%) hyperthyroid, 28 (14%) equivocal and 152 (77%) euthyroid cats. Very few follow-up blood samples were taken.

Hyperthyroidism appears to be not uncommon in Portuguese cats. Getting owners to return for follow-up blood sampling appears to be problematic. Under-reporting of hyperthyroidism appears to be a significant problem in Portugal, as has been reported for some other countries. Thyroid palpation should form part of routine physical examinations, especially of middle aged and older cats. Older cats in Portugal should be screened for hyperthyroidism even in the absence of a detectable thyroid nodule.

Conflicts of interest: Both authors are employees of MSD Animal Health. MSD Animal Health funded the study. MSD Animal Health has an approved veterinary medicinal product for

the treatment of feline hyperthyroidism. This product is available commercially in some EU markets but not in Portugal.

ESVE-P-2 ESTABLISHMENT OF A PROTOCOL FOR THE ISOLATION OF PURE PANCREATIC ISLETS OF LANGERHANS IN CATS. I. Rito Brandao¹, L. Whiting¹, E. Zini², C.E. Reusch², T. Lutz¹, M. Osto¹. ¹Institute of Veterinary Physiology, University of Zurich, Zurich, Switzerland, ²Clinic for Small Animal Internal Medicine, University of Zurich, Zurich, Switzerland

Diabetes mellitus is one of the most commonly encountered endocrinopathies in cats and its prevalence has increased in the past. Similar to human Type 2 Diabetes, feline Diabetes is associated with comparable lesions occurring in the pancreatic islets, namely islet amyloidosis and beta-cell loss. Studying the pathophysiology of feline diabetes and the molecular mechanisms through which glucose metabolism is disturbed is largely hampered by the lack of a method for the isolation of pure pancreatic islets. The aim of this project was to improve a previously established method for the isolation of pancreatic islets; in particular enhancing the purity of isolated islets in this species.

Cats that died or were euthanized due to severe illness other than pancreatitis or other pancreatic disease were enrolled. Pancreata were perfused *post-mortem* with 50 ml Collagenase Type IV (0.5 mg/ml) through the pancreatic duct. The perfused organ was then digested for 30', 40' and 60' at 37 °C in a water-bath and purified using a filtration method. Islet cell viability and purity were determined by Thiazolyl Blue Tetrazolium Bromide (MTT assay) and dithizone staining, respectively.

Perfusing the pancreas through the pancreatic duct allowed collagenase to access the islets using anatomical structures and to improve islet yield compared to previously established protocols in this species. The digestion time of 60' provided the best islet yield. After digestion, feline pancreatic islets remained satisfactorily viable for 5 days in the culture system following regular media changes.

The current study has successfully optimized the isolation, purification and culture maintenance of feline islets. The successful yield and viability of islets isolated through the suggested protocol may provide promising potential as a source of islets for diabetes research in cats.

Conflicts of interest: No conflicts of interest reported.

ESVE-P-3 NESIDIOBLASTOSIS IN A CAT. L.E. Hambrook¹, A.A. Ciavarella¹, J.S. Nimmo², J. Wayne². ¹Advanced Vetcare, Melbourne, Australia, ²Australian Specialised Animal Pathology Laboratory, Melbourne, Australia

Nesidioblastosis describes a syndrome of acquired hyperinsulinaemia and associated hypoglycaemia secondary to focal or diffuse (non-neoplastic) beta cell hyperplasia within the pancreas. Beta cell dysregulation is thought to occur secondary to pancreatic injury. This syndrome has been reported in humans with increasing frequency, but it has not previously been described in domestic pets.

A six year old, de-sexed female British Shorthair cat presented with acute onset weakness and mental dullness. Upon initial presentation the cat was mildly hyperglycaemic (9.9 mmol/L; 3.3-6.7 mmol/L). Over the following 12 hours the cat developed central blindness, tremors, intermittent seizures and opisthotonus. Repeat blood sampling revealed a marked hypoglycaemia (0.8 mmol/L). An insulin level (performed on serum obtained while the cat was hypoglycaemic) was inappropriately elevated (10938 pmol/L; reference range 72 - 583 pmol/L). An intravenous bolus of 5% glucose resulted in rapid resolution of all clinical signs and mild transient hyperglycaemia (12.5 mmol/L). Despite frequent feeding, the hypoglycaemia (2.0 mmol/L) recurred, so an intravenous glucose

continuous rate infusion was commenced. An abdominal ultrasound was unremarkable, although three cranial mesenteric lymph nodes were noted to be prominent (3 mm in width). An exploratory laparotomy revealed a firm and erythematous left limb of the pancreas. The body and right limb of the pancreas appeared grossly normal. Following surgical resection of the left limb of the pancreas, the cat returned to a euglycaemic state after a brief rebound hyperglycaemia. Histopathology revealed pancreatic fibrosis with marked multifocal micronodular hyperplasia of exocrine and endocrine cells, mild lymphoplasmacytic inflammation and ductular ectasia. Synaptophysin immunohistochemistry confirmed nodular beta cell hyperplasia. Mild granulomatous lymphadenitis and hydropic change within hepatocytes was also noted. The cat recovered uneventfully without any further intervention. It gained weight and remained euglycaemic over the following six months.

While beta cell hyperplasia has been reported as an incidental histopathological finding in euglycaemic young Beagles, this is the first reported case of clinically significant hypoglycaemia secondary to nesidioblastosis in a domestic pet. While this condition is rare, nesidioblastosis is being increasingly recognised in the human field and it is an important differential to consider when investigating hypoglycaemia as it cannot be differentiated from insulinoma without histopathological evaluation. Age of onset may provide a clue to this non-neoplastic disease, as this cat was much younger than all previously reported cases of feline insulinoma (all > 12 years of age at diagnosis). While recurrence has been reported in humans, a favourable outcome is anticipated following partial pancreatectomy.

Conflicts of interest: No conflicts of interest reported.

ESVE-P-4

FELINE HYPERTHYROIDISM IN PORTUGAL. H. Vilhena¹, A. Martins², P. Almeida², P. Ferreira³, I. Fonseca⁴, T. Lima², A. Ribeiro⁵, N. Carolino⁶, A.C. Silvestre-Ferreira⁷. ¹Escola Universitária Vasco da Gama / Hospital Veterinário Baixo Vouga / CE-CAV, Portugal, ²Faculdade Medicina Veterinária Universidade Lusófona Humanidades e Tecnologias, Lisboa, Portugal, ³Laboratório Segalab, Leça do balio, Portugal, ⁴Hospital Veterinário do Baixo Vouga, Ageda, Portugal, ⁵Policlínica Veterinária de Aveiro, Aveiro, Portugal, ⁶Instituto Nacional de Investigação Agrária e Veterinária, Santarém, Portugal, ⁷Departamento Ciências Veterinárias Universidade de Trás-os-Montes e Alto Douro, Vila real, Portugal

Hyperthyroidism is the most common feline endocrinopathy of geriatric cats worldwide. Nonetheless, data concerning the accurate prevalence of feline hyperthyroidism (FH) is scarce; and apparently exhibit geographical variation, which can be an important instrument in investigating risk factors through the analysis of exposure to different factors in areas of high and low prevalence. In Europe, FH is considered more frequent in the northern than in the southern countries.

The aims of this study were to determine the occurrence of FH in a region of Portugal, to characterize clinical presentation and potential risk factors.

During an 18-month period, 120 geriatric cats \geq eight years from Aveiro (Central region of Portugal) were selected. Cats were excluded if presented in shock or moribund, or in treatment with drugs that might affect total T4 (TT4) serum concentration. The TT4 concentration was determined through chemiluminescence (Immulite[®], Siemens), and diagnosis of FH established if TT4 serum concentration \geq 4.0 μ g/dl (reference values 0.8–3.9 μ g/dl) associated with compatible clinical signs.

Information on age, gender, breed, weight, housing conditions (indoor vs outdoor), use of external parasiticides, food (dry vs canned food and flavor), use of litter box, environment, clinical signs and laboratory data was collected. All owners gave informed consent.

Population studied included 49 males (40.8%) and 71 females (59.2%), mainly domestic short-haired cats (88.3%). Ages ranged from eight to 21 years old (11.8 \pm 3.0).

Eight (6.7%) cats were diagnosed with hyperthyroidism. If only cats \geq 10 years of age were considered (n = 89), prevalence raised to 9.0%. Hyperthyroid population comprised four males

and four females, ranging from 13 to 21 years old (15.6 \pm 2.7). Increasing age (p = 0.001), polyphagia (p 0.001), weight loss associated with increased (p 0.001) or normal appetite (p 0.001), presence of thyroid uni or bilateral nodules (p 0.001), vomiting (p = 0.04) and hyperactivity (p = 0.006) were significantly associated with hyperthyroidism in the geriatric population studied. Environment was also significantly associated with development of FH (p 0.001), with cats from urban or semi-rural areas at higher risk of developing the disease than cats living in a rural environment. No other significant associations were found between hyperthyroidism and other factors analyzed.

In our knowledge, no epidemiologic studies on FH have been performed in Portugal, a country where the occurrence is believed to be low, but in which the population of pet cats, the feline geriatric population and the clinical cases diagnosed have been increasing.

Conflicts of interest: The study was partially supported by Laboratório Segalab S.A. and Dechra Veterinary Products.

ESVE-P-5

EVIDENCE OF AUTOIMMUNITY IN A POPULATION OF DIABETIC DOGS FROM THE CANARY ISLANDS. Y. Brito-Casillas¹, C. Melán¹, J.C. Wiebe¹, L. López-Ríos¹, O. Quesada², A. Holder³, B. Catchpole³, A.M. Wagner¹. ¹Instituto Universitario de Investigaciones Biomédicas y Sanitarias, Las palmas de gran canaria, Spain, ²Comparative Pathology Dept., Veterinary Faculty, UPLGC, Arucas, Spain, ³Pathology & Infectious Diseases Dept., Royal Veterinary College, Univ. of London, London, United Kingdom

Canine diabetes mellitus (cDM) has been proposed to be a spontaneous animal model of human autoimmune diabetes, and comparative research can be undertaken to investigate the interaction between genetic and environmental factors. Most epidemiological studies of cDM have been performed in northern European and North American populations.

Our aim was to evaluate the epidemiology and clinical features of the diabetic dog population from the Canary Islands, with special focus on immune-mediated disease.

Dogs attending our Veterinary Teaching Hospital were included from January 2009 to January 2012. Previously diagnosed and new cases were considered. Prevalence was calculated as number of cDM/total number of dogs attending the hospital and incidence as newly diagnosed cases divided by the same value per year. Anti-insulin antibodies were assessed by ELISA. Genotyping for dog leukocyte antigen (DLA) and measurement of canine anti-GAD65 and anti-IA2 antibodies by radio-immunoprecipitation assay were performed in dogs with suspected immune-mediated diabetes.

Twenty-nine dogs with cDM were identified from a mean population of 4302 (3741-4581) dogs per year (mean prevalence 0.23% and mean incidence cases per year 16 per 10,000). Age at diagnosis was 9.45 years (range: 3.5-14y). Most dogs were not neutered (87% females; 83% males). Nine breeds were represented, including Poodle (21%) and Andalusian wine-cellar rat-hunting dog (7%). Seasonality was observed in the diagnosis with peaks in December and March-April. Diabetes was classified as dioestrus diabetes (55%), idiopathic/immune-mediated (21%), iatrogenic (7%) and secondary to pancreatitis (14%) or other endocrine disorders (3%).

Insulin-treated dogs were negative for anti-insulin antibodies (n = 19). From the suspected immune-mediated cases (n = 5), autoantibody reactivity was shown in two cases (anti-GAD65, n = 1; anti-IA2, n = 1). No previously described, diabetes-risk DLA-types were identified.

Although age, prevalence and incidence did not differ from previous studies, the high proportion of entire females likely explained the high frequency of dioestrus diabetes. The Andalusian wine-cellar rat-hunting dog was identified as a high-risk breed for cDM. Most of the DLA-types seen have not been previously described, but at least two have been associated with increased risk of autoimmunity in dogs. Further population-based studies are needed in different regions, to assess the heterogeneous nature of this disease.

Conflicts of interest: No conflicts of interest reported.

ESVE-P-6
COMPARISON OF THE PLASMA CORTISOL-DEHYDROEPIANDROSTERONE-RATIO IN HEALTHY DOGS AND DOGS WITH HYPERADRENOCORTICISM. S. Hoffrogge, R. Mischke, M. Piechotta. University of Veterinary Medicine Hannover, Foundation, Hannover, Germany

The cortisol-dehydroepiandrosterone (DHEA)-ratio is widely used in human medicine as a marker for stress however it is not clear whether it could also help in distinguishing hyperadrenocorticism (HAC) from other diseases which might have a negative impact on the outcome of a dexamethasone low dose test. Therefore the aim of the study was to evaluate the cortisol-DHEA-ratio as an additional diagnostic marker for HAC in dogs. To achieve this aim, a reference range of this ratio depending on the sex should be evaluated in healthy dogs and compared with dogs having a HAC. In 55 healthy dogs (age: 1 - 11.4 years) and in 20 dogs with HAC (age: 7.1 - 14.6 years) of different breeds the plasma concentration of cortisol (Immulite System, Siemens Healthcare Diagnostics) and DHEA (Beckman Coulter) was measured and the ratio was calculated. All dogs were patients of the Small Animal Clinic except five of the healthy dogs which were recruited from the Institute of Pharmacology, Toxicology and Pharmacy of the University. With these data the cortisol-DHEA-ratio was calculated for male dogs (healthy dogs $n = 18$; dogs with HAC $n = 3$), neutered males (healthy dogs $n = 9$; dogs with HAC $n = 5$), female dogs (healthy dogs $n = 21$; dogs with HAC $n = 5$) and spayed females (healthy dogs $n = 7$; dogs with HAC $n = 7$). The statistical analysis was performed with Sigma Stat. The plasma cortisol-DHEA-ratio of healthy male dogs was the lowest ratio of all sexual categories (mean average 84.8 ± 128) and it differed significantly to all other sexes (neutered males = 231 ± 138 , $P = 0.002$; females = 244 ± 124 , $P < 0.001$ and spayed females (183 ± 60.0 , $P = 0.006$). The cortisol-DHEA-ratio showed no significant difference between male and female dogs with HAC. Spayed females with HAC had significantly higher cortisol-DHEA-ratios (501 ± 310) than healthy spayed females ($P = 0.035$) but no significant differences were found in other sexual categories. This preliminary data indicates that the cortisol-DHEA-ratio might not be a very promising tool for the diagnosis of HAC. In addition, the significant gender-dependency of this parameter has to be considered and may generally limit its clinical usefulness. This study is financially supported by the Bruns-Stiftung.

Conflicts of interest: No conflicts of interest reported.

ESVE-P-7
PREVALENCE OF FELINE HYPERTHYROIDISM AND INTRINSIC RISK FACTORS IN A CLINIC POPULATION IN SOUTHERN GERMANY. A. Wehner, I.I. Koehler, K. Hartmann. Clinic of Small Animal Medicine, Munich, Germany

Hyperthyroidism is common in older cats. The aim of this study was to assess the prevalence of feline hyperthyroidism and potential intrinsic risk factors in a hospital population in Southern Germany.

Total thyroxine (T4) was prospectively measured by enzyme immunoassay (EIA) in sera of 425 cats older than 8 years that were presented to the Clinic of Small Animal Medicine. A standardized physical examination was performed, and body condition score (BCS) and thyroid palpation score (TPS) were assessed. Association between signalement, BCS and TPS was analyzed by Student's unpaired t-test, Chi-Square, and Mann-Whitney test. Level of significance was set at 0.05.

Fifty nine cats were diagnosed with hyperthyroidism leading to a prevalence of 13.9% (CI 10.9-17.5). Hyperthyroid cats were older than non-hyperthyroid cats ($p < 0.0001$) and more often female ($p = 0.024$, odds ratio 1.920). Domestic short or long hair cats were more often affected than pedigrees ($p = 0.015$). Hyperthyroid cats had higher TPS ($p < 0.0001$) and lower weight than non-hyperthyroid cats ($p < 0.0001$) although BCS was not different ($p = 0.290$). In 11 (2.5%) cats, the elevated T4 was an incidental finding. In 3 of those, the disease was confirmed later (the others were dead due to unrelated diseases). In 152 patients, hyperthyroidism was considered a differential diagnosis and was confirmed in 32 (21.1%) cats although in 5 cats additional diagnostic means were necessary.

Older female domestic cats are predisposed to hyperthyroidism which is frequently diagnosed after the initial clinical suspect. In a few affected cats an elevated T4 is not present or can precede clinical signs.

Conflicts of interest: The study was partially funded by MSD Intervet.

ESVE-P-8
ENHANCED DIAGNOSIS SYSTEM FOR FELINE DIABETES MELLITUS. M. Rosca¹, L. Ferariu², A. Burlacu², G. Solcan¹. ¹USAMV, Iasi, Romania, Iasi, Romania, ²Gheorghe Asachi Technical University of Iasi, Iasi, Romania

The main endocrinopathy affecting both humans and pet felines is diabetes mellitus. Accurate diagnosis is the most important aspect in the future outcome of the disease. A computer based Decision Support System (DSS) is targeted on assisting clinicians with one or more steps of the diagnostic process. The novelty of our DSS emerges from the possibility of assisting both clinical and paraclinical diagnosis stages of diabetes mellitus and all common combination of disorders associated with this endocrinopathy. The motivation behind the development of such system is the desire to maximize the reliability of clinical decisions.

The design of our feline diabetes mellitus DSS emerges from the syndrome of polyuria-polydipsia, with the possibility of spotting the accompanying pathologies. Fuzzy logic is used for dealing with knowledge representation and uncertainty. The fuzzy rules proposed to represent this knowledge emerge from anamnesis, clinician's input, clinical and paraclinical description, and confirmation diagnostic tests. Clinical signs such as polyuria-polydipsia, persistent hyperglycemia, polyphagia, weight fluctuations, administration of drugs with a diabetogenic potential, were considered decisive in the pattern of diagnosis establishment. Registered medical records of 29 cats, 16 males and 13 females, whit ages from 7 to 18 years old, were analyzed in order to validate the DSS. Using Matlab software, the DSS was implemented and tested. For any case with polyuria-polydipsia the system provides, via a friendly graphical user interface, the diagnosis with the highest probability. The set of diagnoses which can be generated by the DSS consists in: a) diabetes mellitus; b) diabetes mellitus induced by (b.1) hypersomatotropism, (b.2) hyperthyroidism, (b.3) hyperadrenocorticism and (b.4) diabetogenic medication; c) diabetes mellitus in association with (c.1) chronic kidney failure and (c.2) heart failure; d) ketoacidotic diabetes mellitus; e) pancreatitis. The DSS was applied with success on all 29 cases, revealing the following diagnoses / no of cases: (a) - 8, (b.1) - 2, (b.2) - 1, (b.4) - 8, (c.1) - 3, (c.2) - 3, (d) - 4.

An adequate treatment protocol requires an accurate and complete diagnosis. Advanced computational systems accompany clinicians in their decision making, leaving a reduced space for medical errors and superfluous, expensive and time consuming tests. Future work will be targeted on exploring the possibilities of combining the DSS with an artificial neural network model for diabetes mellitus. This can be the foundation of a complete case oriented management system for feline diabetes mellitus and associated disorders.

Conflicts of interest: No conflicts of interest reported.

ESVIM-P-1
ACTIVIN B, BUT NOT ACTIVIN A, IS UPREGULATED IN LUNGS OF WEST HIGHLAND WHITE TERRIERS WITH CANINE IDIOPATHIC PULMONARY FIBROSIS. L.I.O. Lilja-Maula¹, P. Syrjä¹, H.P. Laurila¹, E. Sutinen², M. Palviainen³, K. Kolli³, M.M. Rajamäki¹, M. Myllärniemi². ¹University of Helsinki, Helsinki, Finland, ²University of Helsinki and Helsinki University Central Hospital, Helsinki, Finland, ³Haartman Institute, University of Helsinki, Helsinki, Finland

Activins are cytokines belonging to the transforming growth factor (TGF)- β superfamily. It is thought that activins may be

the key intermediary in TGF- β 1 mediated fibrotic response. Activin A has been suggested to participate in the pathogenesis of human idiopathic pulmonary fibrosis (IPF), but studies regarding the role of activin B are still sparse. Canine IPF (CIPF) is a chronic, incurable interstitial lung disease occurring particularly in West Highland White Terriers (WHWTs). During the disease course, acute exacerbations (AEs), with poor prognosis, can occur. Histopathologically AEs of CIPF are featured by diffuse alveolar damage, which is also a key feature in acute respiratory distress syndrome (ARDS). Our objective was to study the expression of activin A and B by immunohistochemistry in the lung tissue of CIPF WHWTs (n = 5), CIPF WHWTs with concurrent AE (n = 4), and dogs of various breeds with ARDS (n = 4), and to compare these findings to healthy WHWTs (n = 3). In addition, western blot analysis of activin B from bronchoalveolar lavage fluid (BALF) of CIPF WHWTs (n = 6) and healthy WHWTs (n = 6) was conducted. We demonstrated that activin B, but not activin A, is strongly expressed in the altered alveolar epithelium in lungs of diseased WHWTs as well as in ARDS lungs. Furthermore, activin B was detected in BALF of CIPF WHWTs, most notably in samples from dogs with AE, but not in BALF of healthy WHWTs. This novel finding suggests that activin B participates in the pathophysiology of CIPF and might act as a potential marker of alveolar epithelial damage.

Conflicts of interest: No conflicts of interest reported.

ESVIM-P-2
MORBIDITY IN THE NOVA SCOTIA DUCK TOLLING RETRIEVER WITH A FOCUS ON IMMUNE-MEDIATED DISEASE. H. Bremer¹, Å. Vilson¹, B.N. Bonnett², H. Hansson-Hamlin¹. ¹Swedish university of agricultural sciences, Uppsala, Sweden, ²B.Bonnett Consulting, Georgian bluffs, Canada

Dogs of the breed Nova Scotia Duck Tolling Retriever (NSDTR) are affected by several immune-mediated diseases, in particular steroid-responsive meningitis-arteritis (SRMA) and an immune-mediated rheumatic disease (IMRD). IMRD is a systemic lupus erythematosus-related disease characterized by chronic stiffness and pain in several joints. The aim of this study was to investigate the morbidity in NSDTRs and to test the hypothesis that NSDTRs are predisposed to SRMA and IMRD. Insurance data from a Swedish insurance company (Agria insurance company, Stockholm, Sweden) from 1995-2006 was used for the study. Approximately one third of Swedish dogs are insured by Agria and the insurance database is a validated tool for epidemiological studies. Assessment of morbidity was based on veterinary care events. Disease diagnoses were grouped in both general and specific disease categories. Individual diagnoses that were likely to represent IMRD were combined. Morbidity was defined as incidence rates and presented as number of cases per 10 000 dog years at risk (DYAR). Relative risk (RR) for NSDTRs compared to other breeds combined was calculated. The study included 445 336 dogs, 2890 were NSDTRs. The most common general causes of veterinary care for NSDTRs were injuries followed by gastrointestinal and musculoskeletal disorders with significant increased risk (RRs between 1.2 and 1.3) for NSDTRs compared to other breeds. The highest relative risk for NSDTRs was for systemic lupus erythematosus (RR 19.0). Compared to other breeds, NSDTRs had an increased risk for SRMA (RR 11.5) and IMRD (RR 11.8) with an incidence rate of 19.6 cases per 10 000 DYAR for SRMA and 8.8 cases per 10 000 DYAR for IMRD. The incidence rate for SRMA and IMRD in NSDTRs were also compared to dogs of other retriever breeds. The comparison revealed that NSDTRs also had a significant increased risk for both SRMA (RR 20.8) and IMRD (RR 10.1) when compared to other retrievers only. This study is the first to investigate the morbidity for IMRD in NSDTRs, which is important for further research and breeding practice. For several reasons the incidence rates might be underestimated and exact numbers should be interpreted with caution. However underestimation of incidence rates should not differ between dogs of different breeds, therefore not affecting the risk calculations. It can be concluded that NSDTRs are predisposed to the diseases SRMA and IMRD with an increased risk compared to other breeds and to other retrievers.

Conflicts of interest: Brenda N. Bonnett consults with Agria insurance company on various projects. Agria insurance company has also funded work leading to the development of the insurance data base that my study was based on.

ESVIM-P-3
VIRAL CO-INFECTIONS IN DOGS WITH BACTERIAL PNEUMONIA. S.J. Viitanen, M.M. Rajamäki. University of Helsinki, University of helsinki, Finland

Canine infectious respiratory disease (CIRD) is a multifactorial contagious disease caused by respiratory viruses and selected bacterial pathogens. CIRD has been shown to be a predisposing factor in the development of bacterial pneumonia (BP) in dogs housed in dense populations such as kennels and rehoming centers. The aim of this study was to determine the prevalence of viral co-infection and to assess its effects on disease severity in household dogs diagnosed with BP.

A prospective cross-sectional observational study was conducted and 20 dogs diagnosed with BP caused by opportunistic bacteria were included. 13 dogs with chronic (> 30 days) tracheobronchitis caused by *Bordetella bronchiseptica* were included as controls for virus analysis. Diagnosis was confirmed by thorough clinical examinations as well as with cytological and bacterial analysis of bronchoalveolar lavage (BAL) or transtracheal wash (TTW) samples. Canine parainfluenzavirus (CPIV), Canine adenovirus, Canine herpesvirus, Canine distempervirus, Canine respiratory coronavirus (CRCoV) and Canine pneumovirus were analysed in BAL or TTW samples using RT-PCR assay.

CPIV was detected in 7/20 (35%) and CRCoV in 1/20 (5%) respiratory samples in dogs with BP. Respiratory viruses were not detected in dogs with chronic tracheobronchitis. There were no significant differences in the duration of hospitalization (p = 0.427) or arterial pO₂ at presentation (p = 0.343) between BP dogs with and without a viral co-infection. These results indicate that co-infections with respiratory viruses are common also in household dogs with BP. Additionally, viral co-infections did not cause a more severe course of BP.

Conflicts of interest: The author's research is financially supported by the Finnish Foundation of Veterinary R and the Finnish Veterinary Foundation.

ESVIM-P-4
CAUSES OF CANINE ANEMIA IN TAIWAN: A FIVE-YEAR RETROSPECTIVE SURVEY. E.C.Y. Lin¹, P.C. Liu¹, L.L. Chueh¹, B.L. Su². ¹Graduate Institute of Veterinary Medicine, National Taiwan University, Taipei, Taiwan, ²Institute of Veterinary Clinical Sciences, National Taiwan University, Taipei, Taiwan

Anemia is a common hematologic disorder in dogs, however, few data are available regarding epidemiology and causes in Taiwan. To investigate the causes of anemia, 3174 anemic cases (PCV < 37%) collected between January 2008 and December 2012 at National Taiwan University Veterinary Hospital (NTUVH) were analyzed. Most dogs (72.1%, n = 2889) presented with a mild form (25% ≤ PCV < 37%), which was followed by a moderate form (15% ≤ PCV < 25%; 20.8%, n = 660) and a severe form (PCV < 15%; 7.1%, n = 225). Among the 2037 dogs with identifiable causes, 70.4% (1435 dogs) were induced by single cause, whereas 29.6% (602 dogs) by multiple causes. Neoplasia-related anemia (n = 460), infectious pathogens-related anemia (n = 287), renal disease-related anemia (n = 251) and post-surgery/trauma-related anemia (n = 182) account for 32.1, 20.0, 17.6 and 12.7% of single-cause cases, respectively. Furthermore, 123 of them (8.6%) presented with severe anemia. Severe anemia primarily resulted from infectious disease-related anemia (61.8%), followed by IMHA (11.4%), and tumor-related anemia (11.4%). Of the 76 infectious disease-related severe anemic dogs, the most common diagnosed pathogen was *Babesia gibsoni* (88.2%, n = 67), followed by *Ehrlichia canis* (6.6%, n = 5) and *Babesia canis* (3.9%, n = 3). Taken together, tumors, infectious diseases, and renal failure are the most frequently causes of canine anemia in

Taiwan, furthermore, *B. gibsoni* appeared to be the most important infectious pathogen causing severe anemia which may be associated with the climate in this geographical area.

Conflicts of interest: No conflicts of interest reported.

ESVIM-P-5

DETECTION OF MYCOPLASMA CANIS AND MYCOPLASMA CYNOS BY SPECIFIC QUANTITATIVE POLYMERASE CHAIN REACTION ASSAYS IN THE BRONCHOALVEOLAR LAVAGE FLUID IN DOGS INFECTED WITH BORDETELLA BRONCHISEPTICA. M. Canonne-Guibert¹, E. Roels¹, E. Ramery¹, F. Billen¹, I.P. Peters², C. Clercx¹. ¹University Teaching Hospital, Liège, Belgium, ²TDDS Ltd., The Innovation Centre, University of Exeter, Exeter, United Kingdom

Bordetella bronchiseptica (*Bb*) is one of the primary causative agents of canine infectious respiratory disease (CIRD). This contagious disease, commonly seen in young dogs, is often self-limiting, although a wide range of respiratory signs can be found, from mild illness to severe pneumonia leading to death. Although *Mycoplasma cynos* (*M. cynos*) was recently identified as an emerging and possibly lethal pathogen in CIRD¹, the role of *M. canis* and *M. cynos* as primary respiratory pathogens still remains unclear. Detection of these bacteria is now improved by quantitative polymerase chain reaction (qPCR). In dogs with CIRD due to *Bb*, the frequency of co-infection with *Mycoplasma spp.* in particular *M. cynos*, and their possible role in the severity of the clinical signs are unknown.

The aim of the present study was to investigate the presence of *M. canis* and *M. cynos* in a population of dog infected with *Bb*, compared with 2 other populations: healthy dogs and dogs with bacterial bronchopneumonia where *Bb* was not involved (BBP).

Therefore, *Bb*, *M. canis* and *M. cynos* were detected by qPCR in the bronchoalveolar lavage fluid (BALF) sample in 16 dogs with *Bb* (mean age = 0.9 y, mean BW = 8.8 kg), 10 dogs with BBP (3.9 y, 15.0 kg), and 10 healthy dogs (6.1y, 20.2 kg). Bordetellosis was diagnosed based on clinical findings together with demonstration of pleomorphic cocci/coccobacilli adhering to the cilia of the epithelial cells on cytospin BALF preparations, and positive qPCR on BALF. A clinical severity index (CSI 0 to 12) was assigned based on clinical signs (cough 0-2, dyspnea 0-2, lethargy 0-1, fever 0-1), thoracic radiographic pattern (0-3), and BALF score (0-3). BBP was diagnosed based on clinical findings, BALF cytology and culture.

M. canis was indifferently detected in healthy (5/10, 50%), BBP (4/10, 40%) and *Bb* dogs (3/16, 19%) while *M. cynos* tended to be more frequently detected in *Bb* group (8/16, 50%) than in healthy (2/10, 20%) and BBP dogs (1/10, 10%) ($\text{K}\chi^2$ test, $p = 0,068$). In *Bb* dogs, no correlation could be detected between CSI and presence of *M. cynos* ($\text{K}\chi^2$ test $p = 0,26$).

In conclusion, the present data suggest that, in CIRD, coinfection with *Bb* and *M. cynos* is frequent, but is not correlated with clinical disease severity. Further studies are required to investigate whether coinfection of *Bb* and *M. cynos* deserves specific therapeutic considerations.

Conflicts of interest: No conflicts of interest reported.

ESVIM-P-6

DETECTION OF BORDETELLA BRONCHISEPTICA, MYCOPLASMA CANIS AND MYCOPLASMA CYNOS BY SPECIFIC QUANTITATIVE POLYMERASE CHAIN REACTION ASSAYS IN THE BRONCHOALVEOLAR LAVAGE FLUID OF DOGS WITH EOSINOPHILIC BRONCHOPNEUMOPATHY. M. Canonne-Guibert¹, E. Roels¹, F. Billen¹, I.P. Peters², C. Clercx¹. ¹University Teaching Hospital, Liège, Belgium, ²TDDS Ltd., The Innovation Centre, University of Exeter, Exeter, United Kingdom

Canine idiopathic eosinophilic bronchopneumopathy (EBP) is a disease characterized by eosinophilic infiltration of the lung and bronchial mucosa in young adults. Aetiology remains

unclear although immunologic hypersensitivity is clearly suspected, while inciting antigens are generally unidentified. In humans as in cats, infections with *Mycoplasma spp.* have been discussed as potential triggers in inflammatory bronchial disease^{1,2}. *Bordetella bronchiseptica* (*Bb*) is a recognized pathogen agent of canine infectious tracheobronchitis. Detection of *Bb* and *Mycoplasma spp.*, especially *Mycoplasma cynos* (*M. cynos*), and their potential role of in canine inflammatory bronchitis, have not been investigated.

The aim of the present study was to investigate the frequency of *Bb*, *Mycoplasma canis* (*M. canis*) and *M. cynos* in canine EBP. Therefore, presence of *Bb*, *M. canis* and *M. cynos* were retrospectively assessed by quantitative polymerase chain reaction (qPCR) in bronchoalveolar lavage fluid (BALF) samples from 18 dogs with EBP (mean age = 4.5 y, mean body weight = 21.1 kg) as well as in 8 dogs with aspecific chronic bronchitis (7.3 y, 22.3 kg). Based on clinical signs, a clinical severity score (CSS, 0-5) was assigned each EBP dog.

Although all BALF culture and cytology were negative for this bacteria, *Bb* was more frequently detected by qPCR in EBP dogs (5/18, 28%) than in CB dogs (0%) ($\text{K}\chi^2$ test, $p = 0,009$). Presence of *Bb* in EBP dogs was independent of age but significantly associated with CSS ($\text{K}\chi^2$ test, $p = 0,041$).

Results of qPCR were positive for *M. canis* and *M. cynos* in 6 (33%) and 2 (11%) EBP dogs and in 1 (13%) and 1 (13%) CB dogs, respectively. There was no difference between the 2 groups for any of the organisms. Any relation between age or CSS and presence of *M. spp* in EBP dogs was observed.

In conclusion, *M. canis* and *M. cynos* do not seem to be predominantly involved in the pathogenesis of canine EBP. However, *Bb* is more frequently detected in BALF from EBP dogs than from dogs with aspecific CB and its presence is associated with clinical severity. Whether *Bb* is able to trigger eosinophilic inflammation or is only more easily collected in an inflamed environment is unclear. But EBP dogs could potentially act as *Bb* carriers and source of infection. Therefore, *Bb* should be systematically searched for in canine EBP cases and treated accordingly.

Conflicts of interest: No conflicts of interest reported.

ESVNU-P-1

IS DISTAL RENAL TUBULAR ACIDOSIS UNDERDIAGNOSED IN DOGS PRESENTING WITH IMHA? M.D. Alferez-Reyes¹, B. Griensteidl², S. Kilpatrick¹, A. Ridyrd¹. ¹University of Edinburgh, Roslin, United Kingdom, ²Ambivet Veterinary Group, Heanor, United Kingdom

Distal renal tubular acidosis (dRTA) was recently reported in three dogs with IMHA. The purpose of this study was to explore the hypothesis that dRTA is an underdiagnosed concurrent disorder in dogs with IMHA. We report the clinical presentation and outcome of three dogs where the combination of IMHA and dRTA was strongly suspected.

The medical records of dogs diagnosed with IMHA at the University of Edinburgh Hospital for Small Animals between January 2008 and May 2013 were reviewed to identify cases where venous blood gas analysis and urinalysis had also been carried out. For the purpose of this retrospective study IMHA was defined by the presence of anaemia with PCV < 30%, and one or more of the following criteria; a positive slide agglutination test, positive Coombs' test or moderate to marked spherocytosis. The criteria for diagnosis of dRTA included moderate to marked hyperchloremic metabolic acidosis with a normal anion gap; urine pH (> 6.0) in the face of metabolic acidosis; hypokalaemia (< 3.5 mmol/l).

Fifty-seven records were evaluated, with 39 cases being excluded due to insufficient clinical information, including inability to determine urinary pH due to the severity of pigmenturia in four cases. Of the 18 cases where there was sufficient clinical data to assess the likelihood of dRTA only one case fulfilled all the criteria; two cases fulfilled all but one of the criteria and dRTA was strongly suspected based on clinical progression and persistence of urine pH > 6 in the face of severe metabolic acidosis.

Of the three cases where concurrent IMHA and dRTA was suspected, two survived to discharge; one was still alive at the time of writing (13 months after discharge) and the other was euthanased 2 months after discharge following the development of multiple joint effusions and skin lesions suggestive of SLE.

Venous blood gas analysis and assessment of urine pH should be considered in all cases of IMHA to exclude the possibility of concurrent dRTA, particularly where persistent hypokalaemia is detected. Prospective evaluation of a larger cohort of IMHA cases is required to determine the actual incidence of concurrent dRTA.

Conflicts of interest: No conflicts of interest reported.

ESVNU-P-2

PREVALENCE OF PROTEINURIA IN CATS AFFECTED WITH CHRONIC KIDNEY DISEASE. P. Scarpa, E. Lazatera, T. Vitiello. UNIVERSITY OF MILAN, Milano, Italy

Persistent renal proteinuria is considered an early marker of chronic kidney disease (CKD) and it is listed among the initiation factors and progression factors according to KDOQI guidelines. Nevertheless, few data are available about the prevalence of proteinuria in cats affected with CKD, in which it is assumed that nephropathy is mainly characterized by tubulointerstitial damage.

The aim of this study is to determine the prevalence of proteinuria in cats affected with CKD and to evaluate the relations between urine protein to creatinine ratio (UPC) or IRIS substaging by proteinuria, towards purebred, sex, age, haematology, biochemistry and urinalysis. Wilcoxon test, linear regression and chi-square test were used for the statistical analysis.

Data from 251 cats were considered. Non-renal proteinuria was an exclusion criterion.

Proteinuric cats (UPC>0.4) were 15.4% in CKD cats, while 15.9% could be substage as borderline proteinuric (0.2)

In cats, proteinuria tends to increase with aging ($p < 0.0001$) and with worsening of the nephropathy ($p = 0.002$). Proteinuria was related to the anaemic state in CKD cats: UPC significantly increases with RBC count, Hb, Ht and MCH decreasing ($p < 0.0001$ and $p = 0.049$ respectively). Proteinuria tends to increase with WBC count ($p = 0.0001$) and neutrophils increasing ($p = 0.0001$), while tends to decrease with lymphocytes increasing ($p = 0.008$). Furthermore, UPC significantly increases in presence of an inflammatory serum protein electrophoretic pattern.

UPC tends to increase with phosphorus and ALP increasing ($p < 0.0001$ and $p = 0.0005$ respectively); while the role of phosphorus in CKD is well known, the increase of ALP is questionable: it has been hypothesized that higher ALP levels in CKD could be related to B-ALP increase due to bone remodelling in secondary renal hyperparathyroidism. Considering urine parameters, UPC increases when urinary specific gravity and pH decrease (probably related to worsening of CKD and development of a metabolic acidosis) and when glycosuria is present, regardless of the cause.

Furthermore, proteinuria increases in presence of RBC in urinary sediment and in samples where casts were observed, in particular when RBC casts (considered always pathological and indicative of glomerular damage) were present.

UPC values assessed in proteinuric cats and data analysis suggest the need of deepen the analytical variability of UPC and the opportunity to reconsider the intervals of substaging by proteinuria in cats.

Conflicts of interest: No conflicts of interest reported.

ESVNU-P-3

URINARY TRACT INFECTIONS IN DOGS AND CATS: URINE CULTURE VERSUS URINALYSIS. P. Scarpa, T. Vitiello, L. Riedo, M. Persico, P.A. Martino. UNIVERSITY OF MILAN, Milano, Italy

Urinary tract infections (UTIs) are a big challenge in clinical practice because of the variability of symptoms and laboratory findings.

The aim of this retrospective study was to evaluate: a) the relations between urine culture results and urinalysis parameters; b) the results of the antimicrobial susceptibility tests.

Urine samples were collected by cystocentesis from 252 dogs and 52 cats, whose diagnostic workup included a differential diagnosis of UTI: all samples underwent a complete urinalysis, UPC ratio assessment and urine culture. Infected vs sterile results were related to urine physical, chemical parameters and observations from urinary sediment analysis. Statistical analysis was performed using JMP 7.0 (SAS Institute Inc.). A p value < 0.05 was considered significant.

Urine culture resulted positive in 104 dogs (41%) and 18 cats (33%). The presence of UTI was significantly related to urine physical properties (color and turbidity), USG and leukocyturia: infections tended to be more frequent in urine samples characterized by a light yellow color, cloudy or sub-limpid aspect and low USG. Nevertheless, urine was limpid in 30% of infected samples, and a normal USG was found in 28.6% of dog's UTI but only in 4.8% of cats. Although leukocyturia tends to become higher in infected samples both in dogs and cats ($p < 0.0001$), in 23.1% of infected sediments WBC count was normal. Haematuria detected by dipstick was significantly related to UTI in dogs but not in cats, nevertheless the RBC count in sediment was not related to infection in both species: RBC count was normal in 30.4% of infected feline samples and in 36.4% of canine samples. No significant relation between presence or absence of UTI and albuminuria, bilirubinuria, glycosuria was detected, while UPC tends to become significantly higher in dogs. Although the chi-square test showed a significant relation between infection and the detection of bacteria in urinary sediment, a pseudobacteriuria was found in 20.5% of samples; furthermore bacteria weren't observed in 25.1% of infected samples (USG<1.013).

E. coli was isolated in the majority of samples (45.9%), compared to other species: *Staphylococcus* (13.6%), *Proteus* (10.2%) and *Streptococcus* (8.5%). The urinalysis pitfalls and the high antibiotic resistance verified towards the most widely used molecules (penicillins, cephalosporins, quinolones) strongly indicates the importance to perform antimicrobials susceptibility tests to avoid the risk of failure associated with the use or abuse of empiric therapies in UTIs.

Conflicts of interest: No conflicts of interest reported.

ESVNU-P-4

THE EVALUATION OF RENAL FUNCTION AFTER MITRAL VALVE REPAIR IN DOGS. K. Harada, Akiko Nishikawa, Masami Uechi. Japan, Tokyo, Japan

Azotemia in dogs with chronic heart failure may reflect impaired renal function not only because of inadequate renal perfusion, but also due to organic renal injury. Impaired renal function is observed in 50% of dogs with heart failure. Altered renal hemodynamics due to decreased cardiac output results in renal hypoperfusion, and resultant elevation of blood urea nitrogen and creatinine, defined as azotemia. Azotemia is a prognostic factor in dogs with mitral regurgitation, therefore, preservation and/or restoration of renal function is thought to improve prognosis. Medical treatment for heart failure, however, includes angiotensin converting enzyme inhibitors and loop diuretics, which has been shown to increase the risk of developing azotemia. We hypothesized that mitral valve repair surgery ameliorates renal function by improvement of systemic hemodynamics. The change in renal function in dogs with mitral regurgitation was assessed by evaluating time-dependent changes in glomerular filtration rate by inulin clearance before and after cardiac surgery. Eighteen dogs with severe mitral regurgitation with azotemia (plasma urea nitrogen level > 28 mg/dL, plasma creatinine level > 1.9 mg/dL) were included in this study. The glomerular filtration rate in all dogs were evaluated by determining inulin clearance before and 3 months after surgery. Serum atrial natriuretic peptide level, plasma NT-pro brain natriuretic peptide level, plasma urea nitrogen concentration, and plasma creatinine concentration were measured at each time point as well as during the initial staging of heart failure based on the International Small Animal Cardiac Health Council (ISACHC). Left atrial/aorta ratio by echocardiography and vertebral heart size by thoracic radiographs were also measured. Glomerular filtration rate significantly increased 3 months after surgery (40.0 ml/min/m² [25.6 - 123.0], 2.7 ml/min/kg [1.0 - 5.3]) compared to before surgery (38.4 ml/min/m² [12.6 - 50.3], 2.2 ml/min/kg [0.7 - 3.8])

($P < 0.05$). The ISACHC stage of heart failure was improved at 3 months after surgery compared to before surgery. In addition, serum atrial natriuretic peptide level, plasma NT-pro brain natriuretic peptide level, plasma urea nitrogen concentration, LA/Ao and VHS significantly decreased after surgery ($P < 0.05$). The use of diuretics decreased after mitral valve repair surgery and consequently, a decrease in plasma urea nitrogen and creatinine levels were observed. Therefore, this suggests that the main cause of azotemia in dogs with mitral regurgitation may be due to inadequate renal blood flow and exacerbation by the use of diuretics.

Conflicts of interest: No conflicts of interest reported.

ESVNU-P-5

VALIDATION OF A HIGH PERFORMANCE CAPILLARY ELECTROPHORESIS METHOD FOR MEASUREMENT OF GLOMERULAR FILTRATION RATE BY SERUM IOHEXOL CLEARANCE IN DOGS. L. Pelligand¹, S. Williams², J. Elliott¹. ¹Royal Veterinary College, Hatfield, United Kingdom, ²DeltaDot Ltd, London, United Kingdom

Glomerular filtration rate (GFR) is generally considered to be the gold standard measurement of kidney function. GFR can be calculated by measuring serum iohexol clearance using concentrations at 2,3 and 4 hours following a bolus injection.

For validation, serum samples were spiked at low (0.017 mg/mL), medium (0.27 mg/mL) and high (2.16 mg/mL) iohexol concentrations. They were analysed, along with standard calibration curves (8 concentrations ranging from 0.017 to 2.16 mg/mL), using deltaDOT's Label-Free high performance capillary electrophoresis (HPCE) system. Data were analysed using deltaDOT's General Separation Transform (GST). Clinical and spiked serum samples were also sent for analysis by mass spectrometry (MS) at a reference laboratory (14 samples for comparison). Concentrations obtained by HPCE and MS were compared in a Bland Altman plot. GFR for clinical samples was calculated from the measured iohexol concentrations using the method reported by Bexfield (2008).

A validated method was produced, with a lower limit of detection of 0.009 mg/mL and an lower limit of quantification of 0.017 mg/mL. The upper limit of quantification was 2.16 mg/mL. The standard curve had excellent linearity ($R^2 = 0.993$). Maximum inaccuracy was less than 11.5% of the true value, except at LLOQ, where it was within 24.2%. Average within day variability was less than 13.1% at all levels, while between day variability was less than 5.6%, except at LLOQ, where it was less than 15.4%. Agreement between the results obtained by measurement with HPCE and MS was good (bias 1.3%, lower and upper limits of agreement of -15.4 and 18.0%, respectively). Method specificity was confirmed by the absence of matrix effect in six serum specimen obtained from clinical dogs. Clinical samples were analysed and GFR reported with a 3 day turnaround time.

In conclusion, HPCE provides an accurate and precise method for measuring iohexol in canine serum.

Conflicts of interest: L Pelligand has the following information to disclose: In receipt of research grant / contract funding from Orion Ltd., Novartis Animal Health, Transpharmation Ltd, DeltaDot Ltd; Acted as a consultant for: Triveritas Ltd. and Novartis Animal Health; S Williams is an employee of the RVC and works in collaboration with deltaDot Ltd; J. Elliott has the following information to disclose: Consultancy: Pfizer Animal Health / Zoetis, CEVA Animal Health, Boehringer Ingelheim, Vetoquinol Ltd, Orion Ltd., Elanco Ltd, Idexx Ltd, Niche Generics Ltd. Triveristas Ltd., Virbac Ltd., Advisory board membership: International Renal Interest Society (supported by Novartis) European Emesis Council (sponsored by Pfizer Animal Health - now Zoetis) Cardiorenal Board - Vetoquinol Ltd. Idexx Renal Advisory Board Research Grants or contracts: Vetoquinol Ltd, Novartis Ltd, Pfizer Animal Health Ltd (now Zoetis), Royal Canin Ltd, Boehringer Ingelheim Ltd, Waltham Centre for Pet Nutrition, CEVA Animal Health Orion Ltd.

ESVONC-P-1

CYSTIC PANCREATIC NEOPLASIA IN CATS. C.M. Borschensky¹, K. Steiger², A. Staudacher³, M. Schlitter², I. Esposito¹, H. Aupperle¹. ¹Laboklin GmbH&Co. KG, Bad Kissingen, Germany, ²TU München, Institute of Pathology, München, Germany, ³Veterinary Clinic Dr. Staudacher, Aachen, Germany

Pancreatic neoplasms in the cat mostly exhibit a solid growth pattern and are diagnosed as carcinomas. In contrast, only few reports about cystic pancreatic lesions exist. Until now, only benign cystic pancreatic lesions are described in the literature. According to the histological pattern, they have been termed as cysts, (acinar) cystadenoma or pseudocysts. In man, cystic pancreatic neoplasms are classified according to the localisation (intra-/extraductal), growth pattern and differentiation (mucinous, (tubulo)papillary, serous, acinar).

The aim of this study was to characterise feline pancreatic neoplasms in more detail, based on the human classification system with a special view on cystic lesions.

Pancreatic masses sent to LABOKLIN from 19 domestic cats (7-14 years) were investigated routinely macroscopically and by histological methods (H.&E. stain).

The neoplasms showed a cystic (n = 8) or solid (n = 11) pattern. Cystic pancreatic tumors were up to 7 cm in diameter and were classified as benign variants in five and malignant variants in three cases. Based on the human classification system, they were classified as tubulopapillary (n = 2), acinar (n = 2) and mixed (n = 1) adenomas and mixed carcinomas (n = 3), respectively. Solid pancreatic nodules were diagnosed as carcinomas with a tubular (n = 5) or acinar (n = 6) differentiation pattern.

In summary, the gross structure (solid versus cystic) seems to be of prognostic relevance. In contrast to solid tumors, cystic pancreatic lesions in the cat behave benign in a higher percentage of cases, resulting in a better prognosis. Therefore, surgical excision of these cystic masses can be recommended. With respect to the human classification system, three different subtypes of cystic pancreatic neoplasms were detected in the cat that have not been described before in veterinary medicine: tubulopapillary, acinar and mixed. To best of our knowledge, this is the first report of cystic adenocarcinomas in feline pancreas.

Further corresponding clinical and histological investigations are needed for a better diagnostic (ultrasound, MRI) and prognostic characterisation of cystic lesions in feline pancreas.

Conflicts of interest: No conflicts of interest reported.

ESVONC-P-2

ROLE OF CYCLOOXYGENASE-2 IN PROGNOSIS OF CANINE MAST CELL TUMOURS. H. Gregorio, J. Prada, I. Pires, F.L. Queiroga. University of Trás-os-Montes and Alto Douro, Vila real, Portugal

The immunohistochemical detection of Cyclooxygenase-2 (Cox-2) expression in canine mast cell tumours was recently described by our team (Prada et al., 2012). However its prognostic value needs to be established. The aim of the present work was study the prognostic value of Cox-2 expression by investigating the relationship with several clinical and pathological variables including the Overall Survival (OS) time.

We included 57 dogs with mast cell tumours (18 grade I; 21 grade II and 18 grade III). Cox-2 immunohistochemical expression was carried out by a streptavidin-biotin method. For the Cox-2 immunoreactivity evaluation were considered the number of positive cells (Cox-2 extension), the intensity and the score of Cox-2. The following clinical and pathological features were considered: animal age, sex, tumour anatomical location, tumour size, skin ulceration, histological grade, histological safety margins and number of mitosis. Cox-2 expression was correlated with the clinical and pathological data and with the overall survival.

Cox-2 intensity was statistical significantly associated with skin ulceration ($p = 0,009$); histological grade ($p = 0,006$) and absence

of histological safety margins ($p = 0,043$), high mitotic number ($p = 0,024$) and with overall survival ($p = 0,02$). Both Cox-2 extension and Cox-2 immunohistochemical score present no statistical relationship with the variables considered neither with the overall survival.

Our results suggest that Cox-2 have an important role in dog mast cell tumours progression and could constitute a promising therapeutic target in this neoplasia. However, our study also demonstrated that in MCTs, is the Cox-2 intensity that has the prognostic value, not the number of Cox-2 positive cells (Cox-2 extension) and not the Cox-2 immunohistochemical score. Consequently, Cox-2 intensity should be elected for evaluating the Cox-2 positivity in MCTs immunohistochemical studies.

Conflicts of interest: Meril provided financial support for immunohistochemical analysis. The research centres has also received financial support from CECAV, CECA and CITAB.

ESVONC-P-3

RADIOTHERAPY IN THE TREATMENT OF 32 CANINE SUBCUTANEOUS SOFT TISSUE SARCOMAS. M. Kleiter¹, M. Moser², I. Fliçkinger², M. Pagitz², M. Willmann², A. Tichy³, B. Wolfesberger². ¹University of Veterinary Medicine Vienna, Vienna, Austria, ²Department for Companion Animals and Horses, Vienna, Austria, ³Department for Biomedical Sciences, Vienna, Austria

Canine soft tissue sarcomas represent approximately 7-15% of all skin tumors. Wide surgical excision is the treatment of choice for this tumor entity. Radiotherapy is recommended for patients with incomplete resections and adjuvant chemotherapy for high grade sarcomas. The aim of this study was to analyse the first patient cohort treated at the University of Veterinary Medicine Vienna since installation of a linear accelerator in 2006.

Dogs which were radiated for a subcutaneous sarcoma between 2006 and 2011 were included. Medical records were reviewed and patient characteristics, treatment protocols, adjuvant therapies and outcome were analysed. Follow-up information was obtained from medical records and by phone conversations with veterinarians or pet owners.

Thirty-two dogs were included into this study. Mean age was 9 years and mean body-weight was 32 kg. Male dogs were slightly overrepresented (59.4%). Curative intent radiotherapy was applied in 22 dogs and palliative intent in 10 dogs with a mean total dose of 51 and 28.4 Gray, respectively. In 17 dogs microscopic disease was radiated. Five dogs received liposomal doxorubicin concomitantly with radiotherapy, two received adjuvant doxorubicin and one intravesical cisplatin. Overall median survival time was 914 days with curative and 513 days with palliative treatment. Overall median survival time in dogs with macroscopic disease was 369 days and in patients with microscopic disease it was not reached.

Radiotherapy was generally accepted as new treatment modality by pet owners and referring veterinarians. Comparable to the literature, best outcome was achieved for dogs radiated with microscopic disease

Conflicts of interest: No conflicts of interest reported.

ESVONC-P-4

ONCEPT® VACCINATION IN 34 DOGS - THE SOUTH AFRICAN EXPERIENCE. J.L. Mclean, R. Lobetti. Bryanston Veterinary Hospital, Johannesburg, South Africa

Oncept®, is indicated for the treatment of stage II or III oral melanoma after local control with survival times significantly increased following vaccination. A similar improvement in survival times has also been reported with digit melanoma.

Medical records of dogs diagnosed with melanoma between March 2009 and December 2013 were retrospectively evaluated. Inclusion criteria were a histopathological diagnosis of melanoma, surgical excision of the tumour, and vaccination using the Oncept® vaccine. 34 dogs met the inclusion criteria.

23 dogs had stage II and III oral melanoma with a median age of 9.1 years (range 5-14). Currently 7 still alive with a median survival time of 25 months (range 2-40) and 16 dead - 13 with progressive disease and the other 3 from unrelated causes (one with gastric torsion, one with severe degenerative joint disease and one with osteosarcoma of the humerus). Median survival time in this group was 10 months (range 5-25). Sex distribution was 11 males and 12 females of various breeds - Dachshund (3), GSD (3), Spaniel (3), Pekinese (2), Rottweiler (2), Staffordshire terrier (2), Bouvier, Giant Schnauzer, Maltese, Irish setter, Kerry blue terrier, Golden retriever, Scottish terrier, and Great Dane (1 of each).

6 dogs had stage II digit melanoma with an equal sex distribution and a median age of 7.8 years (range 3-10). Currently 5 still alive and one dead, the latter following surgery for resection of a rib osteosarcoma. Median survival time of the 5 survivors was 23 months (range 5-40). Survival time of the dog that died was 12 months. Breeds were Golden retriever (2), Bouvier (1), Chow (1), Schnauzer (1) and Shar Pei (1).

5 dogs had stage II-IV melanoma at various other sites with a sex distribution of 3 females and 2 males with a median age of 10.4 years (range 9-11). All tumours were infiltrative and involved the inguinal area, ventral abdomen, axilla, forelimb and hindlimb. Survival times of the 3 survivors are 16, 38 and 39 months. Survival time of the 2 dogs that died was 2 and 3 months. Breeds were Dachshund (2), Staffordshire terrier (2) and Ridgeback (1).

Of the 34 dogs vaccinated, 15 are still alive and 19 dead. The median survival time of the dogs still alive is 25.5 months (range 5-40) versus 13.4 months (range 2-25) for the dogs that have died. None of the dogs showed any adverse effect to the vaccine.

Conflicts of interest: No conflicts of interest reported.

ISCAID-P-1

MORPHOLOGICAL RENAL LESIONS IN DOGS SEROPOSITIVE FOR CANINE VISCERAL LEISHMANIASIS. M.G. Monteiro Carvalho Mori Cunha, C. Lacerda Soares I, G. Bezerra de Moraes, F. Antônio Félix Júnior, I. Lira Borges, I. Neto Góes da Silva, D. Araújo Viana, J. serra azul Monteiro Evangelista. Universidade Estadual do Ceará, Fortaleza, Brazil

Canine Visceral Leishmaniasis (CVL) is a serious chronic disease that affects different animal species. Infected macrophages can cause injury in different organs, including the kidney. CVL is known as a common cause of glomerulonephritis. Thus, this study aimed to investigate and characterize the renal lesions in dogs seropositive for *Leishmania sp.* in Brazil. This project was approved by the animal ethics committee of UECE, Brazil. Twenty adult dogs seropositive for CVL from Center for Zoonosis Control were randomly selected for this experiment. CVL was diagnosed by immunofluorescence and ELISA. Urine and blood sampling and kidney harvesting were performed immediately after euthanasia. Urinalysis and serum creatinine levels were evaluated. Hematoxilin and eosin stained kidneys were assessed by a pathologist in a blind fashion. Dogs were classified into four stages according to the International renal interest society staging system for chronic kidney disease (CKD), based on serum creatinine concentrations. The most frequent histopathological changes observed was mild interstitial fibrosis in 55% (11/20) and glomerulosclerosis and chronic multifocal lymphoplasmacytic interstitial nephritis in 70% (14/20) of the dogs. Urinalysis showed the presence of proteinuria (70%), casts (55%), hematuria (30%) and bacteriuria (15%). Twelve dogs were classified in the stage 1 of CKD, five in stage 2, two in stage 3 and only one in stage 4. Regarding the histopathologic changes, 95% (19/20) had at least one glomerular lesion. Glomerulosclerosis (35%) and multifocal glomerulonephritis (15%), glomerular fibrosis (20%) and thickening of Bowman's capsule (15%) were also observed. One dog showed only tubular and interstitial lesions. An expressive renal impairment and signs of chronic injury such as fibrotic areas were also observed, even in the animals in stage 1 of CKD. The glomerular thickening observed in animals with CVL is due to deposition of proteinaceous material or antibodies. The histopathological changes observed in this study demonstrated a sig-

nificant renal involvement in dogs with CVL. This result indicates that the glomerulonephritis is a common sequelae related to leishmaniasis infection. Even dogs in stage I of CKD showed significant renal histopathological changes. Animals infected with *Leishmania sp.* may have severe renal damage and risk of progressive chronic kidney disease even when no increase of creatinine levels or proteinuria is detected.

Conflicts of interest: The authors received funding to pay the PhD scholarship of one student (Conselho Nacional de Pesquisa e Desenvolvimento - CNPq- Brazil) and received a research grant from Fundação Cearense de Apoio ao Desenvolvimento Científico e Tecnológico - FUNCAP.

ISCAID-P-2

PREVALENCE OF ANAPLASMA PHAGOCYTOPHILUM IN HEALTHY CANINE BLOOD DONORS. A. Chirek¹, C. Silaghi², K. Pfister², B. Kohn¹. ¹Germany, Clinic of Small Animals, Faculty of Veterinary Medicine, Freie Universi, Berlin, Germany, ²Institute of Comparative Tropical Medicine and Parasitology, LMU Munich, Munich, Germany

Anaplasma phagocytophilum, the causative agent of canine granulocytic anaplasmosis, is an obligatory intracellular bacterium transmitted by *Ixodes* ticks. Transmission via blood transfusion has rarely been described in human medicine and once in a dog. In the Berlin/Brandenburg area the seroprevalence rate in dogs was 43% regardless of health status.

The aim of this study was to evaluate PCR screening results for *A. phagocytophilum* in canine blood donors between 2006-2012 in order to estimate the risk of transfusion-transmitted infection.

917 EDTA blood samples from 517 dogs were submitted for *A. phagocytophilum* real-time PCR testing (targeting the *msp2*-gene). Altogether 158 dogs were tested up to 11 times. Clinical and laboratory data were examined before each donation. Statistical analysis was performed using SPSS 17.0.

The PCR test was positive for 21 of the 917 samples. None of the dogs tested PCR positive more than once. Positive results were most often detected in June (8), May (5), and July (3), but also in five other months. In three dogs a mild increased in rectal temperature ($\geq 39,0^{\circ}\text{C}$) was documented. Mild laboratory abnormalities were noted in 11 dogs: thrombocytopenia (3), leukocytosis (2), leukopenia (2), anemia (1) and hyperproteinemia (6/18); four dogs had more than one abnormality. There was no significant difference between the PCR negative and positive blood samples in regard to laboratory abnormalities.

Altogether, 2.3% of blood samples from healthy canine blood donors were PCR positive for *A. phagocytophilum*. Therefore, blood donors should be screened by PCR in endemic areas all year round.

Conflicts of interest: No conflicts of interest reported.

ISCAID-P-3

STUDY OF RISK FACTORS IN CATS WITH URTD, CONJUNCTIVITIS AND CHRONIC GINGIVOSTOMATITIS IN SPAIN. M. Fernandez Aragonés¹, E.G. Manzanilla¹, A. Lloret¹, M. León², J.C. Thibault³. ¹Fundació Hospital Clínic Veterinari, Bellaterra, Spain, ²Merial Laboratorios, Barcelona, Spain, ³Merial SAS, Lyon, France

A large multicentric study was conducted in Spain to evaluate risk factors for upper respiratory tract disease (URTD), conjunctivitis and chronic gingivostomatitis (CGS), as well as for the presence of four pathogens [feline herpesvirus-1 (FHV-1), feline calicivirus (FCV), *Chlamydomphila felis* and *Mycoplasma felis*].

The study population consisted of 358 cats, including 98 control cats recruited from 29 veterinary practices across the country. Among the 260 disease cats, 127 cats presented URTD, 149 cats had conjunctivitis and 153 cats suffered chronic gingivostomatitis, many of them presenting more than one clinical sign.

PCR for the above-mentioned pathogens was performed from pooled conjunctival and oropharyngeal swabs for each cat. A questionnaire regarding signalment (age, breed, sex, neuter status), environment (indoor, number of cats in household) and vaccination history was obtained. Data was analysed by multivariable logistic regression with alpha equal to < 0.05 .

The prevalence for the four pathogens has been previously reported in detail. Briefly, the prevalence among the four groups (including controls) ranged from 6 to 28% for FHV-1, 15-58% for FCV, 2-20% for *Chlamydomphila felis* and 20-46% for *Mycoplasma felis*.

In the univariate analysis, age, neutering status, being purebred, indoor keeping, number of cats in the household and body weight were variably associated with the different groups of disease and the presence of the pathogens. In the multivariable analysis, only the following factors remained significant. In the multivariable analysis, only the following factors remained significant: URTD was significantly associated with positive results for FHV-1, *Chlamydomphila felis* and *Mycoplasma felis* (in addition to being male and not castrated); conjunctivitis was significantly associated to positive results for FHV-1 and *Chlamydomphila felis* (in addition to being young, not castrated and purebred) and CGS was significantly associated to positive results for FCV (in addition to being young, male and purebred). Not being properly vaccinated was a significant risk factor only when all three groups were analyzed together. The number of cats in the household was an independent risk factor for detecting each of the pathogens studied. The age was also a significant factor in cats with FCV and *Chlamydomphila felis*, being older cats predisposed to FCV and younger cats predisposed to *Chlamydomphila felis*.

The present study describes important epidemiological data for cats presenting URTD, conjunctivitis and/or CGS, and emphasizes the complex interrelationships occurring among the different pathogens. Our results also support the role of FCV in cats with chronic gingivostomatitis.

Conflicts of interest: The study was funded and designed by Merial Laboratories.

ISCAID-P-4

HIGH FREQUENCY OF COLONIZATION BY METHICILLIN-RESISTANT STAPHYLOCOCCI IN HEALTHY HUMANS IN DAILY CONTACT WITH ANIMALS IN PORTUGAL. C. Pomba, A.C. Rodrigues, N. Couto. Faculty of Veterinary Medicine, Lisboa, Portugal

Reports of methicillin-resistant staphylococci (MRS) in animals have become more frequent in last years. Various studies have demonstrated the transmission of MRSA between animals and humans in daily contact with animals, however there is only limited data so far available on the transmission of methicillin-resistant coagulase-negative staphylococci between animals and humans. The objective of this study was to investigate the frequency of methicillin-resistant staphylococci (MRS) carriage in healthy veterinarians, veterinary nurses, veterinary assistants, veterinary students and farm workers from several veterinary hospitals, clinics and farms.

Nasal swabs were collected from 72 veterinarians (60 small animal veterinarians and 12 pig veterinarians), 8 veterinary nurses, 53 veterinary students, 4 veterinary assistants and 11 farm workers. MRS were screened on BrillianceTM MRSA2 agar (Oxoid) or ChromIDTM MRSA (bioMérieux). After 24-48 h of incubation at 37°C, suspected colonies on both media were subcultured onto blood agar plates. Species identification was obtained by species-specific PCR. Methicillin-resistance was confirmed by PCR amplification of the *mecA* gene. MRSA isolates were characterized by MLST.

Thirty-nine MRS were identified in 38 humans (23 veterinarians, 2 veterinary nurses, 7 veterinary students, 2 veterinary assistants and 5 farm workers). The MRS isolates were identified as *Staphylococcus aureus* (MRSA, n = 23), *S. epidermidis* (MRSE, n = 9), *S. pseudintermedius* (MRSP, n = 1), *S. haemolyticus* (MRSH, n = 1) and 5 MRS coagulase-negative staphylococci. The frequency of colonization by MRS was similar in both small animals and pigs veterinarians ($\pm 30\%$). One veterinary student was colonized simultaneously with an MRSE and an MRSH. The predominant ST in humans in contact with small animals was ST22 and in humans in contact with pigs was ST398.

In our study the frequency of colonization by MRSA was high, but the frequency of MRSE should not be underestimated. MRSA isolates in this work belonged mainly to the ST22 lineage which is the most frequent in small animals and humans in Europe. Humans in daily contact with animals can become colonized by MRS of animal origin and thus are important keys for infection control programs in Veterinary Hospitals and Farms.

Conflicts of interest: Dr Pomba currently receives research funding from the government and national programmes (Fundação para a Ciência e a Tecnologia). In the past, she has occasionally received research support or honoraria for lectures from pharmaceutical companies including Zoetis and Atral Cípan. She is vice-chair of the Antimicrobial Working Party (AWP) and member of the Antimicrobial Advice ad hoc Expert group (AMEG) of the European Medicines Agency (EMA).

ISCAID-P-5
INFECTION WITH HEMOPLASMA SPECIES IN 17 CATS WITH ANEMIA. C. Weingart¹, S. Tasker², B. Kohn¹. ¹Faculty of Veterinary Medicine, Freie Universität Berlin, Berlin, Germany, ²School of Veterinary Science & The Feline Centre, Langford Veterinary Science, Bristol, United Kingdom

The clinical course of natural infections with feline hemotropic mycoplasmas (hemoplasmas) has rarely been described. Hemoplasmosis ('*Candidatus Mycoplasma haemominutum*' [CMhm] n = 11, *Mycoplasma hemofelis* [Mhf] n = 3, species not determined n = 3) was diagnosed in 17 anemic cats (age 0.6-16 years, median 8; hematocrit 0.05-0.25 l/l, median 0.16) between 2005 and 2013. Seven of the cats (CMhm [5], Mhf [1], species not determined [1]) had concurrent disease, whilst 10 cats (CMhm [6], Mhf [2], species not determined [2]) did not. All 17 cats underwent antibiotic treatment (doxycycline or a fluoroquinolone); 12 cats received blood transfusions and/or Oxyglobin[®]. Three cats were euthanized within 11 days due to concurrent disease (FIV, pancreatitis/cholangitis) or financial constraints, one cat due to persistent anemia after 11 weeks. Four cats were lost to follow-up. The remaining 9 cats underwent follow-up for a period of 11-199 weeks (median 24). Hemoplasma PCR analysis was conducted 2-7 times on blood samples at variable time points from 6 of the 9 follow-up cats. The first negative PCR in 4 cases occurred after 3 (CMhm, during antibiotic treatment), 7 (CMhm, during antibiotic treatment), 10 (CMhm, during antibiotic treatment) and 23 (Mhf, after completion of antibiotics) weeks. One cat remained PCR positive (CMhm) at 4, 13, and 18 (all during antibiotic treatment) weeks, and another cat (CMhm) was PCR positive at 199 weeks. Reactivation of the hemoplasma species (documented by hemolysis and positive PCR) occurred in 2 cats (both CMhm) 1 and 3 times, respectively, up to 177 weeks

after initial presentation. Reactivation was suspected (no PCR testing available) in 2 additional cases (CMhm [1], Mhf [1]). Four of the 9 follow-up cats were euthanized after 14-180 weeks (median 24) due to concurrent disease (cardiomyopathy, immune-mediated thrombocytopenia, postoperative complications, diabetes mellitus). Infection with hemoplasmas is often chronic, can reactivate months later and is rarely the reason for euthanasia.

Conflicts of interest: No conflicts of interest reported.

ISCAID-P-6
CLINICAL APPLICATION OF PORCINE ANTI CDV ANTI-BODY SUBUNIT F(ab')₂ IN CANINE DISTEMPER DOGS. P.C. Liu¹, L.L. Chueh¹, C.A. Chen², C.M. Chen³, C.H. Yen³, M.H. Lee³, C.K. Chuang³, J.H. Lin³, C.F. Tu³, B.L. Su². ¹Graduate Institute of Veterinary Medicine, National Taiwan University, Taipei, Taiwan, ²Institute of Veterinary Clinical Sciences, National Taiwan University, Taipei, Taiwan, ³Animal Technology Laboratories, Agriculture Technology Research Institute, Miaoli, Taiwan

Canine distemper (CD) is a worldwide occurring infectious disease caused by a morbillivirus of the family *Paramyxoviridae*. CDV infection can result in a systemic infection. Dogs presented with neurologic signs revealed the terminal stage of the disease and usually failed to therapy. Additional passive immunotherapy is hypothesized to be beneficial in the early stage of CDV infection. Porcine anti-CDV antibody subunit F(ab')₂ [F(ab')₂] was produced by Animal Technology Laboratories, Agriculture Technology Research Institute. Eighteen CDV-naturally infected dogs showing respiratory signs but no neurological signs were treated with the combination of F(ab')₂ and supportive therapy (group 1). Group 2 included 33 dogs in a similar clinical signs (without neurological signs) that received only supportive therapy. The survival rate was 72.2% (13/18) in group 1 and 33.3% (11/33) in group 2, respectively, with a significant difference between the two groups (p < 0.05). The progressive rates of developing neurological signs during therapy of group1 and group2 were 44.4 and 63.6%, respectively. There was no significant difference between the two groups. The survival rates of dogs developing neurological signs during therapy were 50% (4/8) in group 1 and 4.8% (1/22) in group 2, respectively, with a significant difference between the two groups. In conclusion, additional administration of porcine anti-CDV antibody subunit F(ab')₂ before developing of neurological signs could decrease the mortality and furthermore reduce the rate of developing neurological signs.

Conflicts of interest: No conflicts of interest reported.