



Oral Research Communications of the 23rd ECVIM-CA Congress

Oral Research Communications of the 23rd ECVIM-CA Congress
Liverpool, United Kingdom, September 12-14, 2013

LIST OF ORAL RESEARCH COMMUNICATIONS

Number	Day	Time	Presenting Author	Abstract Title
ESVCN – European Society of Veterinary Clinical Nutrition				
ESVCN-O-1	Thursday 12 September	15.40- 15.55	Gant	CAN YOU TELL HOW FAT A CAT IS FROM PHOTOGRAPHS?
ESVNU – European Society of Veterinary Nephrology and Urology				
ESVNU-O-1	Friday 13 September	14.25- 14.40	Pelizzola	COMPARISON OF DIPSTICK PROTEINURIA AND URINARY PROTEIN TO CREATININE RATIO FOR EARLY DETECTION OF ALBUMINURIA: A RETROSPECTIVE STUDY IN 868 CANINE URINE SAMPLES
ESVNU-O-2	Friday 13 September	14.40- 14.55	Manczur	URINARY ENDOTHELIN CONCENTRATION IN VARIOUS CANINE KIDNEY DISORDERS
ESVNU-O-3	Friday 13 September	14.55- 15.10	Sánchez-Lara	FELINE CHRONIC KIDNEY DISEASE IS ASSOCIATED WITH UPREGULATION OF TRANSGLUTAMINASE, A CROSS-LINKING ENZYME
ESVNU-O-4	Friday 13 September	15.10- 15.25	Hill	URETHRAL STENTING FOR TREATMENT OF BENIGN URETHRAL OBSTRUCTION IN DOGS
ESVNU-O-5	Friday 13 September	15.25- 15.40	Bernardin	RISK FACTORS FOR URINARY TRACT INFECTION WITH MULTIDRUG-RESISTANT ESCHERICHIA COLI IN CATS
ESVNU-O-6	Friday 13 September	15.40- 15.55	Brovida	RENAL DISEASE IN 17 BOXER DOGS
ESVNU-O-7	Friday 13 September	16.30- 16.45	Ben Oz	RENAL HEMATURIA IN GOLDEN RETRIEVERS: A NEWLY RECOGNIZED DISORDER
ESVNU-O-8	Friday 13 September	16.45- 17.00	Marynissen	SERUM CYSTATIN C IN DOGS WITH DIABETES MELLITUS OR CUSHING'S SYNDROME
SCH – Society of Comparative Hepatology				
SCH-O-1	Thursday 12 September	14.25- 14.40	Watson	SKYE TERRIER HEPATITIS RE-APPRAISED: IS THIS A CONGENITAL DUCTAL PLATE ABNORMALITY?
SCH-O-2	Thursday 12 September	14.40- 14.55	Fieten	LONG-TERM MANAGEMENT OF COPPER ASSOCIATED HEPATITIS IN THE LABRADOR RETRIEVER
SCH-O-3	Thursday 12 September	14.55- 15.10	Lidbury	EVALUATION OF SERUM PROCOLLAGEN TYPE III N-TERMINAL PEPTIDE AS A POTENTIAL BIOMARKER FOR CANINE HEPATIC FIBROSIS.

SCH-O-4 Thursday 12 15.10- Gow
September 15.25
CHARACTERISATION OF CANINE ADIPOSE AND BONE MARROW-DERIVED MESENCHYMAL STEM CELLS AND HEPATOCYTE DIFFERENTIATION CAPACITY

SCH-O-5 Thursday 12 15.25- Gow
September 15.40
LOW DENSITY LIPOPROTEIN (LDL) AND ACETYLATED-LDL (AC-LDL) UPTAKE OF PRIMARY CANINE HEPATOCYTES AND CANINE MESENCHYMAL STEM CELLS (MSC).

VBPS – Veterinary Blood Pressure Society

ESVIM – European Society of Veterinary Internal Medicine

ESVIM-O-1 Saturday 14 14.25- Roels
September 14.40
IS THE CXC-CHEMOKINE CXCL8 INVOLVED IN THE BREED PREDISPOSITION OF WEST HIGHLAND WHITE TERRIER TO CANINE IDIOPATHIC PULMONARY FIBROSIS?

ESVIM-O-2 Saturday 14 14.40- Krafft
September 14.55
IMMUNOHISTOCHEMICAL ANALYSIS OF TRANSFORMING GROWTH FACTOR BETA 1 AND ITS SIGNALLING PATHWAYS IN CANINE IDIOPATHIC PULMONARY FIBROSIS

ESVIM-O-3 Saturday 14 14.55- Krafft
September 15.10
IS 5-HYDROXYTRYPTAMINE (SEROTONINE) INVOLVED IN THE PATHOGENESIS OF IDIOPATHIC PULMONARY FIBROSIS IN DOGS?

ESVIM-O-4 Saturday 14 15.10- Lilja-Maula
September 15.25
6-MINUTE WALK TEST IN WEST HIGHLAND WHITE TERRIERS WITH IDIOPATHIC PULMONARY FIBROSIS AND CLINICAL FEATURES DURING DISEASE PROGRESSION

ESVIM-O-5 Saturday 14 15.25- Laurila
September 15.40
SURVIVAL AND PROGNOSTIC FACTORS IN WEST HIGHLAND WHITE TERRIERS WITH IDIOPATHIC PULMONARY FIBROSIS

ESVIM-O-6 Saturday 14 15.40- Canonne
September 15.55
LONG TERM FOLLOW UP IN DOGS WITH EOSINOPHILIC BRONCHOPNEUMOPATHY TREATED WITH INHALED GLUCOCORTICOSTEROID THERAPY: PRELIMINARY RESULTS.

ESVIM-O-7 Thursday 12 10.00- Viitanen
September 10.15
C-REACTIVE PROTEIN IN CANINE LOWER AIRWAY DISEASES AND IN CARDIOGENIC PULMONARY EDEMA

ESVIM-O-8 Thursday 12 9.00- Biermann
September 9.15
EFFECTS OF LOW DOSE CLOPIDOGREL ON PLATELET FUNCTION IN HEALTHY DOGS

ESVIM-O-9 Thursday 12 9.15- Senzolo
September 9.30
EVALUATION OF CHANGES IN COAGULATION TIMES (PT AND APTT) AFTER INTRAVENOUSLY VITAMIN K ADMINISTRATION IN 73 DOGS INTOXICATED WITH ANTICOAGULANT RODENTICIDE

ESVIM-O-10 Thursday 12 9.30- Mason
September 9.45
TRAPPED NEUTROPHIL SYNDROME: PRESENTATION AND OUTCOME IN 3 U.K BRED BORDER COLLIES

ESVIM-O-11 Thursday 12 9.45- Girod
September 10.00
EFFECTS OF OMEPRAZOLE ON THE CANINE CEREBROSPINAL FLUID COMPOSITION

ISCAID - International Society for Companion Animal Infectious Diseases

ISCAID-O-1 Thursday 12 14.25- Kuzi
September 14.40
NOSOCOMIAL VIRULENT, MULTI-DRUG-RESISTANT ACINETOBACTER BAUMANNII INFECTION OUTBREAK IN A VETERINARY HOSPITAL

ISCAID-O-2 Thursday 12 10.15- Tebb
September 10.30
ABDOMINAL CRYPTOCOCCOSIS: A RESTROSPECTIVE EVALUATION OF 25 CASES FROM AUSTRALIA

ISCAID-O-3	Thursday 12 September	14.55- 15.10	Nivy	SERUM ACUTE PHASE PROTEINS CONCENTRATIONS IN DOGS WITH SPIROCERCOSIS AND THEIR ASSOCIATION WITH ESOPHAGEAL NEOPLASIA: A PROSPECTIVE OBSERVATIONAL STUDY
ISCAID-O-4	Thursday 12 September	15.10- 15.25	Llewellyn	PREVALENCE OF LEPTOSPIRA URINARY SHEDDING IN HEALTHY DOGS FROM SOUTHERN GERMANY
ISCAID-O-5	Thursday 12 September	15.25- 15.40	Mende	PREVALENCE OF ANTIBODIES AGAINST FELINE PANLEUKOPENIA VIRUS IN CLIENT-OWNED CATS IN SOUTHERN GERMANY
ISCAID-O-6	Thursday 12 September	15.40- 15.55	Grellet	RISK FACTORS OF WEANING DIARRHEA IN PUPPIES FROM FRENCH BREEDING KENNELS
ISCAID-O-7	Thursday 12 September	15.55- 16.10	Geisweid	EVALUATION OF CONJUNCTIVAL SWAB POLYMERASE CHAIN REACTION FOR DETECTION OF LEISHMANIA INFANTUM IN DOGS IN A NON-ENDEMIC AREA
ISCAID-O-8	Thursday 12 September	16.10- 16.25	Mende	EVALUATION OF AN IN-HOUSE DOT ENZYME-LINKED IMMUNOSORBENT ASSAY TO DETECT ANTIBODIES AGAINST FELINE PANLEUKOPENIA VIRUS
ISCAID-O-9	Thursday 12 September	16.25- 16.40	Leal	THE USE OF ORAL RECOMBINANT FELINE INTERFERON-OMEGA IN NATURALLY FELINE IMMUNODEFICIENCY VIRUS INFECTED CATS: NEW INSIGHTS INTO AN ALTERNATIVE IMMUNOMODULATION THERAPY
ISCAID-O-10	Thursday 12 September	14.40- 14.55	Knebl	EFFICACY OF LOCAL APPLICATION OF FELINE INTERFERON-OMEGA IN CATS WITH ACUTE VIRAL UPPER RESPIRATORY TRACT DISEASE

ESVC – European Society of Veterinary Cardiology

ESVC-O-1	Thursday 12 September	14.25- 14.40	Vollmar	ASSOCIATION OF LOW OR NORMAL WHOLE BLOOD TAURINE CONCENTRATION WITH OUTCOME IN IRISH WOLFHOUNDS WITHOUT EVIDENCE OF DCM INITIALLY
ESVC-O-2	Thursday 12 September	14.40- 14.55	Aupperle	PATHOLOGY OF DCM IN GREAT DANES
ESVC-O-22	Thursday 12 September	14.55- 15.10	Spalla	SPECKLE TRACKING ECHOCARDIOGRAPHY IN DOGS WITH PATENT DUCTUS ARTERIOSUS
ESVC-O-4	Thursday 12 September	15.10- 15.25	Santilli	24-HOUR AMBULATORY ELECTROCARDIOGRAPHIC FINDINGS IN ENGLISH BULLDOGS WITH MYOCARDIAL DISORDERS
ESVC-O-5	Thursday 12 September	15.25- 15.40	Pedro	INDICES OF MYOCARDIAL STRAIN AND STRAIN RATE BY TWO-DIMENSIONAL SPECKLE TRACKING ECHOCARDIOGRAPHY IN GREAT DANES
ESVC-O-6	Thursday 12 September	15.40- 15.55	Dirven	ECHOCARDIOGRAPHIC FINDINGS IN 246 ADULT CATS WITH HEART DISEASE IN GENERAL PRACTICE
ESVC-O-7	Thursday 12 September	15.55- 16.10	Creamer	VALIDATION OF SMARTPHONE APPLICATION SOFTWARE IN THE MEASUREMENT OF HEART RATE IN DOGS AND CATS
ESVC-O-8	Thursday 12 September	16.10- 16.25	Hanås	24 HOURS HOLTER MONITORING OF CATS WITH ASYMPTOMATIC HCM BEFORE AND AFTER TREATMENT WITH ATENOLOL

ESVC-O-9	Thursday 12 September	16.25- 16.40	Perego	RESPONSE OF SUPRAVENTRICULAR TACHYCARDIA TO MANUAL CARDIOVERSION (CHEST THUMP)
ESVC-O-10	Saturday 14 September	9.00- 9.15	Wiedemann	EVALUATION OF CARDIAC TROPONIN I FOLLOWING DUAL CHAMBER PACEMAKER IMPLANTATION IN DOGS WITH ATRIOVENTRICULAR BLOCK
ESVC-O-11	Saturday 14 September	9.15- 9.30	Scollan	MEXILETINE SERUM LEVELS WITH TWICE DAILY DOSING IN COMBINATION WITH SOTALOL IN HEALTHY DOGS
ESVC-O-12	Saturday 14 September	9.30- 9.45	Lee	PATHOLOGIC MANIFESTATIONS AND CLINICAL CORRELATES IN CANINE DEGENERATIVE MITRAL VALVE DISEASE AT SURGICAL BIOPSY
ESVC-O-13	Saturday 14 September	9.45- 10.00	Tidholm	DIAGNOSTIC VALUE OF ECHOCARDIOGRAPHIC VARIABLES TO IDENTIFY PULMONARY ARTERIAL HYPERTENSION IN DOGS WITH MYXOMATOUS MITRAL VALVE DISEASE
ESVC-O-14	Saturday 14 September	10.00- 10.15	Locatelli	LONGITUDINAL RIGHT VENTRICLE STRAIN AND STRAIN RATE BY TWO DIMENSIONAL FEATURE TRACKING ECHOCARDIOGRAPHY IN HEALTHY DOGS
ESVC-O-15	Saturday 14 September	10.15- 10.30	Gentile	ECHOCARDIOGRAPHIC ASSESSMENT OF THE CANINE RIGHT HEART: REFERENCE INTERVALS AND REPEATABILITY
ESVC-O-16	Saturday 14 September	11.20- 11.35	Buch	CLINICAL EVALUATION OF A SECOND-GENERATION CANINE NT-PROBNP ASSAY
ESVC-O-17	Saturday 14 September	11.35- 11.50	Santarelli	EFFECTS OF COMBINATION OF ACEPROMAZINE/BUTORPHANOL ON CONVENTIONAL ECHOCARDIOGRAPHIC MEASUREMENTS AND GLOBAL STRAIN IN HEALTHY DOGS
ESVC-O-18	Saturday 14 September	11.50- 12.05	Merveille	PULMONARY VEIN TO PULMONARY ARTERY RATIO IS AN ECHOCARDIOGRAPHIC INDEX OF LEFT CONGESTIVE HEART FAILURE IN DOGS WITH MYXOMATOUS MITRAL VALVE DISEASE
ESVC-O-19	Saturday 14 September	12.05- 12.20	Kiss	HEART SPECIFIC GENE EXPRESSION PROFILING IN PERIPHERAL BLOOD OF DOGS WITH CHRONIC HEART FAILURE
ESVC-O-20	Saturday 14 September	12.20- 12.35	Reimann	BIOPTERIN STATUS IS ASSOCIATED WITH DISEASE SEVERITY AND HUMAN CARDIOVASCULAR RISK FACTORS IN DOGS WITH MYXOMATOUS MITRAL VALVE DISEASE
ESVC-O-21	Saturday 14 September	12.35- 12.50	Drut	PREVALENCE OF PHYSIOLOGIC HEART MURMURS IN HEALTHY YOUNG ADULT DOGS

ESVONC – European Society of Veterinary Oncology

ESVONC-O-1	Saturday 14 September	10.00- 10.15	Voorhorst	BIOAVAILABILITY OF CYCLOPHOSPHAMIDE AND VINCRIStINE AFTER INTRAPERITONEAL ADMINISTRATION IN CATS
ESVONC-O-2	Saturday 14 September	10.15- 10.30	Leo	EVALUATION OF LOW-DOSE CYCLOPHOSPHAMIDE TOXICITY IN CATS WITH MALIGNANT NEOPLASIA
ESVONC-O-3	Saturday 14 September	11.20- 11.35	Zandvliet	THE USE OF PREDNISOLONE WITHIN A MULTIDRUG CYTOSTATIC PROTOCOL FOR THE TREATMENT OF CANINE LYMPHOMA DOES NOT AFFECT THERAPY RESULTS

ESVONC-O-4	Saturday 14 September	11.35- 11.50	Terragni	EXPRESSION OF EGFR, HER-2 AND KRAS GASTRIC EPITHELIAL TUMORS IN DOG: CANINE GASTRIC CANCER COULD BE A MODEL FOR MAN?
ESVONC-O-5	Saturday 14 September	11.50- 12.05	Treggiari	CANINE INFRATENTORIAL (CAUDAL FOSSA) TUMOURS TREATED WITH FRACTIONATED RADIATION THERAPY: FIVE CASES
ESVONC-O-6	Saturday 14 September	12.05- 12.20	Kleiter	ALTERNATIVE LENGTHENING OF TELOMERES WAS DETECTED AS A TELOMERE MAINTENANCE MECHANISM IN CANINE SARCOMA
ESVONC-O-7	Saturday 14 September	12.20- 12.35	Killick	IDENTIFICATION AND GENETIC CHARACTERIZATION OF THE CANINE MAGE-B AND MAGE-C SUB-FAMILIES OF CANCER TESTIS ANTIGENS
ESVONC-O-8	Saturday 14 September	12.35- 12.50	Grant	MULTI-INSTITUTIONAL RETROSPECTIVE EVALUATION OF THE CLINICAL RESPONSE AND TOXICITY ASSOCIATED WITH THE USE OF MASITINIB FOR THE TREATMENT OF MAST CELL TUMOURS IN 34 DOGS

ESCG – European Society of Comparative Gastroenterology

ESCG-O-1	Friday 13 September	14.25- 14.40	Minamoto	EVALUATION OF SERUM METABOLITE CONCENTRATIONS AND FECAL MICROBIOTA IN DOGS WITH IDIOPATHIC INFLAMMATORY BOWEL DISEASE BEFORE AND AFTER TREATMENT
ESCG-O-2	Friday 13 September	14.40- 14.55	Grützner	SERUM HOMOCYSTEINE CONCENTRATIONS IN HYPOCOBALAMINEMIC AND HYPOFOLATEMIC GREYHOUNDS.
ESCG-O-3	Friday 13 September	14.55- 15.10	Unterer	PREVALENCE OF CLOSTRIDIUM PERFRINGENS ENTEROTOXIN AND CLOSTRIDIUM DIFFICILE TOXIN A/B IN DOGS WITH IDIOPATHIC ACUTE HAEMORRHAGIC DIARRHOEA SYNDROME
ESCG-O-4	Friday 13 September	15.10- 15.25	Schmitz	A PILOT STUDY ON THE EFFECT OF PROBIOTIC TREATMENT ON GENE EXPRESSION IN INTESTINAL BIOPSIES FROM DOGS WITH FOOD-RESPONSIVE CHRONIC ENTEROPATHY.
ESCG-O-5	Friday 13 September	15.25- 15.40	Luckschander	THE ROLE OF SIGNAL TRANSDUCER AND ACTIVATION OF TRANSCRIPTION (STAT) 3 IN DOGS WITH CHRONIC ENTEROPATHIES
ESCG-O-6	Friday 13 September	15.40- 15.55	Slovak	DEVELOPMENT AND VALIDATION OF A SIMPLIFIED ENDOSCOPIC ACTIVITY SCORE FOR CANINE CHRONIC ENTEROPATHIES
ESCG-O-7	Friday 13 September	16.30- 16.45	Trehy	SERUM PANCREAS-SPECIFIC LIPASE CONCENTRATIONS IN DOGS WITH UPPER GASTROINTESTINAL FOREIGN BODIES
ESCG-O-8	Friday 13 September	16.45- 17.00	Mortier	IDIOPATHIC ACUTE HEMORRHAGIC DIARRHOEA SYNDROME IN DOGS: 108 CASES
ESCG-O-9	Friday 13 September	17.00- 17.15	Williams	PROTEIN-LOSING ENTEROPATHY COMMONLY CO-EXISTS WITH HIGH FECAL FAT OUTPUT IN GERIATRIC CATS WITH IDIOPATHIC MALABSORPTION AND PERSISTS FOLLOWING CORRECTION OF SUBNORMAL SERUM COBALAMIN CONCENTRATION.

ESVE – European Society of Veterinary Endocrinology

ESVE-O-1	Friday 13 September	9.00- 9.15	Schäfer	EVALUATION OF INSULIN-LIKE GROWTH FACTOR 1 (IGF-1), TOTAL THYROXINE (TT4), FELINE PANCREATIC LIPASE IMMUNOREACTIVITY (FPLI) AND URINARY CORTICOID CREATININE RATIO (UCCR) IN CATS WITH DIABETES MELLITUS IN SWITZERLAND AND THE NETHERLANDS
ESVE-O-2	Friday 13 September	9.15- 9.30	Paepe	ROUTINE KIDNEY PARAMETERS, GLOMERULAR FILTRATION RATE AND URINARY CYSTATIN C IN CATS WITH DIABETES MELLITUS, CATS WITH CHRONIC KIDNEY DISEASE AND HEALTHY CATS
ESVE-O-3	Friday 13 September	9.30- 9.45	Catchpole	INVESTIGATION OF BREED DIFFERENCES AND DLA GENETIC INFLUENCE ON DEVELOPMENT OF ANTI-INSULIN ANTIBODIES IN DIABETIC DOGS
ESVE-O-4	Friday 13 September	9.45- 10.00	van den Berg	SF-1 EXPRESSION IN CANINE CORTISOL-SECRETING ADRENOCORTICAL TUMORS
ESVE-O-5	Friday 13 September	10.00- 10.15	Kool	ACTIVATING MUTATIONS OF GNAS IN CANINE CORTISOL-SECRETING ADRENOCORTICAL TUMORS
ESVE-O-6	Friday 13 September	10.15- 10.30	Cook	INCIDENTAL ADRENAL LESIONS: ULTRASONOGRAPHIC FINDINGS IN 3748 DOGS (2007-2010)
ESVE-O-7	Friday 13 September	11.20- 11.35	van der Kooij	MANAGEMENT OF HYPERTHYROIDISM IN CLIENT-OWNED CATS WITH AN IODINE-RESTRICTED FOOD
ESVE-O-8	Friday 13 September	11.35- 11.50	Vagney	SURVIVAL TIMES FOR CATS WITH HYPERTHYROIDISM TREATED WITH A FIXED LOW-DOSE OF IODINE 131
ESVE-O-9	Friday 13 September	11.50- 12.05	Corradini	PROGNOSTIC FACTORS IN DOGS WITH NEWLY DIAGNOSED PITUITARY DEPENDENT HYPERCORTISOLISM
ESVE-O-10	Friday 13 September	12.05- 12.20	Wehner	ASSOCIATION BETWEEN ACTH STIMULATION TEST, CLINICAL SIGNS, AND LABORATORY PARAMETERS IN DOGS WITH HYPERADRENOCORTICISM TREATED WITH TRILOSTANE
ESVE-O-11	Friday 13 September	12.20- 12.35	Rick	THE MEASUREMENT OF 21-HYDROXYLASE ANTIBODIES IN DOGS VIA ENZYME-LINKED IMMUNOSORBENT ASSAY
ESVE-O-12	Friday 13 September	12.35- 12.50	Roberts	STABILISATION OF DOGS WITH PRIMARY HYPOADRENOCORTICISM, COMPARING ONCE DAILY VERSUS TWICE DAILY ORAL DOSING OF FLUDROCORTISONE ACETATE

ESVCP – European Society of Veterinary Clinical Pathology

ESVCP-O-1	Saturday 14 September	14.25- 14.40	Shibly	PERFORMANCE AND VALUE OF DIRECT COOMBS' TESTING IN CATS WITH POTENTIAL IMHA IN A CLINICAL SETTING
ESVCP-O-2	Saturday 14 September	14.40- 14.55	Zoia	HAEMOSTATIC FINDINGS OF ASCITIC FLUID: A CROSS-SECTIONAL STUDY IN 70 DOGS
ESVCP-O-3	Saturday 14 September	14.55- 15.10	Zoia	ASSOCIATION BETWEEN ASCITES AND PRIMARY HYPER-FIBRINOGENOLYSIS: A CASE CONTROL STUDY IN 210 DOGS
ESVCP-O-4	Saturday 14 September	15.10- 15.25	Zoia	ANTITHROMBIN CONCENTRATION IN DOGS WITH ASCITES: A CASE CONTROL STUDY IN 105 DOGS
ESVCP-O-5	Saturday 14 September	15.25- 15.40	Zoia	UTILITY OF LIGHT'S CRITERIA IN CLASSIFYING PLEURAL EFFUSION IN CATS: A COHORT STUDY IN 19 CATS

ESVCP-O-6	Saturday 14 September	15.40- 15.55	Trehy	DOES HYPERCOBALAMINAEMIA HAVE ANY CLINICAL SIGNIFICANCE IN CATS?
ESVCP-O-7	Saturday 14 September	16.30- 16.45	Amores Fuster	CHARACTERISTICS OF DIAGNOSTIC AND NON DIAGNOSTIC LYMPH NODE CYTOLOGY SAMPLES IN A LARGE FIRST OPINION POPULATION OF DOGS AND CATS
ESVCP-O-8	Saturday 14 September	16.45- 17.00	Murtagh	COMPARISON OF THE NON-VALIDATED IN HOUSE METHOD OF URINE SEDIMENT PREPARATION WITH THE REFERENCE LABORATORY METHOD

ESVCN-O-1

CAN YOU TELL HOW FAT A CAT IS FROM PHOTOGRAPHS? P. Gant¹, S.L. Holden¹, V. Biourge², P.J. Morris³, A.J. German¹. ¹University of Liverpool, WIRRAL, United Kingdom., ²Royal Canin Research Center, AIMARGUES, France., ³WALTHAM Centre for Pet Nutrition, WALTHAM-ON-THE-WOLDS, United Kingdom.

Most veterinarians use body condition scoring to estimate adiposity in cats, and this correlates well with body fat mass measured by dual-energy X-ray absorptiometry (DEXA). However, the technique is operator-dependent, with scores from lay people being least reliable. If body condition could reliably be estimated from photographs, it might be feasible to develop a remote screening tool (i.e. internet web page or smart phone application), whereby photographs are uploaded and reviewed by experts. However, the feasibility of using photographs to determine body composition has never been assessed in cats.

Photographs from overweight and obese cats referred to the Royal Canin Weight Management Clinic, University of Liverpool UK, were reviewed. Images, taken before and after weight loss, were used enabling cats in ideal condition (BCS 3/5), overweight (BCS 4/5) and obese (BCS 5/5) to be examined. Eleven observers, with a range of experience examined the photographs, and estimated body condition (iBCS) using two methods: (1) a standardised assessment of paired dorsal and lateral images, taken against a grid using standardised positioning (iBCS_{dl}); and (2) a subjective estimate of body condition from photographs with no standardised positioning (iBCS_{sub}). Results were compared with body fat percentage measured by DEXA and actual BCS (assessed by an experienced operator).

Moderate, significant, positive associations were seen between body fat percentage and both methods, albeit with marked differences amongst operators (iBCS_{dl} median Kendall's tau 0.54, 0.28-0.72; iBCS_{sub} median Kendall's tau 0.47, -0.16 to 0.64). Overall, there was fair-to-moderate agreement with actual BCS for both methods BCS (iBCS_{dl} median kappa 0.41, 0.19-0.62, $P < 0.001$; iBCS_{sub} median kappa 0.47, -0.04-0.63). Operators scored a similar percentage of cats correctly with both methods (iBCS_{dl} 57%, 39-70%; iBCS_{sub} median 58%, 38-78%, $P = 0.51$). Age, sex, breed, coat length, coat colour, and operator experience had no effect on the performance of either method ($P > 0.10$ for all). However, body fat percentage was positively associated with the number correctly scored using the iBCS_{sub} method (Kendall's tau 0.353, $P = 0.023$), but not using iBCS_{dl} (Kendall's tau 0.172, $P = 0.27$).

An approximate estimate of body composition can be obtained from photographs in cats, although wide operator variability exists, which is marginally less when using standardised images and assessment.

Conflicts of interest: No conflicts of interest reported.

ESVNU-O-1

COMPARISON OF DIPSTICK PROTEINURIA AND URINARY PROTEIN TO CREATININE RATIO FOR EARLY DETECTION OF ALBUMINURIA: A RETROSPECTIVE STUDY IN 868 CANINE URINE SAMPLES. M. Pelizzola, M. Gruarin, L. Conti, R. Troia, M. Giunti, F. Dondi. Alma Mater Studiorum - University of Bologna, OZZANO DELL'EMILIA (BO), Italy

Proteinuria is considered a marker of kidney damage in dogs and urine dipstick test and urinary protein to creatinine ratio (UPC) are commonly used to diagnose and monitor proteinuric patients in the clinical practice. In humans, the urinary albumin to creatinine ratio (UAC) represents the *gold standard* method for the early renal disease detection. Moreover, albuminuria is associated to chronic kidney disease (CKD) progression and mortality.

The aim of the study was to evaluate the ability of a urine dipstick test and UPC to early recognize albuminuric dogs. 868 canine urine samples without active sediment and macroscopic hematuria were collected from January 2002 to December 2012 in our Teaching Hospital. Urinalysis, UPC and UAC were available for 550 samples, while UPC and UAC were performed in 868 specimens. Urine samples were dipstick tested, using an

automated reader. Considering UAC (immunoturbidimetric assay) as the reference method, the diagnostic accuracy of dipstick test and UPC (pyrogallol red- molybdate) was evaluated. UAC reference interval (0-0.024) was determined from 60 healthy dogs. Data collected were categorized using different cut-offs for urine specific gravity (1012 or 1030), dipstick proteinuria (30 or 100 mg/dl), UPC (0.2) and UAC (0.024). Diagnostic agreement (Cohen's *k* coefficient) and Spearman's correlation between dipstick, UPC and UAC were evaluated. The diagnostic accuracy was estimated with the area under the ROC curve (AUC) analysis. Significance level was set at $p < 0.05$.

Independently of urine concentration, diagnostic agreement between dipstick and UPC or UAC was substantial ($k = 0.62$ and $k = 0.61$, respectively; $p < 0.001$) with positive dipstick results ≥ 30 mg/dl, and was fair ($k = 0.27$ and $k = 0.26$, respectively; $p < 0.001$) with positive dipstick results ≥ 100 mg/dl. Diagnostic accuracy of dipstick compared to UPC and UAC was very good (AUC 0.84 and 0.84, respectively; $p < 0.001$) and negative dipstick results presented 100% sensitivity. UPC and UAC were highly correlated ($r = 0.90$; $p < 0.001$). Comparing UPC with UAC, diagnostic accuracy was excellent (AUC 0.94; $p < 0.001$), with maximum sensitivity and specificity for UPC ≥ 0.3 .

In conclusion, for any urine density positive dipstick results (≥ 30 mg/dl) probably detect albuminuria outside the reference interval, and UPC or UAC should be assessed. In contrast, samples with negative dipstick results are unlikely to be proteinuric or albuminuric. Furthermore, UPC could be used to detect albuminuric canine patients with an early kidney damage using thresholds lower than 0.5. Further studies are required to clarify the usefulness of UAC in the veterinary clinical practice.

Conflicts of interest: No conflicts of interest reported.

ESVNU-O-2

URINARY ENDOTHELIN CONCENTRATION IN VARIOUS CANINE KIDNEY DISORDERS. F. Manczur, N. Kubik, M. Kulcsár. Faculty of Veterinary Science, Szent István University, BUDAPEST, Hungary

Endothelins (ETs) are 21 amino acid vasoactive peptides with endocrine, paracrine and autocrine effects. In the kidney, endogenous ET controls water and sodium excretion and acid-base balance, and maintains normal renal cell proliferation and tonic vasoconstriction under physiological conditions.

Increased ET synthesis causes intrarenal vasoconstriction, mesangial cell proliferation, hypertrophy, contraction and extracellular matrix accumulation. These effects induce glomerular damage, and lead to renal inflammatory processes and fibrosis. Since ET causes vasoconstriction, sodium and water retention, it has the potential to raise blood pressure in kidney patients. Many laboratory animal studies and observations on human clinical patients have proved the pathologic roles of ET in hypertension and various renal disorders. Almost 100% of ET detected in urine is originates from the kidney itself (mostly from medulla). Accordingly, the assessment of urinary ET excretion but not of circulating ET concentration has been strongly suggested as a possible marker of early renal damage.

The aim of our research was measure the urinary ET concentration in spontaneous acute and chronic renal diseases of the dog.

Forty three urine samples were obtained by ultrasound guided cystocentesis from 34 dogs during their diagnostic work-up. Seven animals were healthy, while the other 27 suffered from various renal diseases or had hypertension (both renal and extrarenal origin). The measurements were performed by a human commercial RIA-kit according to the manufacturer's protocol. Urinary ET concentrations were expressed after normalization to the urinary creatinine level.

In this study we found the strongest association between urinary ET and sodium concentrations ($R^2 = 0.66$), and fractional excretion of sodium ($R^2 = 0.67$). There was weaker association between the degree of proteinuria and the urinary ET level ($R^2 = 0.3$), however proteinuric dogs had significantly higher urinary ET to creatinine ratio than non-proteinuric animals. Azotaemic dogs also had significantly higher urinary ET concentrations than non-azotaemic ones, but there was no association between the urinary ET expression and the degree of azotaemia.

No association has been found between the urinary ET level and systemic blood pressure.

Our results support previous data about the role of renal ET synthesis in the sodium handling of the kidneys. The found associations between proteinuria, renal diseases and urinary ET excretion should be confirmed on larger study populations.

Conflicts of interest: No conflicts of interest reported.

ESVNU-O-3

FELINE CHRONIC KIDNEY DISEASE IS ASSOCIATED WITH UPREGULATION OF TRANSGLUTAMINASE, A CROSS-LINKING ENZYME. A.C. Sánchez-Lara¹, J. Elliott², H.M. Syme³, J.L. Haylor¹. ¹Academic Nephrology Unit, University of Sheffield, SHEFFIELD, United Kingdom, ²Department of Veterinary Basic Science, Royal Veterinary College, LONDON, United Kingdom, ³Department of Veterinary Clinical Sciences, Royal Veterinary College, LONDON, United Kingdom

Chronic kidney disease (CKD) is a major cause of morbidity and mortality in the cat. Transglutaminase 2 (TG2) is a calcium-dependant enzyme proposed to mediate tubulointerstitial fibrosis by irreversibly cross-linking collagen fibrils.

Post-mortem kidney tissue was analysed from cats with either CKD, based on plasma creatinine concentration $>177 \mu\text{m/L}$ ($n = 10$), or without renal azotaemia ($n = 5$). CKD cats were staged according to the International Renal Interest Society, ranging from stage 2 (sub-stage b) to 4. Kidneys were assessed for extracellular matrix protein deposition by Masson's trichrome staining and collagen immunofluorescence. Total kidney TG enzyme activity and total TG2 protein were measured in tissue homogenates by putrescine incorporation and western blotting respectively. TG2 was quantified from a 75 kDa band on western blotting, detected using a polyclonal rabbit TG2 antibody. Extracellular TG enzyme activity and extracellular TG2 protein were determined *in situ* by fluorescent labelled streptavidin (TG substrate) and immunofluorescence, respectively, using cryostat sections. The cross-sectional data were analyzed by unpaired, two-tailed, t-test with Welch's correction. To evaluate group correlations, r^2 was calculated by linear regression analysis. The level of statistical significance was defined as $P < 0.05$.

Elevated plasma creatinine, urea and phosphate concentrations were associated with tubulointerstitial fibrosis but not glomerular fibrosis. Homogenate studies showed approximately 3-fold greater total TG enzyme activity (37.2 ± 8.1 vs. 10.9 ± 2.3 nmol putrescine per hour at 37°C , $P < 0.02$) and total TG2 protein (60300 ± 14870 vs. 21270 ± 4046 OD $\text{mm}^2/\mu\text{g}$ protein, $P < 0.03$) in the azotemic group when compared to the non-azotaemic group. Fluorescent studies performed *in situ* confirmed a 3-fold increase in extracellular TG enzyme activity (30.1 ± 3.1 vs. 7.9 ± 2.1 area%, $P < 0.0001$) and extracellular TG2 protein (26.4 ± 3.3 vs. 9.7 ± 2.7 area%, $P < 0.005$) in cats with azotaemic CKD. Tubulointerstitial TG2 showed highly significant positive linear correlations with plasma concentrations of creatinine ($r^2 = 0.75$, $P < 0.0001$), urea ($r^2 = 0.81$, $P < 0.0001$) and phosphate ($r^2 = 0.76$, $P < 0.0001$). Significant positive linear correlations could also be demonstrated with Masson's trichrome staining ($r^2 = 0.39$, $P < 0.02$) and abundance of type I collagen ($r^2 = 0.38$, $P < 0.02$).

In cats with azotaemic CKD, both filtration failure and tubulointerstitial fibrosis were associated with the upregulation of TG2, a cross-linking enzyme and the major TG isoform in the kidney. TG2 may provide a new therapeutic target for drug design to stop or slow the progression of feline CKD.

Conflicts of interest: No conflicts of interest reported.

ESVNU-O-4

URETHRAL STENTING FOR TREATMENT OF BENIGN URETHRAL OBSTRUCTION IN DOGS. T.L. Hill¹, A. Berent², C. Weisse². ¹The Royal (Dick) School of Veterinary Studies, EDINBURGH, United Kingdom, ²The Animal Medical Center, NEW YORK CITY, United States of America

Urethral obstructions from benign disease are uncommonly reported in dogs but do result in significant morbidity and

mortality due to challenges with conventional therapies. Treatment of malignant urethral obstructions with intraluminal urethral stents is reported to successfully relieve obstructions.

This retrospective study investigated urethral stent placement in dogs with benign urethral obstructions treated in the authors' practice between 2007 and 2012. Data collected included signalment, cause of benign obstruction, procedure time, size and type of stent, complications, and short- and long-term outcome.

Eleven dogs were included for analysis. Intraluminal urethral stent(s) relieved the obstructions in all dogs. Three dogs had two stents placed in separate procedures due to incomplete patency after treatment or tissue ingrowth through the stent. There were no other perioperative complications in any dog. Four dogs had mild incontinence following stent placement and one dog had moderate to severe incontinence. There was no significant association between sex, duration of clinical signs, length of the stent, and diameter of the stent and incontinence. One dog developed mild stranguria after stent placement. Ten dogs had an excellent outcome > 3 months after treatment. One dog that developed lymphoma was euthanized 4 months after treatment. One dog has chronic urinary tract infections that were present prior to stent placement.

Urethral stents appear to be effective treatments for benign urinary obstructions. Moderate to severe incontinence developed in 1/11 dogs. Stents relieved obstructions in all dogs with good to excellent long-term outcome in 10/11 dogs.

Conflicts of interest: No conflicts of interest reported.

ESVNU-O-5

RISK FACTORS FOR URINARY TRACT INFECTION WITH MULTIDRUG-RESISTANT ESCHERICHIA COLI IN CATS. F. Bernardin¹, J.H. Hernandez¹, D. Bota¹, M.F. Farbos¹, G. Ragetly¹, C.M. Médaille². ¹CHV Frégis, ARCUEIL, France, ²Laboratoire Vebio, ARCUEIL, France

The emergence of multiple drug-resistant (MDR) bacteria is a growing public health problem. In non-hospitalised humans, a certain number of factors that promote the onset of infection with MDR pathogens have been identified: the age of the patient, hospitalisation within the past year, and the administration of antibiotics within the past year. In dogs, it has been demonstrated that the proportion of *Escherichia coli* isolated from the rectum and resistant to at least one antibiotic increases after more than 3 days hospitalisation in a university intensive care unit. The objective of this retrospective study was to identify risk factors associated with MDR *E. coli* infection of the urinary tract in cats. All cats presenting a *E. coli* urinary infection between March 2010 and December 2012 at the Centre Hospitalier Vétérinaire Frégis (France) were divided into two groups: MDR group and non-MDR group. Isolates that were resistant to ≥ 3 antimicrobial classes were considered to be MDR. The effects of different variables on the occurrence of a MDR *E. coli* were evaluated: age, sex, additional diseases, number of antibiotics and number of days of hospitalisation. Fifty-two cats were identified (42 MDR and 10 non-MDR). The number of antibiotic families used within the last 3 months was associated with an increased risk of MDR *E. coli* urinary infection ($p: 0.007$). A trend between the number of days of hospitalisation within the last 3 months and an increased risk of MDR *E. coli* urinary infection was also found ($p: 0.090$). This study provides evidence that systematic urinary culture with antibiotic sensitivity testing should be recommended when treating urinary tract infection if antibiotics have been prescribed within the past 3 months. Moreover, the selection of MDR bacteria through antibiotic use should be considered as a potential risk associated with treatment.

Conflicts of interest: No conflicts of interest reported.

ESVNU-O-6

RENAL DISEASE IN 17 BOXER DOGS. C. Brovida¹, E.A. Galli¹, L. Aresu², R. Heiene³. ¹ANUBI Ospedale per Animali da Compagnia, MONCALIERI, Italy, ²Department of Comparative Biomedicine and Food Science, University of Padua, PADUA, Italy, ³NCVS, OSLO, Norway

Kidney diseases historically termed “juvenile” or “renal dysplasia” have been found in several dogs breeds. In Boxers, renal lesions, termed juvenile nephropathy, have been described, however the literature on this topic is limited and different theories on the pathogenesis are proposed. In one study, data from 37 Boxers younger than five years were collected retrospectively. The dogs showed azotaemia, inappropriately low urine concentrating ability, and ultrasound or radiographic evidence of abnormal kidneys. Renal histopathological findings included pericapsular and interstitial fibrosis, inflammatory cell infiltration, dilated tubules, sclerotic glomeruli and dystrophic calcification (Chandler, 2007). Another study included seven young Boxers and the authors concluded that the end-stage kidney disease (ESKD) likely was the result of a chronic reflux nephropathy causing segmental hypoplasia (Ask-Upmark kidney) (Kolbjørnsen, 2008). The aims of the present study were twofold: to describe the light microscopy lesions in a population of Boxers, and to evaluate the clinical findings and compare them to published data in Boxers and in other dogs with non-specific chronic kidney disease (CKD). We retrospectively evaluated kidney biopsies and clinical data from seventeen Boxers enrolled between 2005-2012 with CKD IRIS stage 1-4. Ages ranged from 1 to 6 years. The dogs were evaluated by use of biochemistry and CBC, urinalysis, urine culture, blood pressure and renal ultrasound. Clinical data were available for an extended period. Eight dogs had renal lesions associated with juvenile nephropathy based on the presence of immature (fetal) glomeruli. The histopathological findings included increased of mesangial matrix and cells, glomerulosclerosis, interstitial fibrosis and tubular atrophy. No immune deposits were observed excluding immune mediated origin of the disease. The most striking features were high number of cystic glomerular atrophy and the alteration in the morphology of the glomeruli characterized by severe podocyte hypertrophy and splitting of the Bowman's capsule with multiple synechiae. Three dogs had Leishmania, one dog had renal lymphoma, and one dog had chronic urinary tract infection. Four dogs had nonspecific ESKD. Clinicopathological findings were characteristic of progressive CKD, with PU/PD, weight loss, anorexia, sometimes vomiting and diarrhea, anemia and hyperphosphatemia. Nine dogs were hypertensive; 16 dogs were proteinuric (UPC>0.5) (range 0.75 to 8.85). Fourteen dogs had isosthenuria and 6 dogs had cylinduria. Average plasma creatinine at the time of biopsy was 5.75 mg/dl (508.3 µmol/l). The pathophysiology of juvenile renal disease in Boxers needs further elucidation.

Conflicts of interest: No conflicts of interest reported.

ESVNU-O-7

RENAL HEMATURIA IN GOLDEN RETRIEVERS: A NEWLY RECOGNIZED DISORDER. J. Ben Oz¹, S. Jessen², N. Kahane¹, G. Segev¹. ¹Koret School of Veterinary Medicine, The Hebrew University of Jerusalem, REHOVOT, Israel, ²Vetcenter clinic, HERZELIA, Israel

Renal hematuria is an uncommon cause of hematuria in dogs. Recently, we have identified chronic renal hematuria of unknown cause in Golden Retrievers. This retrospective study is aimed to characterize the history, physical examination, clinicopathologic, ultrasonographic and histopathologic findings, as well as the disease progression and prognosis of affected dogs. Twenty client owned Golden Retrievers that presented to the Hebrew University Veterinary Teaching Hospital between 2004-2013 with a complaint of persistent hematuria (>30 days) or intermittent hematuria (>1 year), were included. Data were collected from the medical records. Follow up data were obtained via telephonic interview and rechecks at the teaching hospital or the referring veterinarian.

Median age of onset of hematuria was 48 months (range, 6-132). One third of the dogs presented with hematuria at 2 years of age or

younger. Bleeding episodes lasted for a median of 12 months (range, 2 days to 74 months). Hematuria was present for more than 90% of the time in 7/13 dogs. Median hematocrit, creatinine concentration, and UPC at presentation were 46% (range, 24-52%), 1.2 mg/dL (range, 0.78-4.95), and 0.4 (n = 8, range, 0.15-2.58), respectively. Renal ultrasonographic abnormalities were documented in 11/15 dogs, and included mottled areas (60%), poor corticomedullary definition (54%), hyperechogenicity (53%), and heterogenic mass (7%). The median follow-up time (17 dogs) was 35 months (range, 1-66). Eleven dogs were still alive at follow-up. Median creatinine concentration at last follow up was 1.23 mg/dL and 4/20 dogs developed azotemia. The quality of life was graded 4/5 and was related mostly to concurrent diseases. Despite the persistent and severe hematuria, only 2 dogs required blood transfusions. Six dogs were dead at follow-up (4 euthanized), of these, 2 due to progression of renal disease, one due to persistent hematuria, and the others due to unrelated causes. Histopathology was available in 2 dogs; abnormalities included cortical interstitial fibrosis, hematoidin accumulation in tubular epithelium, focal interstitial nephritis, and presence of amorphous material with brown to black-laden macrophages between the tubules.

This is the first report of renal hematuria in Golden Retrievers. Despite persistent hematuria, hematocrit remained stable and blood transfusion were rarely required. Although ultrasonographic changes were common and occasionally severe, kidney function remained stable and good quality of life and longevity were maintained. The mode of inheritance of this disorder needs to be further investigated, and a larger scale surveys are warrant to assess the prevalence of the disorder.

Conflicts of interest: No conflicts of interest reported.

ESVNU-O-8

SERUM CYSTATIN C IN DOGS WITH DIABETES MELLITUS OR CUSHING'S SYNDROME. S. Marynissen¹, P. Smets¹, L. Ghys¹, D. Paeppe¹, J. Delanghe², H.P. Lefebvre³, S. Daminet¹. ¹University of Ghent, faculty of Veterinary Medicine, MERELBEKE, Belgium, ²University of Ghent, Faculty of Medicine and Health Sciences, GHENT, Belgium, ³École Nationale Vétérinaire de Toulouse, TOULOUSE, France

Glomerular and tubular alterations have been described in dogs with Cushing's syndrome or diabetes mellitus. Early diagnosis of renal dysfunction is crucial. Routine renal markers are insensitive and GFR measurements time-consuming. New renal markers, such as serum Cystatin C (sCysC), used as a GFR marker in human nephrology, have gained much interest. Information on sCysC in dogs is scarce. This study aimed to validate a nephelometric assay for canine sCysC measurement and evaluate sCysC in dogs with DM or CS.

Renal function was monitored in 22 dogs with CS and 14 dogs with DM, for 12 and 6 months respectively. Baseline data were collected from seventeen healthy dogs, of comparable age and bodyweight. Serum creatinine (sCr), serum urea and urinary protein-to-creatinine ratio (UPC) were determined. After analytical validation, sCysC levels were measured with a particle-enhanced nephelometric immuno assay (PENIA) using the human BN ProSpec analyzer. A combined plasma exogenous creatinine-iohexol clearance test was performed, to determine GFR, as previously described.

The PENIA measured CysC in a linear ($R^2 = 0.995$) and precise manner (inter-assay coefficient of variation (CV): 1.6%, intra-assay CV: 2.9%). A significant inverse effect of creatinine clearance on sCysC was found for the whole group ($p < 0.05$). When compared with the DM (28%) and healthy group (29%), a significant higher percentage of CS dogs (60%) had proteinuria (UPC>0.5) at baseline ($p < 0.05$). No significant differences were found between the 3 groups for sCysC or GFR at baseline.

No clinically relevant changes were observed for sCr over time. Thirty-three percent of diabetic dogs had persistent proteinuria despite treatment. In the CS group, a significant decrease in UPC ratio was observed after initiating treatment ($p < 0.05$), although 33% had persistent proteinuria. Although a significant decrease over time was observed for sCysC within the DM group ($p < 0.05$), no significant changes were observed for sCysC levels within the CS group. A significant decrease was observed for

GFR within the CS group ($p < 0.05$). No significant changes for GFR were found within the DM group.

Our results show that PENIA is a valid method for measuring sCysC in dogs. CysC could be a useful GFR marker, although the current study could not enhance this. Further research determining the usefulness of sCysC in dogs is necessary. Post-treatment decline of GFR in dogs with CS and persistence of proteinuria both in dogs with CS and DM warrants the clinician's attention.

Conflicts of interest: No conflicts of interest reported.

SCH-O-1

SKYE TERRIER HEPATITIS RE-APPRAISED: IS THIS A CONGENITAL DUCTAL PLATE ABNORMALITY?

P. Watson, M. Reading, C. Constantino-Casas. University of Cambridge, CAMBRIDGE, United Kingdom

Skye terrier hepatitis was first reported in 1988 in nine related dogs, with only a single case report since then in 2003. There was accumulation of copper in the liver which differed from copper storage disease in other breeds. A disorder of intracanalicular cholestasis was proposed.

An 11 month-old entire male Skye terrier intended as a stud dog presented to the first author to investigate a 3 week history of inappetence, lethargy and ascites. Investigations revealed elevations in pancreatic and liver enzymes, marked hypoalbuminaemia, an abdominal transudate and normal coagulation times. Biopsies of liver and pancreas were taken. Bile cytology and culture were negative. The pancreas section was histologically unremarkable. The liver in more normal areas exhibited moderate small irregular bile duct proliferation, endothelial hyperplasia, reduced portal vein profiles and fibrosis. In grossly abnormal areas, hepatocytes were replaced almost entirely by large amounts of fibrous tissue admixed with large numbers of small bile ducts. Inflammation was minimal. Scattered clusters of brown pigment laden macrophages or hepatocytes were present multifocally. Perl's stain (haemosiderin) was strongly positive in these areas and so was rhodanine stain (copper). Bile plugs of green pigment within canaliculi (Fouchet's stain positive) were present. The histological findings were consistent with previously reported juvenile hepatic fibrosis in dogs.

In the light of these findings, the liver of a related dog (5.5 year old female) from the same breeder which had been examined at post mortem was re-assessed. This dog showed similar but more severe changes with marked biliary hyperplasia and abnormal bile duct profiles within large tracts of fibrous tissue. Perl's and rhodanine stains showed marked accumulation of both iron and copper. The findings were consistent with juvenile hepatic fibrosis.

Further questioning of the breeder revealed that the mother of the first case had a clinical diagnosis of 'cirrhosis' but was still alive with no hepatic histology. Anecdotally, there were also reports of a juvenile renal disease in young adult related Skye terriers which has not been previously described.

These findings are strongly suggestive of an inherited ductal plate abnormality in the liver of these Skye terriers. This is not inconsistent with the original published reports and could explain the secondary accumulation of iron and copper. Further studies are underway to obtain further liver and kidney tissues from affected dogs and to stain livers with cytokeratin 7 for juvenile bile ducts.

Conflicts of interest: No conflicts of interest reported.

SCH-O-2

LONG-TERM MANAGEMENT OF COPPER ASSOCIATED HEPATITIS IN THE LABRADOR RETRIEVER.

H. Fieten¹, V.C. Biourge², T.S.G.A.M. van den Ingh³, A.L. Watson², P.A.J. Leegwater¹, J. Rothuizen¹. ¹Faculty of Veterinary Medicine, UTRECHT, The Netherlands, ²Royal Canin Research Centre, AIMARGUES, France, ³TCCI Consultancy BV, UTRECHT, The Netherlands

Introduction: Copper associated hepatitis in the Labrador retriever is a hereditary disease in which both genetic factors and

environmental factors, like dietary intake of copper and zinc, play a role in the pathogenesis^{1,2}. Hepatic copper accumulation is accompanied by an inflammatory infiltrate and copper levels are typically between 1000 and 5000 mg/kg dry weight liver at first presentation. Chelation therapy with D-penicillamine is the initial treatment of choice. However, lifelong continuous chelation therapy carries the risk of overtreatment in this breed and may lead to unwanted low body copper and zinc concentrations.³

Aims: In the current study we evaluated the use of a low copper, high zinc diet in the long term management of copper associated hepatitis in Labrador retrievers.

Methods: Sixteen Labrador retrievers affected with copper associated hepatitis were studied longitudinally after normalization of their hepatic copper due to D-penicillamine treatment. The dogs were maintained on a low copper, high zinc diet. Liver biopsies were taken every 6 months to measure hepatic copper concentration and to histologically evaluate hepatitis and fibrosis.

Results and conclusion: We observed variation in the re-accumulation rate of hepatic copper. Dietary treatment alone was sufficient to maintain hepatic copper concentration at a safe level in the majority of dogs during the follow up period, which ranged from 17 until 39 months in these dogs. Four dogs needed re-treatment with D-penicillamine after a median follow up period of 15.7 months (range 5.9-19.2 months). Re-accumulation of hepatic copper was accompanied by a worsening of hepatitis grade and stage of fibrosis.

We propose the use of D-penicillamine in combination with dietary treatment alternating with dietary treatment alone as a good strategy for long-term management of Labrador retrievers affected with copper associated hepatitis.

Conflicts of interest: Adrian Watson and Vincent Biourge are employed at Royal Canin, which financially supported this study and provided the diet that was used in the study.

SCH-O-3

EVALUATION OF SERUM PROCOLLAGEN TYPE III N-TERMINAL PEPTIDE AS A POTENTIAL BIOMARKER FOR CANINE HEPATIC FIBROSIS.

A. Lidbury¹, A. Rodrigues Hoffman¹, J.K. Fry², J.S. Suchodolski¹, J.M. Steiner¹. ¹Texas A&M University, COLLEGE STATION, United States of America, ²Gulf Coast Veterinary Specialists, HOUSTON, United States of America

Serum biomarkers of hepatic fibrosis have been developed for use in human patients. Pro-collagen type III N-terminal peptide (PIIINP) is a component of the extracellular matrix. During fibrosis greater amounts of this protein are released into the blood stream due to an increased rate of extracellular matrix turnover. In humans the reported sensitivities and specificities of serum PIIINP concentration for differentiating patients with no to mild hepatic fibrosis from those with moderate to severe hepatic fibrosis range from 60-80% and from 74-87%, respectively. The aim of this study was to assess the utility of PIIINP as a biomarker for canine hepatic fibrosis.

Serum samples from 53 dogs with hepatic disease were collected. These were divided into groups as follows: 18 dogs (34%) with chronic hepatitis, 17 (32%) with hepatic neoplasia, 6 (11%) with congenital portosystemic shunts, and 12 (22%) with other hepatobiliary diseases. Liver biopsy specimens from 43 of these dogs were available for assessment. Serum samples from 50 healthy control dogs were also collected. All hepatic samples were evaluated histologically to determine the degree of fibrosis. Fibrosis was staged using a 5-point scheme: normal, minimal, mild, moderate, or severe. Serum PIIINP concentrations were measured using a commercially available RIA (UniQ PIIINP RIA, Orion Diagnostica). The data were tested for normality and the differences in PIIINP concentrations between the groups of dogs were analysed using Mann-Whitney or Kruskal-Wallis tests as appropriate. Significance was set as $P < 0.05$.

The median (min-max) serum PIIINP concentrations for dogs with liver disease and healthy dogs were 9.3 (3.1-50.0) and 12.2 (3.8-50.0) $\mu\text{g/L}$, respectively ($P < 0.01$). The median serum PIIINP concentrations for dogs with chronic hepatitis, hepatic neoplasia, congenital portosystemic shunts, and other hepatobiliary diseases were 9.6 (3.1-50.0), 8.1 (3.3-35.3), 12.7 (4.1-23.6),

and 8.9 (3.3-50.0) µg/L, respectively ($P = 0.56$). The median serum PIIINP concentrations for dogs with no to mild hepatic fibrosis ($n = 30$) and dogs with moderate to severe hepatic fibrosis ($n = 13$) were 9.4 (3.3-12.8) and 10.9 (3.1-50.0) µg/L, respectively ($P = 0.68$).

Contrary to what we expected serum PIIINP concentrations were significantly higher in healthy dogs than in dogs with liver disease. There was no significant difference in serum PIIINP concentration between dogs with no to mild and dogs with moderate to severe hepatic fibrosis. The results of this study do not support the utility of serum PIIINP as a biomarker for canine hepatic fibrosis.

Conflicts of interest: No conflicts of interest reported.

SCH-O-4

CHARACTERISATION OF CANINE ADIPOSE AND BONE MARROW-DERIVED MESENCHYMAL STEM CELLS AND HEPATOCYTE DIFFERENTIATION CAPACITY. A. Gow¹, D. Hay², D.J. Argyle¹. ¹The Roslin Institute and R(D)SVS, EDINBURGH, United Kingdom, ²Centre for Regenerative Medicine, EDINBURGH, United Kingdom

An in-vitro supply of functional canine hepatocytes would be an invaluable resource for drug and toxicology testing. As hepatocyte function declines precipitously in-vitro, a renewable supply from stem cells is an attractive option. Reports of mesenchymal stem cell (MSC) to hepatocyte differentiation has been reported in a variety of species, including one report of hepatic differentiation of canine amniotic MSC which showed gene expression of albumin, tyrosine aminotransferase, and alpha-1 antitrypsinase. Hepatocyte differentiation of MSC from a more widely available source such as adipose tissue or bone marrow would be more useful.

Canine adipose and bone marrow-derived MSC's were characterised by their ability to adhere to plastic, differentiate into osteocytes, adipocytes and chondrocytes as demonstrated by positive alizarin red, oil red O, and toluidine blue staining. Both MSC types were shown to express cell surface markers CD105, CD44, STRO-1 and lack CD45, CD19 and CD14 expression.

A three stage hepatocyte differentiation protocol consisting of serum-free DMEM (SF-DMEM), EGF and bFGF for 48 hours, SF-DMEM, HGF, nicotinamide and bFGF for 7 days and finally hepatocyte culture media (Lonza) with oncostatin M, dexamethasone and DMSO for 41 days. Time points 0,14, 30 and 50 days were used for gene expression and functional characteristics such as PAS staining for glycogen storage, indocyanine green (ICG) uptake, cytochrome P450 (CYP) activity. Canine primary hepatocytes were used as a positive control.

Over the differentiation protocol, cell morphology changed dramatically to polygonal cells with a large amount of cytoplasm. Faint PAS staining and ICG uptake/excretion was demonstrated by day 30. No CYP activity was found at any time point. Using real-time PCR, no evidence of hepatocyte gene expression was demonstrated over the length of the protocol.

Using this protocol, no evidence of hepatocyte-like induction of canine adipose or bone marrow-derived MSC's was demonstrated. Alternative methods of in-vitro hepatocyte production will be explored.

Conflicts of interest: No conflicts of interest reported.

SCH-O-5

LOW DENSITY LIPOPROTEIN (LDL) AND ACETYLATED-LDL (AC-LDL) UPTAKE OF PRIMARY CANINE HEPATOCYTES AND CANINE MESENCHYMAL STEM CELLS (MSC). A. Gow¹, D. Hay², D.J. Argyle¹. ¹The Roslin Institute and R(D)SVS, EDINBURGH, United Kingdom, ²Centre for Regenerative Medicine, EDINBURGH, United Kingdom

Demonstrating that stem cells have been differentiated into hepatocyte-like cells requires a battery of functional tests. One functional test frequently used is uptake of fluorescent labelled

LDL. Both labelled native LDL and Ac-LDL are commercially available. Cultured murine and human fibroblasts have been reported to internalise DiI-LDL. Ac-LDL is reported to be taken up by macrophages and endothelial cells. Hepatocytes possess a specific LDL receptor that should fail to recognise the acetylated ligand. Despite this, papers describing hepatocyte differentiation of stem cells in various species use the two compounds with almost equal frequency.

This study was designed to investigate LDL and Ac-LDL uptake of canine primary hepatocytes to assess which compound should be utilised in demonstration of canine hepatocyte-like qualities. Undifferentiated canine adipose and bone marrow-derived MSC were also tested.

Canine primary hepatocytes were cultured from three dogs euthanased for behavioural reasons using the wedge perfusion method. Adipose and bone marrow-derived MSC's were also cultured from two dogs. After 48 hours in culture, cells were incubated in media containing 20 µg/ml of DiI-LDL or DiI-Ac-LDL for 3 hours. Wells were then washed three times, fixed in 4% formaldehyde and nuclear counterstaining performed with DAPI. Wells were then examined for lipid uptake using fluorescent microscopy.

All three canine primary hepatocyte cultures demonstrated strong uptake of DiI-LDL with only sporadic cells showing Ac-LDL uptake. Both undifferentiated adipose and bone marrow mesenchymal stem cells contained DiI-LDL but not DiI-Ac-LDL.

These results show that for low-density lipoprotein uptake to demonstrate canine hepatocyte-like properties during stem cell differentiation, documentation of DiI-LDL and not Ac-DiI-LDL uptake is required. The sparse Ac-LDL uptake in hepatocyte culture is likely to be contaminating Kupffer and/or endothelial cells. Furthermore, both canine adipose and bone marrow-derived MSC's show strong DiI-LDL uptake in their undifferentiated state, preventing this from being a useful test in canine MSC to hepatocyte differentiation protocols.

Conflicts of interest: No conflicts of interest reported.

ESVIM-O-1

IS THE CXCL8-CHEMOKINE INVOLVED IN THE BREED PREDISPOSITION OF WEST HIGHLAND WHITE TERRIER TO CANINE IDIOPATHIC PULMONARY FIBROSIS ? E. Roels¹, E. Krafft¹, H.P. Laurila², M.M. Rajamäki², N. Antoine¹, M. Massart¹, C. Clercx¹. ¹University of Liège, LIÈGE, Belgium, ²University of Helsinki, HELSINKI, Finland

Canine idiopathic pulmonary fibrosis (canine IPF) is a progressive interstitial lung disease of unknown aetiology and pathophysiology, mainly described in middle-aged to old West Highland white terriers (WHWT). CXCL8 (IL-8) is a proinflammatory chemokine probably involved in the pathogenesis of human IPF, where it appears to be a diagnostic and prognostic biomarker in serum and BALF. In dogs, little is known about the role of CXCL8 in the pathogenesis of IPF. Recently an increased CXCL8 gene expression has been shown in lung tissues from IPF WHWT compared to control dogs from other breeds. The aim of the present study was to compare serum CXCL8 levels in healthy WHWT versus WHWT with IPF and healthy dogs from terrier breeds other than WHWT and from non-terrier breeds.

Ten WHWT with IPF (mean age 10 years, range 5-14) and 71 healthy dogs, including 12 WHWT (9, 3-17), 10 Scottish terrier (6, 1-10), 10 Jack Russell terrier (7, 1-12), 10 Maltese (6, 1-13), 9 Cavalier King Charles Spaniel (4, 1-8), 10 Labrador Retriever (6, 2-12) and 10 Malinois Belgian Shepherd (5, 3-8) entered the study. Health status was based on clinical examination, serum biochemistry and haematology, as well as high-resolution computed tomography in 9/12 WHWT. IPF was confirmed by histopathology. Serum CXCL8 concentrations were determined by ELISA (Canine CXCL8/IL-8 Quantikine ELISA Kit, R&D systems). Results between IPF and healthy WHWT, and between healthy dogs of different breeds were compared using a global linear model (SAS[®] software) incorporating the effects of covariates age and gender; $p \leq 0.05$ was chosen as level of significance.

Serum CXCL8 concentrations are expressed in pg/ml and given as $\text{lsmean} \pm \text{standard error}$. No difference in serum CXCL8 levels was detected between WHWT with IPF ($4634,3 \pm 541,5$) and healthy WHWT ($3806,7 \pm 369,4$). However, serum CXCL8 concentration was significantly higher in healthy WHWT compared to each of the other groups of healthy dogs. Moreover, serum CXCL8 concentrations were higher in Scottish terrier ($2685,2 \pm 314,4$) and Maltese ($2563,8 \pm 339,7$) than in Cavalier King Charles Spaniel ($1527,5 \pm 348,8$) in which the lowest concentration was found.

Results of the present study show that serum CXCL8 concentration is high in both healthy and IPF WHWT compared to healthy dogs from other breeds. Therefore, CXCL8 (1) cannot be used as a serum diagnostic biomarker for IPF; (2) might be related to the breed predisposition of the WHWT for IPF.

Conflicts of interest: No conflicts of interest reported.

ESVIM-O-2

IMMUNOHISTOCHEMICAL ANALYSIS OF TRANSFORMING GROWTH FACTOR BETA 1 AND ITS SIGNALLING PATHWAYS IN CANINE IDIOPATHIC PULMONARY FIBROSIS. E. Krafft¹, H.P. Laurila², M.M. Rajamäki², M. Myllärniemi³, M.J. Day⁴, P. Lybaert⁵, C. Clercx¹. ¹Faculty of veterinary medicine, University of Liège, LIÈGE, Belgium, ²Department of Equine and Small Animal Medicine Faculty of Veterinary Medicine, HELSINKI, Finland, ³Department of Medicine, Division of Pulmonary Medicine, University of Helsinki, HELSINKI, Finland, ⁴School of Veterinary Sciences University of Bristol, BRISTOL, United Kingdom, ⁵Faculté de Médecine, Université Libre de Bruxelles, BRUXELLES, Belgium

The aetiology and pathogenesis of canine idiopathic pulmonary fibrosis (IPF) are poorly understood. A genetic basis is suspected because of the predisposition of the West Highland white terrier (WHWT). In previous studies, we have showed that serum concentration of the profibrotic protein transforming growth factor beta 1 (TGFB1) is elevated in both healthy WHWTs and WHWTs with IPF, and that TGFB1 activating pathways are modified in IPF lungs. The present study compared the expression and localization of TGFB1 and of proteins involved in TGFB1 signalling: TGF receptor type 1 (TBR1) and the intracellular messenger, phosphorylated Smad2/3 protein (P-Smad2/3) in the lungs of healthy dogs and dogs with IPF.

Sections from formalin-fixed lung tissues were obtained from ten WHWTs with IPF and six control dogs of various breeds. The primary rabbit antibodies used allowed detection of TGFB1, TBR1 and P-Smad2/3. Bound antibodies were visualized using diaminobenzidine. A qualitative assessment was made by comparing localisation and intensity of labelling for each primary antibody in the lungs of both groups of dog.

In sections from control dogs, strong and diffuse TGFB1 labelling was seen in the layer of fibrous connective tissue surrounding bronchi and bronchioles and weaker granular expression was seen in bronchial and vascular smooth muscles. In contrast, bronchial and bronchiolar epithelial cells had distinct apical expression of TBR1 but there was only weak granular labelling of the smooth muscles and no expression within the layer of fibrous connective tissue. In IPF lung sections, the same pattern of immunoreactivity was observed for both antibodies in tissues surrounding the bronchi and bronchioles. In areas of pulmonary fibrosis, there was a strong and diffuse expression of TGFB1 in the fibrous matrix. In contrast, TBR1 was not visible within the fibrotic tissue, but there was very strong labelling of individual alveolar epithelial cells, particularly within hyperplastic pneumocytes. Some alveolar macrophages also appeared to express TBR1. P-Smad2/3 nuclear labelling was observed in both groups, but in IPF lungs intense positive nuclear labelling was observed in altered alveolar epithelial cells.

In IPF lungs, there was intense TGFB1 labelling in the fibrous matrix, consistent with a high amount of TGFB1 in fibrotic areas and increased TGFB1 extracellular storage. Hyperplastic pneumocytes appeared to be an important target for TGFB1 in canine

IPF. The finding of intense nuclear labelling for P-Smad2/3 in the alveolar epithelium confirmed active TGFB1 signalling in affected lung tissue.

Conflicts of interest: No conflicts of interest reported.

ESVIM-O-3

IS 5-HYDROXYTRYPTAMINE (SEROTONINE) INVOLVED IN THE PATHOGENESIS OF IDIOPATHIC PULMONARY FIBROSIS IN DOGS? E. Krafft¹, E. Roels¹, HP. Laurila², MM. Rajamäki², F. Farnir¹, N. Antoine¹, C. Clercx¹. ¹Faculty of veterinary medicine, University of Liège, LIÈGE, Belgium, ²Department of Equine and Small Animal Medicine Faculty of Veterinary Medicine, HELSINKI, Finland

Idiopathic pulmonary fibrosis (IPF) in dogs is an emerging chronically progressive interstitial lung disease of unknown aetiology, characterized by irreversible scarring of the lung that mainly occurs in the West Highland White Terrier (WHWT). Like in humans, IPF is unresponsive to current therapy and the basic mechanisms underlying pathogenesis remain unknown. Among the many factors (growth factors, cytokines and chemokines) that have been involved in the pathogenesis of IPF in humans, the role of serotonin (5-hydroxytryptamine 5HT) is currently investigated. The aim of the present study was to determine serum 5HT level in dogs with IPF versus healthy dogs and dogs with other chronic pulmonary diseases and to evaluate lung expression of proteins involved in 5HT signalling: 5HT receptor type 2A (HTR2A) and type 2B (HTR2B) and 5HT transporter (5HTT).

Serum samples were collected from 13 dogs with IPF (10 WHWTs, 2 Scottish terriers, 1 Yorkshire terrier, mean age 11.6 years), 9 dogs with chronic bronchitis (CB) (various breeds, 9.3 years), 10 dogs with eosinophilic bronchopneumopathy (EBP) (various breeds, 7.5 years), 9 healthy WHWTs (9.4 years). Serum 5HT concentrations were determined by ELISA. Group influence on serum 5HT concentration was analyzed by covariance analysis (ANOVA). Total RNA was extracted from lung tissue from 14 dogs with histopathologically-confirmed IPF (12 WHWTs, 1 Scottish terrier, 1 Lhasa Apso, mean age 13 years) and 11 control dogs (various breeds, 6.3 years). Expression of HTR2A, HTR2B and 5HTT was measured by qRT-PCR. For each gene, a relative copy number was calculated for each sample and results were normalized using two stably expressed housekeeper genes (RPS18 and TBP). Lung expression levels in the two groups were compared using a Student t-test or a Mann-Whitney Rank sum test.

Serum 5HT concentration in IPF dogs was not statistically different from healthy WHWTs, dogs with CB or with EBP. Expression of all genes was not statistically different between IPF and control lung tissues.

Results of the present study excluded 5HT as a potential blood biomarker for canine IPF and are not in favour of a major implication of 5HT pathways in the pathogenesis of canine IPF.

Conflicts of interest: No conflicts of interest reported.

ESVIM-O-4

6-MINUTE WALK TEST IN WEST HIGHLAND WHITE TERRIERS WITH IDIOPATHIC PULMONARY FIBROSIS AND CLINICAL FEATURES DURING DISEASE PROGRESSION. L.I.O. Lilja-Maula, H.P. Laurila, A.K. Lappalainen, P. Syrjä, M.M. Rajamäki. University of Helsinki, HELSINKI, Finland

Our objective was to evaluate exercise tolerance in West Highland White Terriers (WHWTs) with idiopathic pulmonary fibrosis (IPF) using six-minute walk test (6MWT) and describe changes in chosen parameters during disease progression. 12 IPF WHWTs, (median follow-up time 16 (range 4-40) months, controlled regularly until death or study endpoint), and for 6MWT, 5 age-matched healthy WHWTs were recruited. Dogs were walked along a 64-m straight corridor at their own pace for 6 minutes. Distance covered (6MWD) and heart rate (HR), body temperature, oxygen saturation (SpO₂) and arterial blood gases were measured pre- and post-walking. Nonparametric statistics was used for 6MWT results and changes in clinical parameters from initial to last available measurement. IPF WHWTs (6/12)

walked a shorter distance, median 398 m (range 273-519 m), than controls, median 492 m (420-568 m), $P = 0.05$. Arterial partial pressure of oxygen (PaO_2) in diseased dogs seemed to have a moderate positive correlation with 6MWD ($\tau = 0.69$, $P = 0.06$). Post-HR was significantly higher than pre-HR in IPF WHWTs ($P = 0.04$). During disease course, no change was noted in body weight, $P = 0.31$, or hematocrit, $P = 0.06$. Severity of bronchointerstitial radiographic pattern, initially classified as moderate or severe, varied to either direction. A declining trend in PaO_2 values and increase in alveolar-arterial oxygen gradient were detected, and were significant in WHWTs dying of IPF-related causes (5/12), $P = 0.04$. 6MWT proved to be a well-tolerated test to evaluate exercise tolerance in IPF WHWTs. 6MWD could serve as a noninvasive parameter to evaluate lung function during disease, in addition to accurate PaO_2 .

Conflicts of interest: No conflicts of interest reported.

ESVIM-O-5

SURVIVAL AND PROGNOSTIC FACTORS IN WEST HIGHLAND WHITE TERRIERS WITH IDIOPATHIC PULMONARY FIBROSIS. H.P. Laurila¹, L.I.O. Lilja-Maula², A.K. Lappalainen², P. Syrjä³, E. Krafft⁴, C. Clercx⁴, M.M. Rajamäki². ¹Faculty of Veterinary Medicine, University of Helsinki, HELSINKI, Finland, ²Department of Equine and Small Animal Medicine, Faculty of Veterinary Medicine, UNIVERSITY OF HELSINKI, Finland, ³Department of Basic Veterinary Sciences, Faculty of Veterinary Medicine, UNIVERSITY OF HELSINKI, Finland, ⁴Department of Clinical Sciences, Faculty of Veterinary Medicine, UNIVERSITY OF LIÈGE, Belgium

Idiopathic pulmonary fibrosis (IPF) is an incurable interstitial lung disease affecting mainly older West Highland White Terriers (WHWTs). Its effect on lifespan is unclear. Our objective was to evaluate survival and prognostic factors in WHWTs with IPF. We conducted a prospective case-control study (years 2007-2012). We recruited privately owned WHWTs, 15 with IPF and 10 age-matched healthy controls. IPF was confirmed by histopathology or high-resolution computed tomography (HRCT). Descriptive statistics and Kaplan-Meier (KM) survival curves with Cox proportional hazard ratios were performed for survival analysis. The chosen potential prognostic factors predicting survival were arterial partial pressures of oxygen and carbon dioxide, alveolar-arterial oxygen gradient, serum endothelin-1 concentration, severity of changes in thoracic radiographs, HRCT Hounsfield unit values and complete blood cell count values. KM curves, Cox-regression analysis or logistic regression models were used for prognostic factor analysis. The median all-cause survival of deceased WHWTs with IPF (13/15) from onset of symptoms was 27 months (range 2-51 months) and the median IPF-specific survival (7/15) was 32 months (range 2-51 months). The risk of death in WHWTs with IPF was significantly higher (hazard ratio 4.6; 95% confidence interval 1.05-19.74, $P = 0.04$) than in control WHWTs. No significant prognostic factors were identified. Four of the five dogs euthanized due to acute dyspnea had diffuse alveolar damage in the lungs. IPF had a negative impact on life expectancy of WHWTs, but individual survival varied considerably. IPF in WHWTs may therefore have both a rapid and a slowly progressive disease course like human IPF.

Conflicts of interest: No conflicts of interest reported.

ESVIM-O-6

LONG TERM FOLLOW UP IN DOGS WITH EOSINOPHILIC BRONCHOPNEUMOPATHY TREATED WITH INHALED GLUCOCORTICOSTEROID THERAPY: PRELIMINARY RESULTS. A.M. Canonne, F. Billen, C. Clercx, Faculty of Veterinary Medicine, University of Liège, Belgium, LIÈGE, Belgium

The classic treatment of dogs with idiopathic eosinophilic bronchopneumopathy (EBP) consists in long-term oral administration of prednisolone at the minimum effective dose. Unfortunately, chronic systemic glucocorticosteroid therapy may lead

to iatrogenic hyperadrenocorticism. Therefore, alternative treatment with inhaled steroid administration has been increasingly used, although the clinical response to this therapy has not been evaluated in dogs with EBP. The aims of this study were (1) to assess the clinical response to long-term inhaled steroid therapy with fluticasone (IST), and (2) to investigate possible side effects including inhibition of pituitary-adrenal axis (PAA).

Six dogs with EBP (2 Whippet, 2 Brittany Spaniel, 1 Border Terrier and 1 Husky, 4 females and 2 males, median age = 47 months [12 - 85]), diagnosed based on clinical, blood, radiographical, bronchoscopic examinations and bronchoalveolar lavage fluid analysis, and treated with IST only for more than 1 month, were retrospectively recruited. At the time of diagnosis, duration of clinical signs varied from 2 to 12 months (median = 7.5); a clinical severity index (CSI), assigned based on clinical signs (cough 0-3, retch 0-1, exercise intolerance 0-1) was calculated and varied between 3 and 5.

At the time of recheck (median 30.5 months [6 weeks - 4 years]), a CSI was calculated and an ACTH stimulation test was performed. Fluticasone dosage varied between 100 and 250 μg BID. CSI decreased in all dogs and varied from 1 to 3. Three dogs showed persistent cough under medication (once a week in 2 dogs, daily in one). In the 3 other dogs, cough reappeared as soon as treatment was not regularly administered. In two dogs treated with IST for more than 2 years, ACTH stimulation test revealed inhibition of PAA; only one dog had overt clinical signs of iatrogenic hyperadrenocorticism.

This preliminary study indicates that treatment of EBP with long term IST does not appear to allow a proper management in all affected dogs. Besides, long term IST may induce inhibition of PAA. A prospective study including a higher number of dogs treated with IST and with regular control of the PAA is warranted.

Conflicts of interest: No conflicts of interest reported.

ESVIM-O-7

C-REACTIVE PROTEIN IN CANINE LOWER AIRWAY DISEASES AND IN CARDIOGENIC PULMONARY EDEMA. S.J. Viitanen, H.P. Laurila, L.I.O. Lilja-Maula, M.A. Melamies, M.M. Rajamäki. University of Helsinki, Faculty of Veterinary Medicine, UNIVERSITY OF HELSINKI, Finland

C-reactive protein (CRP) is a major acute phase protein having low serum concentrations in healthy dogs and rising rapidly after inflammation. The aim of the study was to investigate CRP levels in various canine respiratory diseases and to examine whether CRP can be used as a biomarker in the diagnostics of pneumonia.

A prospective cross-sectional observational study was conducted and a study population of 106 dogs with different respiratory diseases (22 with bacterial pneumonia (BP), 17 with primary bacterial tracheobronchitis (PBTB), 20 with eosinophilic bronchopneumopathy (EBP), 20 with chronic bronchitis (CB), 12 with idiopathic pulmonary fibrosis (IPF), and 15 with cardiogenic pulmonary edema (CPE)) was collected. Diagnosis was confirmed in all dogs by thorough clinical examinations and additionally by bronchoalveolar lavage, histopathology or echocardiography. 72 healthy blood donor dogs were included as controls. CRP was measured using quantitative immunoassay (LifeAssays[®] Canine CRP Test Kit). The differences between groups in CRP were evaluated with analysis of variance models (ANOVA).

Dogs with BP had significantly higher serum CRP concentrations (median 121 mg/l, interquartile range 68-178 mg/l) compared to dogs with PBTB (23, 15-38, $p = 0.0003$), CB (13, 8-14, $p < 0.0001$), EBP (5, 5-15, $p < 0.0001$), IPF (17, 10-20, $p < 0.0001$), CPE (19, 13-32, $p < 0.0001$) and to healthy controls (14, 8-20, $p < 0.0001$). Dogs with PBTB had significantly higher CRP -levels compared to dogs with CB ($p = 0.001$), EBP ($p < 0.0001$) and to healthy controls ($p = 0.03$). These results indicate that CRP has potential to be used as an additional biomarker especially in the diagnostics of bacterial pneumonia.

Conflicts of interest: No conflicts of interest reported.

ESVIM-O-8
EFFECTS OF LOW DOSE CLOPIDOGREL ON PLATELET FUNCTION IN HEALTHY DOGS. K.M. Biermann, R. Mischke. University of Hannover, HANNOVER, Germany

Clopidogrel is a potent platelet inhibitor that is widely used in humans and has increasing relevance in dogs and cats. The aim of this study was to assess platelet function in healthy dogs treated with three different doses of clopidogrel using different methods for evaluation of platelet function.

Eighteen healthy dogs of different breeds were divided equally into 3 groups and were treated orally with 0.5 mg/kg clopidogrel (group 1), 1 mg/kg (group 2) and 2 mg/kg (group 3) over 7 days. Blood sampling was performed before treatment and on day 1, 2, 3, 4 and 7. Platelet function was assessed by using the Multiplate[®] impedance aggregometer with three different agonists: collagen (COL), adenosine diphosphate (ADP) and aspirin (ASPI). Additional tests of primary hemostasis included platelet count, capillary bleeding time, automated platelet function analyser (PFA-100[®]) and turbidimetric aggregation. Data were analysed by 2-way ANOVA for repeated measures and subsequent 1-way ANOVA for individual groups followed by t-tests as indicated. Alpha was set at 5%.

2-way ANOVA revealed significant changes over time for ADP- and ASPI-induced impedance aggregometry (area under the curve [AUC] values), capillary bleeding time and closure time measured with the PFA collagen/ADP cartridge. Significant differences between dosage groups were demonstrated only for capillary bleeding time and ADP-induced impedance aggregometry. ASPI-induced impedance aggregometry decreased significantly starting from day 1 in group 2 and 3 and from day 2 in group 1. Both, ADP-induced impedance aggregometry and closure time of the PFA collagen/ADP cartridge showed only tendencies towards lower values in groups 2 and 3, receiving higher dosages. Only in group 3, capillary bleeding time increased significantly compared to baseline.

In conclusion, results of this study reveal a limited effect of low dosages such as 0.5 mg/kg clopidogrel on platelet function tests in dogs. However, higher dosages of at least 2 mg/kg seem to be necessary to significantly inhibit platelet function for clinical use.

Conflicts of interest: No conflicts of interest reported.

ESVIM-O-9
EVALUATION OF CHANGES IN COAGULATION TIMES (PT AND APTT) AFTER INTRAVENOUSLY VITAMIN K ADMINISTRATION IN 73 DOGS INTOXICATED WITH ANTICOAGULANT RODENTICIDE. M. Senzolo¹, F. Gentilini², A. Zoia³, M. Caldin³. ¹University of Bologna/Clinica Veterinaria San Marco, PADOVA, Italy, ²University of Bologna, BOLOGNA, Italy, ³San Marco Veterinary Clinic, PADUA, Italy

Intoxication with anticoagulant rodenticide may cause haemorrhagic diathesis in dogs. No information is available on how long clotting times take to normalised after vitamin K treatment and literature suggest that intravenously (IV) vitamin K administration may cause anaphylaxis, but no references for this statement is provided. The aim of this study was to evaluate time to normalisation of activated partial thromboplastin time (aPPT) and prothrombin time (PT) after intravenously vitamin K treatment in dogs with naturally anticoagulant rodenticide (AR) intoxication. Inclusion criteria for AR intoxication were: detection of internal or external bleeding, prolonged coagulation times consistent with AR intoxication, exclusion of any other reasons for the clinical bleeding, a complete clinical record, and a coagulation profiles (PT, aPTT, antithrombin%, fibrinogen, FDPs and D-dimer concentration) collected at presentation (T0). Vitamin K was administered at 5 mg/kg intravenously, over 15 minutes, protected from light. Four hours and 8 hours post-vitamin K administration (T4 and T8) a coagulation profile was repeated. Seventy-three dogs with AR poisoning referred to the San Marco Veterinary Clinic met inclusion criteria. aPTT and PT values were compared at T0, T4 and T8 with "repeat measures analysis of variances" and "Friedman test", respectively. At T0 no dogs

had PT and aPTT within reference range (6.8-8.6 seconds, and 10.2-12.3 seconds respectively) and mean aPPT and median PT was 45.9 and 120 seconds respectively; at T4, mean aPPT and median PT was 14.5 and 8.5 seconds, respectively; and at T8 (these data were available only in 43 out of the 73 dogs) mean aPPT and median PT was 13.7 and 8.3 seconds respectively. There was a significant decreased in aPTT and PT between T0 and T4 and between T4 and T8 ($p < 0.0001$ for all comparison). All 73 dogs survived to discharge, none received blood or plasma transfusion or had an adverse reaction after vitamin K IV supplementation, and by T4 no dogs showed clinical signs of ongoing of external bleeding. This protocol with vitamin K administration seems to be safe and effective in treatment of dogs with naturally occurring AR intoxication.

Conflicts of interest: No conflicts of interest reported.

ESVIM-O-10
TRAPPED NEUTROPHIL SYNDROME: PRESENTATION AND OUTCOME IN 3 U.K BRED BORDER COLLIES. S.L. Mason¹, R. Jepson², M. Maltman³, D. Batchelor¹. ¹University of Liverpool, NESTON, United Kingdom, ²RVC, NORTH MYMMS, United Kingdom, ³Maltman Cosham Veterinary Clinic, HORSHAM, United Kingdom

Trapped neutrophil syndrome (TNS) is an autosomal recessive condition reported in Border collies and characterised by reduced circulating neutrophil numbers and intramedullary myeloid hyperplasia. Affected dogs present with fever, neutropenia and failure to thrive at a young age

TNS was originally identified in dogs from Australia and New Zealand and has been reported in Japan. There are no clinical reports of TNS in the United Kingdom, although there is an active screening programme and a reported carrier rate of 11.3%. Although the Border collie breed is known to be affected by cyclic neutropenia (Grey collie syndrome), veterinarians may not be familiar with TNS, for which management and prognosis are different. The objective of this study is to report the presentation and management of three unrelated dogs in the UK with confirmed TNS.

Three black and white Border collie puppies were presented for assessment of pyrexia and musculoskeletal signs. All three dogs were pyrexia on presentation and had at least one grossly swollen and painful joint. All were small in stature with facial dysmorphism typical of TNS (elongated and narrowed "ferret-like" facial features).

CBC revealed neutropenia in all (neutrophil count $1-2 \times 10^9/L$ (RI: 3.5-12.5)). Biochemistry was mostly normal. Serum cobalamin was normal. Bone marrow aspiration in two cases revealed expansion of the myeloid series. Arthrocentesis was consistent with immune-mediated polyarthritis. Genetic testing by PCR confirmed that the three puppies were homozygous for the mutation which causes TNS, and did not carry the mutation which causes cyclic neutropenia.

All dogs were initially treated with intravenous fluid therapy and antibiotics with no significant improvement and subsequently received prednisolone at immunosuppressive doses. Marked improvement in mentation and ambulation was observed within 24 hours in all three.

In case one prednisolone was tapered and withdrawn and the dog remains clinically stable 7 months after initial presentation. In case two prednisolone was tapered and discontinued after one year. The dog has remained clinically well for 21 months despite intermittent episodes of inappetence and pyrexia. In case three prednisolone and antibiotics were continued indefinitely and the patient survived for 39 months.

TNS should be a differential diagnosis for U.K. Border collies with neutropenia. This study supports the previous Australian study in describing successful medical management of this condition, although prognosis remains guarded and owners must be counselled regarding the requirement for on-going medication and occasional hospitalisation.

Conflicts of interest: The primary author's residency is partially funded by BSAVA Petsavers.

ESVIM-O-11

EFFECTS OF OMEPRAZOLE ON THE CANINE CEREBROSPINAL FLUID COMPOSITION. M. Girod¹, F.J.W. Allerton¹, K. Gommeren¹, A.C. Tutunaru¹, J. de Marchin², I. van Soens¹, D. Peeters¹. ¹University of Liege, LIEGE, Belgium, ²CHR citadelle, LIEGE, Belgium

The use of omeprazole, an inhibitor of K⁺-H⁺-ATPase, has been advocated in dogs to reduce cerebrospinal fluid (CSF) production in conditions where an accumulation of CSF may be associated with intracranial hypertension (hydrocephalus, syringomyelia). Two previous studies have shown a significant reduction in CSF production (26-35%) following ventriculocisternal or intravenous administration of omeprazole in dogs and rabbits. However, the underlying mechanism remains undetermined. Prior work in humans suggests that albumin quotient (QAlb: the ratio between CSF and serum albumin concentrations) is the best parameter to evaluate CSF flow, with decreasing CSF production resulting in an increased QAlb. The present study evaluated the impact of omeprazole administration on canine blood and CSF composition, including QAlb.

Fifteen healthy experimental beagle dogs received omeprazole (1.2 mg/kg/day) orally for 14 days. In each dog, 2.3 mL of CSF was collected from the cerebello-medullary cistern and 10 mL of blood was obtained from the jugular vein before and after treatment. Albumin concentrations were evaluated by immunoturbidimetric assay (gold standard in human medicine) and high-resolution protein electrophoresis (HRE) on paired CSF and serum samples, according to guidelines from the International Consensus Group for CSF Analysis. Erythrocyte and leukocyte counts, pH, lactate, glucose, total protein and electrolyte concentrations were also measured.

There were no significant differences in the composition of CSF before and after omeprazole administration except for a mild increase in sodium ($p = 0.002$). A strong linear correlation was found between albumin concentrations measured with immunoturbidimetry and HRE ($p < 0.0001$, correlation coefficient = 0.86). Regardless of methodology, QAlb did not change significantly ($p = 0.918$). Omeprazole induced a statistically significant increase in venous pH (7.43 ± 0.05 to 7.50 ± 0.04) ($p < 0.001$).

The lack of change in QAlb in the present study suggests that there is no change in the CSF flow following oral therapy with omeprazole, in contradiction with previous studies that detected a reduced CSF flow shortly after administration. Therefore, the present study provides no evidence of scientific benefit of long-term omeprazole treatment to reduce CSF production in dogs. Further research is warranted to explain these findings.

Conflicts of interest: No conflicts of interest reported.

ISCAID-O-1

NOSOCOMIAL VIRULENT, MULTI-DRUG-RESISTANT ACINETOBACTER BAUMANNII INFECTION OUTBREAK IN A VETERINARY HOSPITAL. S. Kuzi¹, N. Kahane¹, G. Segev¹, S. Bloom², I. Aroch¹. ¹Koret School of Veterinary Medicine, The Hebrew University of Jerusalem, REHOVOT, Israel, ²Kimron Veterinary Institute, Department of Bacteriology, Israel

Acinetobacter baumannii (*Acb*) is a Gram-negative, strictly aerobic nosocomial pathogenic coccobacillus, responsible for fatal pneumonia and other infections outbreaks in humans, with mortality rates of 30-75%. It is highly resistant to major antimicrobials, dry environment and disinfectants. *Acb* infections are increasingly reported in dogs and cats. This report describes a severe nosocomial outbreak of a multidrug resistant (MDR) *Acb* isolate, inducing a systemic, highly fatal illness, in a veterinary hospital that had occurred in several clusters (08/2011 to 11/2012). The affected 19 dogs and 4 cats were all microbiologically positive for this isolate. *Acb* was isolated from urine (9 animals), respiratory tract (11), tissues (3) and blood (1). The most common underlying primary diagnoses included seizures of various causes (4 animals), pneumonia, lung contusion, lymphoma, acute lung injury, feline idiopathic cystitis, heatstroke, urinary tract infection (UTI) and intervertebral disk herniation (2 each). The overall mortality rate was 70% (16/23 animals). *Acb* infection

played a major part in the mortality of 5 animals, and contributed to the mortality in 10. The infection-associated clinical and laboratory signs included fever, tracheal tube purulent discharge, hypotension, neutropenia, neutrophilia and neutrophilic cytoplasmic toxicity, which were absent previously during hospitalization. The *Acb* associated infections included pneumonia, UTI, cellulitis and sepsis. Most infected animals were hospitalized in the Intensive Care Unit (ICU), however, infections were identified in 5/23 animals in other wards, including one dog hospitalized in isolation. Mortality was unassociated with hospitalization length, urinary catheter placement, tracheal intubation or ventilation. Mortality rate tended to increase ($P = 0.09$) in respiratory *Acb* infections compared to other organs infection. The isolated *Acb* was resistant to numerous antibiotics, excluding amikacin, imipenem, ticarcillin and polymyxin-B. During the outbreak, an environmental investigation was performed, and samples from cages, rails, phones, sinks and ventilator were obtained, and yielded pure *Acb* cultures. These isolates had identical MDR profiles as determined in the *Acb* isolated from the infected animals. An infection control protocol was implemented, including aggressive disinfection of surfaces and equipment, and staff education regarding personal hygiene and antiseptics, which subsequently, led to a sharp decrease in the incidence MDR *Acb* infections in the hospital. Outbreaks of MDR *Acb* infections may be highly fatal and difficult to irradiate, warranting education for antiseptic techniques and judicious antibiotic use, especially in veterinary ICUs.

Conflicts of interest: No conflicts of interest reported.

ISCAID-O-2

ABDOMINAL CRYPTOCOCCOSIS: A RESTROSPECTIVE EVALUATION OF 25 CASES FROM AUSTRALIA. A. Tebb¹, A. Kessel², S. Mcgill³, T. King⁴, R. Malik⁵. ¹Murdoch University, MURDOCH, PERTH, Australia, ²Charles Sturt University, WAGGA WAGGA, Australia, ³Small Animal Specialist Hospital, SYDNEY, Australia, ⁴Veterinary Specialist Services, BRISBANE, Australia, ⁵University of Sydney, SYDNEY, Australia

Cryptococcosis affects cats and dogs worldwide. It has a predilection for the respiratory tract and is four-times more common in cats than dogs. Abdominal cryptococcosis is rare in cats, dogs and people, but more common in ferrets and horses. Databases from four referral centres and three veterinary laboratories were searched (2000-2012) for cryptococcosis primarily affecting intra-abdominal structures. Signalment, clinical findings, antigen titres, culture, treatment and outcomes were reviewed. Twenty-five cases (24 dogs, one cat) met the inclusion criteria. Median age and weight at diagnosis for the dogs was 2 years (range:8 months-8 years) and 20.9 kg (range:8-57 kg), respectively. Breeds included Staffordshire Bull Terriers (4), Bulldogs (3), seven other purebreds and 10 crossbreds, with 11 males (5 castrated) and 13 females (9 spayed). The cat was a 4-year-old male neutered Domestic Shorthair. The majority of cases were from Western Australia (20), four were from Queensland and one each from Victoria, New South Wales and South Australia. Presenting complaints included inappetence (8), lethargy (13), vomiting (15), hematemesis (1), diarrhoea (5), melaena (3), dyschezia (1), weight loss (8) and a mass noted by the owner (3). Physical findings included fever (1), abdominal distension (2) or pain (5), abdominal mass (11) or effusion (1) and ataxia/back pain (2). Cryptococcal lesions involved the stomach (1), small (13) or large (1) bowel, ileocaecocolic junction (1), abdominal cavity/mesentery (18), kidney (1), liver (1) and pancreas (2). Eight cases had thoracic imaging, three with lesions. The antigen titres ranged from 1:2 to 1:65,536 (median 2048) in 11 dogs tested. Culture was attempted in eight patients, yielding *C.gattii* (4) and *C.neoformans* (3); one case failed to grow *in vitro*. Untreated cases were euthanased after diagnosis. Treatment was attempted in 14 patients, consisting of complete surgical excision (2), surgical excision or de-bulking plus anti-fungal therapy (5) and anti-fungal therapy alone (7). Median survival for all treated patients was 805 days (range:2-2737). Survival of the two cases treated with surgery alone was 40 and 2737 days. Median survival for medically treated cases

was 103 days (range:2-1460) and for combined therapy was 1029 days (range:132- 1984). Abdominal cryptococcosis is a life-threatening infection which is much more common in dogs than cats. Successful therapy requires aggressive management. We hypothesise that the scavenging nature of dogs predisposes to transient intestinal colonisation with mucosal injury permitting invasion into the gut wall, with subsequent spread to contiguous structures and/or dissemination via lymphatic or portal venous pathways.

Conflicts of interest: No conflicts of interest reported.

ISCAID-O-3

SERUM ACUTE PHASE PROTEINS CONCENTRATIONS IN DOGS WITH SPIROCERCOSIS AND THEIR ASSOCIATION WITH ESOPHAGEAL NEOPLASIA: A PROSPECTIVE OBSERVATIONAL STUDY. R. Nivy¹, E. Lavy¹, M. Caldin², K. Shaabon¹, G. Segev¹, I. Aroch¹. ¹Hebrew University Veterinary Teaching Hospital, REHOVOT, Israel, ²Clinica San Marco, PADOA, Italy

Spirocerca lupi infection in dogs typically results in esophageal nodules. Malignant esophageal nodule transformation to sarcoma occurs in up to 26% of the cases. Definitive ante mortem discrimination between benign and malignant esophageal masses is difficult, requiring endoscopy and histopathology of esophageal biopsies, or post-mortem microscopic examination of esophageal specimens. Serum acute phase proteins (APPs) concentrations in dogs change in response to inflammation and are utilized as diagnostic and prognostic markers in various canine diseases. This study aimed to characterize serum APPs concentrations in dogs with esophageal spirocercosis and to investigate their discriminatory performance between malignant and benign esophageal nodules. The study included 78 client-owned dogs with esophageal spirocercosis, of which 23 re-presented during therapy. Serum C-reactive protein (CRP), haptoglobin (Hp), serum-amyloid A (SAA) and albumin concentrations were measured at presentation and at follow-up visits, and were compared with those of healthy dogs, and between malignant and benign esophageal spirocercosis cases. Receiver operator characteristics (ROC) analyses were used to assess the diagnostic utility of APPs and the white blood cell count (WBC) in discriminating malignant from benign cases. Serum Hp, CRP and SAA concentrations at presentation and at follow-up visits were significantly higher, and albumin concentration was significantly lower in infected dogs compared to their reference intervals (RIs) ($P < 0.001$ for all), but did not change significantly during anthelmintic therapy. Compared to benign cases, neoplastic cases had significantly higher levels of CRP ($P = 0.011$), Hp ($P = 0.008$) and SAA ($P = 0.05$), while albumin concentration was significantly lower ($P = 0.012$). However, serum concentrations of these APPs and the leukocyte count only moderately discriminated between suspected neoplastic and benign esophageal nodules (area under the ROC curve, 0.70, 0.69, 0.71, 0.68 and 0.74, respectively). Nevertheless, there were no neoplastic cases with concentrations of all four APPs within reference interval concurrently. In conclusion, canine esophageal spirocercosis is characterized by an acute phase reaction, at presentation and during anthelmintic therapy, but they are not useful to monitor response during treatment. When the concentrations of all four are within RI, esophageal malignancy can be ruled out. Although concentrations of all positive APPs were significantly higher in neoplastic compared to benign cases, moderate discriminatory power precludes their individual clinical use in spirocercosis.

Conflicts of interest: No conflicts of interest reported.

ISCAID-O-4

PREVALENCE OF LEPTOSPIRA URINARY SHEDDING IN HEALTHY DOGS FROM SOUTHERN GERMANY. J.R. Llewellyn¹, I. Krupka-Dyachenko², A.L. Rettinger², V. Dyachenko², T. Stamm³, P.A. Kopp³, R.K. Straubinger², K. Hartmann¹. ¹Clinic of Small Animal Internal Medicine Ludwig-Maximilians-University, MUNICH, Germany, ²Bacteriology and Mycology, Institute for Infectious Diseases and Zoonoses, MUNICH, Germany, ³Vet Med Labor GmbH Division of IDEXX Laboratories, LUDWIGSBURG, Germany

Leptospirosis is a re-emerging zoonotic disease among humans and dogs worldwide, but the role of healthy dogs as source of infections is unknown. Thus, the aim of this study was to evaluate the number of healthy dogs that shed leptospires in their urine and to identify the genomospecies of those leptospires. As a second aim, antibody prevalence was evaluated.

Urine samples of 200 healthy dogs from Southern Germany that were prospectively included in the study were evaluated by a quantitative real-time polymerase chain reaction (PCR) specific for the *lipL32* gene of *Leptospira* (*L.*) spp. Positive samples were tested *via* multilocus sequence typing (MLST) to identify the genomospecies. A microscopic agglutination test (MAT) was performed to determine antibody titers.

Three urine samples were PCR-positive for leptospires, indicating a prevalence of urinary shedding of 1.5% (95% confidence interval 0.3%-4.5%). All three dogs were vaccinated with "old" vaccines containing only two serovars. One dog shed leptospires of the genomospecies *L. interrogans*, and two from the genomospecies *L. borgpetersenii*. Of all dogs, 17.0% (95% confidence interval 12.3%-22.8%) had antibody titers $\geq 1:100$ to non-vaccinal serogroups, and 3.5% (95% confidence interval 1.5%-7.1%) had titers $\geq 1:400$.

As 1.5% of healthy dogs in Southern Germany shed leptospires of pathogenic genomospecies, dogs represent a potential risk for their owners and other animals. The study emphasizes the importance of using vaccinations that contain a broader range of serovars and do not only protect against disease, but also prevent shedding.

Conflicts of interest: No conflicts of interest reported.

ISCAID-O-5

PREVALENCE OF ANTIBODIES AGAINST FELINE PANLEUKOPENIA VIRUS IN CLIENT-OWNED CATS IN SOUTHERN GERMANY. K. Mende¹, B. Stuetzer¹, R.S. Mueller¹, C. Sauter-Louis², T. Homeier-Bachmann³, U. Truyen³, K. Hartmann². ¹Ludwig-Maximilians-Universitaet/Clinic of Small Animal Medicine, MUNICH, Germany, ²Ludwig-Maximilians-Universitaet, Munich/Clinic of Ruminants and Herd Management, MUNICH, Germany, ³University of Leipzig/Institute of Animal Hygiene and Veterinary Public Health, LEIPZIG, Germany

Feline panleukopenia (FPV) is a frequent and commonly fatal disease in cats. Recently published studies raise suspicion that some cats fail to develop antibodies after vaccination. The purpose of this study was to assess the prevalence of antibodies against FPV in cats in Southern Germany, and to identify factors that are associated with a lack of antibodies.

Three hundred fifty cats in which blood was taken for various unrelated reasons (e.g., health check, test for feline retrovirus infections, different clinical signs) were randomly included in the study. Prior power analysis had determined a minimum number of 323 cats to be included. Information regarding signalement, origin, environment, lifestyle, housing conditions, health status, chronic diseases, glucocorticoid therapy, and vaccination status were collected. Antibodies were detected by hemagglutination inhibition test. Asymptomatic Chi-square tests and univariable logistic regression were used to investigate associations between lack of antibodies and parameters obtained from signalement and history. Parameters determined to be statistically significant (with a p-value < 0.1) were verified by a multivariable logistic regression analysis.

One hundred and three of 350 cats (29.4%) had no antibodies, and even in 23.4% of properly vaccinated cats, no antibodies

were detected. Chronic kidney disease, neoplasia, glucocorticoid therapy, and vaccination status were significantly associated with a lack of antibodies.

In this study, nearly one third of the cats had no antibodies and was likely to have inadequate immunity against panleukopenia. Cats with chronic diseases and cats that receive glucocorticoids were protected less frequently.

Conflicts of interest: No conflicts of interest reported.

ISCAID-O-6
RISK FACTORS OF WEANING DIARRHEA IN PUPPIES FROM FRENCH BREEDING KENNELS. A. Grellet¹, C. Robin², A. Feugier¹, C. Boogaerts², C. Boucraut-Baralon³, D. Grandjean², B. Polack², S. Chastant-Maillard⁴. ¹Royal Canin, AIMARGUES, France, ²Université Paris-Est, Ecole Nationale Vétérinaire d'Alfort, MAISONS-ALFORT, France, ³Scanelis, TOULOUSE, France, ⁴Unité Toulousaine d'Élevage et Reproduction (UTER), UMR INRA/ENVT 1225 IHAP, TOULOUSE, France

Introduction: Puppies weaning diarrhea is a frequent disease, with deleterious consequences on growth [1]. Recently a fecal scoring scale for puppies was developed, providing an objective definition of an abnormal fecal score [1]. Rather than due to one pathogen, weaning diarrhea results from interaction between infectious agents, host immunity, and management procedures. The aim of this study was to identify risk factors for pathological feces in puppies in French breeding kennels.

Materials and methods: 33 French breeding kennels were included. For each kennel, data on environmental factors (size of the kennel and number of puppies in the litter), management of the kennel and puppies (number of meals per day, cleaning and disinfection procedures, access to outdoor, number of persons dedicated to the puppies) and puppies' characteristics (age, breed, sex) were collected. Stool quality was scored for each puppy [1]. Parasites excretion was evaluated by the McMaster flotation technique. The presence of copro-antigens of *Giardia intestinalis*, and *Cryptosporidium parvum* was evaluated by the ProSpecT-Giardia/Cryptosporidium Microplate Assay (Remel, Lenexa, KS, USA). Canine parvovirus type 2 (CPV2) and canine coronavirus (CCV) excretions were evaluated respectively by qPCR and qRT-PCR. Several separate univariate and three binary logistic regression models (SAS software) were used to evaluate factors influencing feces quality.

Results: 307 puppies between 5 and 14 weeks of age were included. 46.9% (144/307) of the puppies were housed in breeding kennels producing more than 30 puppies per year. The prevalence of enteropathogens was: 40.9% (124/303) for *Giardia sp.*, 37% (111/302) for CCV, 29% (87/301) for CPV2, 28.2% (84/298) for *Isospora ohioensis* complex, 25.7% (78/304) for *C. parvum*, 14.1% (42/298) for *Toxocara canis*, and 12.1% (36/298) for *Isospora canis*. 82 out of 307 feces evaluated (26.7%) were classified as abnormal. Among enteropathogens, CPV2 was the unique pathogen associated to a pathological fecal score. Among environmental factors, the number of puppies produced per year in the kennel and the number of meals per day were the only two factors having an impact on feces quality. In the final model including enteropathogens and environmental factors, CPV2 was the unique factor with a significant impact.

Conclusion: This study underlines the high prevalence of enteropathogens in puppies. CPV2 circulation needs to be suspected in case of weaning diarrhea even in absence of typical clinical signs or mortality.

[1] Grellet A et al. Prev. Vet. Med. 106 (2012) 315-323

Conflicts of interest: Employed by Royal Canin.

ISCAID-O-7
EVALUATION OF CONJUNCTIVAL SWAB POLYMERASE CHAIN REACTION FOR DETECTION OF LEISHMANIA INFANTUM IN DOGS IN A NON-ENDEMIC AREA. K. Geisweid, K. Weber, C. Sauter-Louis, K. Hartmann. LMU University of Munich, MUNICH, Germany

Due to increasing numbers of dogs that are imported from or have visited Mediterranean countries, canine leishmaniasis has become an important infectious disease in Germany. Although conjunctival swabs have recently been described as useful diagnostic tool in endemic areas, their value in non-endemic areas is unknown.

The aim of this study was to evaluate sensitivity and specificity of conjunctival swab polymerase chain reaction (PCR) in dogs in a non-endemic area. Seventy four dogs that were presented for suspected canine leishmaniasis or for screening purposes after travelling to endemic countries were prospectively included. PCR results from conjunctival swabs were compared to those from bone marrow, lymph node, blood and to antibody results determined by immunofluorescence assay or enzyme-linked immunosorbent assay. Dogs were considered infected if bone marrow and/or lymph node and/or blood PCR was positive and were defined as not infected if bone marrow PCR (considered the test with the highest sensitivity) was negative.

Sensitivity and specificity of conjunctival swab PCR were 78.4% (CI 95% 62.8-88.6) and 93.8% (CI 95% 79.8-98.3). There was substantial agreement between PCR results from conjunctival swabs and lymph nodes ($\kappa = 0.642$), fair to moderate agreement between conjunctival swab and bone marrow ($\kappa = 0.483$) and almost no agreement between conjunctival swab and blood ($\kappa = 0.070$).

Conjunctival swab PCR is a good non-invasive test to diagnose Leishmania infection in dogs in non-endemic countries.

Conflicts of interest: No conflicts of interest reported.

ISCAID-O-8
EVALUATION OF AN IN-HOUSE DOT ENZYME-LINKED IMMUNOSORBENT ASSAY TO DETECT ANTIBODIES AGAINST FELINE PANLEUKOPENIA VIRUS. K. Mende¹, B. Stuetzer¹, U. Truyen², K. Hartmann³. ¹Ludwig-Maximilians-Universitaet/Clinic of Small Animal Medicine, MUNICH, Germany, ²University of Leipzig/Institute of Animal Hygiene and Veterinary Public Health, LEIPZIG, Germany, ³Ludwig-Maximilians-Universitaet, Munich/Clinic of Ruminants and Herd Management, MUNICH, Germany

Routine titer testing to determine the specific immune status instead of regular vaccination has not been established in most European countries so far. An in-house test for detection of antibodies against feline panleukopenia virus (FPV), the ImmunoComb[®] Feline VacciCheck, is now available in several European countries. The aim of this study was to evaluate the performance of the in-house test ImmunoComb[®] Feline VacciCheck in comparison with the gold standard hemagglutination inhibition (HI).

Serum samples from randomly selected cats (n = 347) that were obtained for other unrelated reasons were included. HI was used as gold standard for antibody detection. Sensitivity, specificity, positive and negative predictive value, and overall accuracy of the ImmunoComb[®] Feline VacciCheck were assessed. Cohen's kappa was used to assess agreement between different methods.

Prevalence of antibodies detected by HI was 64.8%. The ImmunoComb[®] Feline VacciCheck showed a sensitivity of 86.7% and a specificity of 81.1%. Based on an antibody prevalence of 64.8%, the positive predictive value was 89.4% and the negative predictive value was 76.7%. The overall accuracy of the test was 84.7%. Kappa (κ) was 0.633 (standard error (SE) 0.045, 95% confidence interval (CI) 0.545 - 0.721).

For the intended use of the test to replace regular booster vaccination in veterinary practice, a low number of false positive test results is important. Thus, the specificity of the test in the present study was not good enough and, therefore, the test cannot be fully recommended to replace vaccination, especially not for cats at risk.

Conflicts of interest: Biogal Galed Laboratories, Kibbutz Galed, Israel sold the test at a reduced price for this study.

ISCAID-O-9

THE USE OF ORAL RECOMBINANT FELINE INTERFERON-OMEGA IN NATURALLY FELINE IMMUNODEFICIENCY VIRUS INFECTED CATS: NEW INSIGHTS INTO AN ALTERNATIVE IMMUNOMODULATION THERAPY. R.A.O. Leal¹, S. Gil¹, D. McGahie², N. Sepúlveda³, A. Duarte¹, M.M.R.E. Niza¹, L. Tavares¹. ¹CIISA/Faculty of Veterinary Medicine - Technical University of Lisbon, LISBON, Portugal, ²Virbac, FRANCE, Portugal, ³London School of Hygiene and Tropical Medicine/Centro de Estatística Univ Lisboa, LONDON, United Kingdom

Recombinant-Feline Interferon-Omega (rFeIFN- ω) is an immunomodulator drug often used in feline medicine. Although alternative oral trials have been successfully applied in some viral infections, only the licensed protocol has been recommended to Feline Immunodeficiency Virus (FIV) infected cats. This protocol has been shown to improve clinical signs, reduce concurrent viral excretion and increase levels of Acute Phase Proteins (APP). Despite these effects, its cost can be limiting and alternative protocols are required.

This study aimed to evaluate the clinical improvement of naturally FIV-infected cats treated with an oral rFeIFN- ω protocol (0.1MU/cat per os daily for 90 days) in comparison to the licensed one (3 cycles of 5 subcutaneous daily injections at Day (D) 0, 14 and 60).

11 FIV naturally-infected cats were treated with oral rFeIFN- ω protocol (PO Group). 6 cats were indoor single-housed animals and 5 lived in a multi-cat/outdoor environment. Clinical signs were monitored at D0 (before therapy), 10, 30, 65 and 90 (end of therapy) using a previously validated score scale which included the most relevant signs for FIV infection. Results were compared to previous clinical data of 7 naturally FIV-infected cats, living in an animal shelter and treated with the licensed rFeIFN- ω protocol (SC Group). According to EMEA-guidelines, this group was considered an external positive control. For both groups, clinical improvement was classified as 'marked' (> 50% improvement of the initial score), 'mild' (up to 50% improvement), 'stable' (same final and initial score) or 'worse' (final score higher than the initial).

There was no difference between groups in the proportion of cats showing improvement (Pearson's Chi-square test adjusted for small samples; $p = 0.95$) or in the grade of clinical improvement (Pearson's Chi-square test; $p = 0.23$). In detail, 9/11 (82%) cats improved their overall scores with oral rFeIFN- ω therapy. Specifically: 3/11 (27%) showed a marked improvement, 6/11 (55%) a mild improvement and 2/11 (18%) remained stable. In the SC Group, 5/7 (71%) cats improved their overall score. In particular: 4/7 (57%) had marked improvement, 1/7 (14%) mild improvement and 2/7 (29%) remained stable. No worsening was observed in both groups.

Independently of the protocol applied, this study showed that rFeIFN- ω induced an overall significant clinical improvement of treated cats, supporting a potential immune-stimulation. Although the licensed protocol is better recommended in more symptomatic animals, in cases where cost might be an issue, oral rFeIFN- ω may be considered as an alternative therapy in the management of FIV-infected cats.

Conflicts of interest: McGahie, D: employee Virbac, Carros, France. His contribution for this work was mainly as a Consultant.

This work was supported by the project CIISA 10.

Rodolfo Leal is a PhD fellow (FCT SFRH / BD / 62917 / 2009 Portugal); Solange Gil is a research assistant under Programa Ciência 2007 (FCT-Portugal).

ISCAID-O-10

EFFICACY OF LOCAL APPLICATION OF FELINE INTERFERON-OMEGA IN CATS WITH ACUTE VIRAL UPPER RESPIRATORY TRACT DISEASE. A. Knebl¹, B. Schulz¹, C. Helps², C. Sauter-Louis³, R. Mueller¹, K. Hartmann¹. ¹Clinic of Small Animal Medicine, LMU University of Munich, MUNICH, Germany, ²Department of Clinical Veterinary Science, University of Bristol, BRISTOL, United Kingdom, ³Clinic for Ruminants, LMU University of Munich, OBERSCHLEISSHEIM, Germany

Feline interferon omega (feIFN- ω) is used in the treatment of acute virus-induced feline upper respiratory tract disease (FURTD), despite lack of controlled studies confirming its efficacy. This study aimed to evaluate, whether local feIFN- ω improves clinical signs in cats with acute FURTD caused by feline calicivirus (FCV) or feline herpesvirus 1 (FHV-1) and reduces the number of cats shedding FCV over a 42 day period.

The study was designed as a prospective, randomized, placebo-controlled, double-blinded clinical trial including 37 cats with acute FCV and/or FHV-1 infection and clinical signs of FURTD. Presence of FCV and FHV-1 was determined by quantitative polymerase chain reaction of oropharyngeal and conjunctival swabs. Cats were assigned to either treatment with feIFN- ω (2.5 MU/kg SC once, followed by 0.5 MU locally q8 h, divided conjunctivally, intranasally, and orally) or placebo for 21 days. Cats of both groups received standard treatment in addition. Clinical signs and FCV shedding were evaluated over 42 days.

Cats of both groups responded rapidly with a significant improvement in general health status, nasal and ocular discharge. Clinical and laboratory variables did not differ among groups. Intragroup analysis of changes over time within the feIFN- ω group revealed a significant decrease of relative FCV copy numbers by day 21, and an improvement of oromucosal ulcerations on day 42. These differences were not present in the placebo group.

Local administration of feIFN- ω is not an effective treatment for cats with acute viral FURTD, but might lead to improvement of chronic oral changes through immunomodulatory mechanisms.

Conflicts of interest: The study was partially supported by Virbac Tierarzneimittel GmbH, Bad Oldesloe, Germany.

ESVC-O-1

ASSOCIATION OF LOW OR NORMAL WHOLE BLOOD TAURINE CONCENTRATION WITH OUTCOME IN IRISH WOLFHOUNDS WITHOUT EVIDENCE OF DCM INITIALLY. C. Vollmar¹, P. Fox². ¹Small Animal Vet. Clinic, BONN, Germany, ²The Animal Medical Center, NEW YORK, United States of America

Whole blood taurine (WBT) concentration was measured in 115 IW dogs that received cardiovascular examination including echocardiograms.

Results: 49 (42.6%) had DCM while 66 (57.4%) had no detectable heart disease. WBT concentration was: severely reduced (<130 nmol/mL) in 4 dogs with DCM and in 4 without heart disease; moderately low (130-179.9 nmol/mL) in 12 of 49 with DCM (41.3%) and 20 of 66 (30.3%) without heart disease. At time of blood collection, prevalence of WBT deficiency was similar in IWs with and without DCM.

Follow up information was available for 46 of the 66 IW dogs without DCM at time of blood sample collection for WBT analysis. Ten of these 46 dogs (21.7%) followed up to 7 years, developed DCM between 6 months and 7 years after initial examination. Of the four with severely reduced WBT, one developed DCM with AF 3 years later; 2 had no evidence of DCM (one was healthy 5 years later; the other died of non-cardiac disease 2 years later); the fourth dog was lost to follow. Of the 20 dogs with moderately reduced WBT, 2 developed AF without DCM; 5 developed DCM; 2 died of non-cardiac death 2 and 3 years later; 5 had normal hearts; and 6 were lost to follow. Of the 12 dogs with marginal WBT, 4 developed DCM and AF; 3 had normal hearts; and 5 were lost to follow. Out of the 30 dogs with normal WBT, 1 developed AF without DCM; 15 had normal hearts, 6 died of non-cardiac death 2 to 4 years later; and 8 were lost to follow. The proportion of these 46 dogs which

had developed DCM or AF at subsequent follow-up was compared with the proportion with normal hearts in two separate categories. When all dogs which had marginal, moderate, or severe WBT deficiency combined were compared to all dogs having normal WBT, the proportion that developed DCM or AF was significantly higher in the low WBT cohort ($P < 0.001$). When all dogs which had moderate-severe WBT deficiency were compared with all dogs with marginal or normal WBT combined, the proportion that developed DCM or AF was significantly higher in the moderate-severe WBT deficiency cohort ($P = 0.044$). None of 22 dogs with normal WBT developed DCM; but one developed AF.

Conclusion: Further study is required to investigate potential relationships between prolonged taurine deficiency, myocardial dysfunction, and predisposition to develop DCM in the IW breed.

Conflicts of interest: Taurine measurements had been carried out by the laboratory from Royal Canin company.

ESVC-O-2

PATHOLOGY OF DCM IN GREAT DANES. H. Aupperle¹, I. März², K. Baldauf², N. Roggon³, J.G. Kresken², ¹Laboklin GmbH & Co.KG, BAD KISSINGEN, Germany, ²Klinik für Kleintiere der Universität Leipzig, LEIPZIG, Germany, ³Tierärztliche Klinik für Kleintiere am Kaiserberg, DUISBURG, Germany

Introduction: Dilative Cardiomyopathy (DCM) is characterized by systolic dysfunction as well as ventricular and often atrial dilation.

Pathological findings are mainly described for the DCM in the Doberman Pinscher. Typical histological findings include the attenuated wavy fiber type (wType) or the fatty infiltration degenerative type (fType).

Reports about pathological findings in dogs of giant breeds like Great Danes concerning heart diseases are rarely to find.

Material and methods: Hearts of 13 Great Danes (0.5-9.0 years old) were investigated clinically, on gross pathology and histopathologically using HE and Picrosirius red stain.

Results: Six dogs without heart disease but suffering from neoplastic diseases or pachymeningitis ossificans served as controls.

Seven dogs with diagnosed DCM were in congestive heart failure and had atrial fibrillation. Echocardiographic investigation of these dogs showed that the diameter of left ventricle in systole (DCM 7.2 ± 0.8 cm vs. controls 3.3 ± 0.5 cm, $P = 0.001$) and diastole (DCM 7.9 ± 1.07 cm vs. controls 4.8 ± 0.46 cm, $P = 0.001$) as well as the Cornell-index (DCM 2.28 ± 0.31 vs. controls 1.41 ± 0.2 $P = 0.001$) were significantly increased compared to the controls.

Gross investigations of hearts with DCM showed a moderately dilated left ventricle with flattened papillary muscles. In contrast to echocardiography, gross investigations detected significantly thinned left ventricular wall in DCM group (DCM 1.4 ± 0.31 cm vs. controls 1.9 ± 0.25 cm, $P = 0.016$). Relative heart weight was not different between the groups (DCM $0.74 \pm 0.09\%$ vs. controls $0.64 \pm 0.08\%$, $P = 0.75$).

Histopathological findings were not uniform. In one dog, similar tissue changes as in the Doberman Pinschers wType morphology ($n = 1$) were diagnosed. In three dogs a mixture of wType and fType with mainly fibrotic lesions was seen. Moderate to marked replacement fibrosis was present in the right ventricular myocardium ($n = 1$) or in both ventricles ($n = 1$). One Great Dane showed clinically typical phenotype of DCM but clinical symptoms did not aggravate during the next three years. When the dog died from gastric dilatation-volvulus syndrome, pathological investigation showed marked left ventricular fibrosis, thus diagnosis of DCM is questionable from retrospective view.

Conclusion: In conclusion these interdisciplinary investigations showed that in Great Danes the typical clinical and gross findings of DCM are correlated with varying histopathological alteration. Histological findings in Great Danes are different from those in Doberman Pinscher DCM. Beside the two known major pathological changes in DCM, extended myocardial fibrosis seems to be the predominant alteration in Great Danes.

Conflicts of interest: No conflicts of interest reported.

ESVC-O-4

24-HOUR AMBULATORY ELECTROCARDIOGRAPHIC FINDINGS IN ENGLISH BULLDOGS WITH MYOCARDIAL DISORDERS. R.A.S. Santilli¹, S.B. Bassi², M.P. Perego¹, ¹Clinica Veterinaria Malpensa, SAMARATE, Italy, ²Ospedale Veterinario I Portoni Rossi, ZOLA PREDOSA, Italy

English bulldog (EB) can be affected by a segmental form of arrhythmogenic right ventricular cardiomyopathy (ARVC) with a characteristic dilation of the right ventricular outflow tract and monomorphic ventricular tachycardia with a left bundle branch block morphology.

The aim of this study was to evaluate arrhythmias prevalence during ambulatory electrocardiography in EB with myocardial disorders compared to normal dogs of the same breed. Medical records of 136 EB referred for cardiac consultation were retrospectively analyzed, and dogs with congenital heart diseases, advanced atrioventricular block and accessory pathway were excluded. Forty-five dogs, 35 males and 10 females with a mean age of 83.3 ± 38.2 (SD) months and a mean body weight of $26.5 \text{ kg} \pm 5.8$, were included in the study. According to instrumental findings at admission, the dogs were divided into two groups: normal dogs (10) and dogs with primary myocardial disorders (35), 14 of which with signs of congestion. Normal distribution of values was assessed by the Shapiro-Wilk W-test. Normally distributed data were tested using Student's t-test and non-normally distributed data using Wilcoxon's sum rank test. Multivariate analysis was performed with GLM and non-parametric ANOVA to test the influence of each variable and of congestive cardiac failure on the results. Dogs with cardiomyopathy resulted older 97.5 months (CI 95% 88-107.1) vs. 33.3 months (CI 95% 14.9-51.7) ($p = 0.0007$), with a higher count of total beats 175393 (CI 95% 156625-194261) vs. 135230.4 (CI 95% 112238-158223) ($P = 0.0175$), ectopic beats 6900.7 (CI 95% 2518.7-11282.7) vs. 1.9 (CI 95% 0.3-3.5) ($p < 0.001$), a higher average ventricular rate 132.2 bpm (CI 95% 121.3-143.2) vs. 104.5 bpm (CI 95% 74-135) ($p = 0.0405$), a longer supraventricular tachycardia 643.2 (CI 95% 0-1357.4) vs. 671 (CI 95% 147.7-1194.3) ($p = 0.0348$), a lower count of ventricular pause 0.6 (CI 95% 0.2-1) vs. 38 (CI 95% 0 - 109.2) ($p = 0.0131$) and of sinus bradycardia 30.7 (CI 95% 0-79.1) vs. 314.5 (CI 95% 0 -748.8) ($p = 0.0319$). Multivariate analysis showed a trend ($p = 0.06$) of higher VPCs count, ventricular tachycardia runs and allorhythmic forms in dogs with signs of congestion. Similarly to Boxer and human beings with ARVC, also EB can be affected by a cardiomyopathy characterized by ventricular and supraventricular arrhythmias and high sympathetic tone. Further studies, with larger sample, are needed to evaluate the influence of different factors and to expand our results to the general population.

Conflicts of interest: No conflicts of interest reported.

ESVC-O-5

INDICES OF MYOCARDIAL STRAIN AND STRAIN RATE BY TWO-DIMENSIONAL SPECKLE TRACKING ECHOCARDIOGRAPHY IN GREAT DANES. B. Pedro, H. Stephenson, C. Linney, J. Dukes-McEwan. Small Animal Teaching Hospital, University of Liverpool, CHESHIRE, United Kingdom

Dilated cardiomyopathy (DCM) is an adult-onset disease with high prevalence in Great Danes (GD). It has a long occult phase requiring serial echocardiography to detect dogs at risk. Strain (St) and strain rate (SR) imaging based on two-dimensional speckle tracking (STE) methods can assess myocardial function. Its advantage over Tissue Doppler assessment is its independence of translational movement and insonation angle. Values for St and SR vary between dogs of different size, therefore breed-specific reference intervals are required.

Between 2008 and 2012, 127 GD were assessed echocardiographically. Images obtained underwent analysis of radial and circumferential St and SR by STE. Dogs were classified as normal (NORM), equivocal (EQUIV) or affected (AFX). Forty-five dogs had at least 2 echocardiographic examinations. Dogs with other significant congenital or acquired, cardiac or systemic diseases were excluded, as were EQUIV dogs for the purpose of the

present analysis. Echocardiographic studies with unsatisfactory image quality were also excluded.

Forty-two GD were NORM and thirty-eight AFX. NORM group consisted of fifteen males and twenty-seven females; mean age: 76.9 months (48-138), mean weight: 64.4 kg (50.6-87.5). AFX group consisted of twenty-three males and fifteen females; mean age: 76.4 months (48-141), mean weight: 64.6 kg (52.4-88.0). Fifteen NORM dogs had repeated echocardiography.

Radial St(RSt) and SR(RSR) were obtained from right parasternal short axis view at papillary muscles level for 21/42NORM dogs and 6/38AFX dogs. Circumferential St and SR were obtained from right parasternal short axis view at the level of mitral valve (MVCSt and MVCSR, respectively) and apex (ApCSt and ApCSR, respectively) in 35/42NORM and 14/38AFX dogs. Segmental values were averaged to obtain global St and SR.

Results (mean±standard deviation) for NORM and AFX groups, respectively, for the different parameters were: RSt $45.79 \pm 10.51\%$ and $32.65 \pm 12.48\%$ ($p = 0.051$); RSR systolic $2.61 \pm 0.80s^{-1}$ and $2.02 \pm 0.54s^{-1}$ ($p = 0.058$), early diastolic $-1.81 \pm 0.78s^{-1}$ and $-1.57 \pm 0.40s^{-1}$ ($p = 0.309$), late diastolic $-1.64 \pm 0.57s^{-1}$ and $-1.09 \pm 0.40s^{-1}$ ($p = 0.021$); MVCSt $-14.84 \pm 3.21\%$ and $-12.56 \pm 3.33\%$ ($p = 0.039$); MVCSR systolic $-2.01 \pm 0.46s^{-1}$ and $-1.69 \pm 0.43s^{-1}$ ($p = 0.025$), early diastolic $1.51 \pm 0.38s^{-1}$ and $1.22 \pm 0.50s^{-1}$ ($p = 0.061$), late diastolic $1.18 \pm 0.48s^{-1}$ and $0.87 \pm 0.46s^{-1}$ ($p = 0.047$); ApCSt $-21.77 \pm 5.90\%$ and $-19.09 \pm 4.31\%$ ($p = 0.087$); ApCSR systolic $-2.60 \pm 1.28s^{-1}$ and $-1.97 \pm 0.60s^{-1}$ ($p = 0.023$), early diastolic $2.58 \pm 0.87s^{-1}$ and $2.21 \pm 0.82s^{-1}$ ($p = 0.167$), late diastolic $1.54 \pm 0.66s^{-1}$ and $0.87 \pm 0.46s^{-1}$ ($p = 0.198$).

This study demonstrates that STE assessment of radial and circumferential St and SR is feasible in GD. CSt and CSR appear to be most useful in differentiating NORM and AFX dogs. Normal reference intervals in GD may allow early identification of systolic dysfunction in occult DCM or EQUIV dogs.

Conflicts of interest: No conflicts of interest reported.

ESVC-O-6 ECHOCARDIOGRAPHIC FINDINGS IN 246 ADULT CATS WITH HEART DISEASE IN GENERAL PRACTICE. M.J.M. Dirven. Dierenkliniek Rijen, RIJEN, The Netherlands

Most studies concerning cats with heart disease relate to referral populations. Characterization of feline heart disease in general practice remains ill-defined. Echocardiography is considered the method of choice to diagnose feline heart disease and echocardiographic evidence of left atrial enlargement is considered a poor prognostic variable. The aims of the study were to assess the relative frequencies of different forms of heart disease encountered in general practice and to describe the presence of left atrial enlargement in different heart diseases.

Case records of cats that underwent an echocardiographic examination in Dierenkliniek Rijen between May 2009 and December 2012 were reviewed retrospectively ($n = 818$). Referral cases and screenings ($n = 43$), follow-up examinations ($n = 122$) and examinations of kittens ($n = 217$) were excluded. In 436 cases heart disease was suspected based on the presence of a heart murmur, gallop or arrhythmia and/or dyspnea, syncope or cold and paralyzed extremities. Age ranged from 6-222 months.

In 246/436 cases an echocardiographic diagnosis of heart disease was made being congenital heart disease in 8 cats and acquired heart disease in 238 cases.

Concentric left ventricular hypertrophy (cLVH) was found in 200/238 cases with or without dynamic left ventricular outflow tract obstruction (DLVOTO) in 105 cases and 95 cases respectively. Isolated DLVOTO was found in 12 cats. Unclassified, Restrictive, Dilated and Arrhythmogenic Right Ventricular Cardiomyopathy were identified in 7,4,1 and 1 cats.

Left atrial enlargement (LAE) was found in 40/200 cats with cLVH. LAE was present in 6/7 UCM, 4/4 RCM, 1/1 DCM and 1/1 ARVC cases.

Concentric left ventricular hypertrophy is by far the most frequently identified cardiac abnormality in cats in general practice. Other forms of heart disease are relatively rare. LAE is more

commonly found in cats with UCM, RCM, DCM or ARVC than in cats with cLVH.

Conflicts of interest: No conflicts of interest reported.

ESVC-O-7 VALIDATION OF SMARTPHONE APPLICATION SOFTWARE IN THE MEASUREMENT OF HEART RATE IN DOGS AND CATS. K. Creamer, D.N. Clements, R. Mellanby, G. Culshaw. R(D)SVS Hospital for Small Animals, University of Edinburgh, EDINBURGH, United Kingdom

The hypothesis of this study was that smartphone application software (Apps) can provide an easy, cheap, accurate, reliable and readily available method for owners to monitor their animal's heart rates (HRs) at home. The aims of this study were to determine the degree of correlation between App-derived heart rates and ECG-derived heart rates and to determine whether the strength of the correlation was influenced by the following variables: species, body size, panting or vocal, clipped or unclipped coat, anaesthetised or conscious.

Heart rates were obtained with 3 different commercially available Apps and compared with those from simultaneously recorded ECGs in 7 cats and 37 dogs referred to the Royal (Dick) School of Veterinary Studies. The Apps used were the "Instant Heart Rate Monitor"(IHRM) which uses the camera to detect a pulse and the "Heart Beat Monitor"(HTM) and "Heart Monitor"(HM) which use the microphone to detect heart sounds. Animals were selected according to demeanour and absence of an auscultatable arrhythmia.

Using Pearson's correlations, there was no significant correlation between HM and ECG for heart rates in dogs ($r = -0.096$, $p = 0.613$) and cats ($r = 0.622$, $p = 0.136$), for HTM in dogs ($r = -0.208$, $p = 0.353$) and cats ($r = -0.069$, $p = 0.871$) or for IHRM in cats ($r = -0.402$, $p = 0.371$). For IHRM in dogs, there was significant correlation with the ECG derived heart rate ($p = 0.036$) but with $r = -0.497$, suggesting the IHRM was half-sampling the heart rate. Using Bland-Altman plots for all 3 Apps, a high proportion of data points fell outside 95% confidence intervals suggesting that errors were not consistent and a correcting factor could not be used.

Comparing HRs with a 2-sample t-test, for the HM App, size had a significant effect in dogs ($p=0.026$) with the error increasing significantly in large dogs versus small dogs. For the HBM app, the error increased significantly comparing large dogs ($p = 0.022$) and medium dogs ($p = 0.039$) versus small dogs. There was also a greater error in awake versus anaesthetised cats and clipped versus non clipped cats (both $p = 0.007$). None of the variables significantly affected the error in the IHRM app.

In conclusion, this study failed to demonstrate that the 3 selected smartphone Apps were reliable or accurate in calculating heart rate in cats or dogs. Further development in smartphone technology is required to allow owner assessment of heart rate in dogs and cats.

Conflicts of interest: No conflicts of interest reported.

ESVC-O-8 24 HOURS HOLTER MONITORING OF CATS WITH ASYMPTOMATIC HCM BEFORE AND AFTER TREATMENT WITH ATENOLOL. A.S. Hanäs¹, A.E.M. Tidholm¹, B.E. Ström Holst². ¹Djursjukhuset Albaño, STOCKHOLM, Sweden, ²Swedish University of Agricultural Sciences, UPPSALA, Sweden

Heart rate and presence of arrhythmia in cats with asymptomatic hypertrophic cardiomyopathy (HCM) has not been well investigated. We performed a double blinded, randomized, and placebo controlled pilot study using 24 hour ECG system, i.e. Holter monitoring. The aim was to determine heart rate, number of ventricular ectopic complexes (VECs), supraventricular ectopic complexes (SVECs), and blood pressure before and after treatment with atenolol or placebo. Inclusion criteria: Left ventricular end-diastolic lateral wall (LWVd) and/or interventricular septal

diameters (IVSd) >6 mm and systolic blood pressure <170 mmHg in absence of other cardiac or systemic disease or congestive heart failure (CHF). Blood was analysed for creatinine, ALAT, total T4 and glucose. Echocardiography with 12 MHz probe was used. Seven electrodes were placed on ventral thorax providing a 3 lead ECG. The cats were released to roam freely in the home environment during 24 hours. Fifteen privately owned HCM cats of 6 different breeds, age 1.4-11.2 years (mean 5.1), five females and ten males, were included. At initial visit, echocardiography, blood pressure and Holter was performed. IVSd and LWVd were 6-7.2 mean 6.5 mm. Mean Holter time was 23 hrs 11 min. Cats were treated with a tablet once daily for 30 days (atenolol 6.25 mg or placebo). At revisit blood pressure was measured and Holter again recorded (mean 23 hrs 32 min). Holter data were manually analysed by one of the authors (SH) using Welch Allyn's PCH100 Holter analysis system. Three cats were excluded from the study due to CHF, not being able to medicate properly, or computer problems. Before treatment, mean heart rate was 158 bpm SD 24. The cats had normal sinus rhythms with tall R waves or left anterior fascicular block appearance and mild to moderate sinus arrhythmia. Median number of VECs was 3 IQR 1-29. Median number of SVECs was 0 IQR 0-0. Mean blood pressure 140 mmHg SD 13.7 cats received placebo and 5 cats received atenolol. No significant differences were detected within groups before compared to after treatment or between the groups after treatment with respect to heart rate, VECs, SVECs or blood pressure.

In conclusion, there was no effect of atenolol on heart rate, number of VECs, SVECs or blood pressure. The results of the present study do not support routine treatment of asymptomatic HCM cats with atenolol. However, a larger number of cats may be needed to obtain conclusive results.

Conflicts of interest: No conflicts of interest reported.

ESVC-O-9

RESPONSE OF SUPRAVENTRICULAR TACHYCARDIA TO MANUAL CARDIOVERSION (CHEST THUMP). M.P. Perigo, R. Ferriani, R.A.S. Santilli. Clinica Veterinaria Malpensa, SAMARATE, Italy

Manual cardioversion (MC) is a procedure that can be used to treat supraventricular tachycardias (SVT). To perform a MC, an operator strikes a blow with the fist to a specific place on the thorax. The object of the study was to describe the efficacy of MC in interrupting SVTs and the electrocardiographic variations of SVTs sequence post-MC. Twelve-lead electrocardiograms of 74 dogs underwent electrophysiological study were retrospectively analyzed. Nineteen dogs of different breeds with a mean age (+SD) 3.07 ± 2.8 years, a mean body weight of 35.3 ± 11.7 kg, a M:F of 0.6 were selected. In thirteen cases, MC was performed during orthodromic atrioventricular reciprocating tachycardia (OAVRT) and in 6 cases during focal atrial tachycardia (FAT). Normal distribution of values was assessed by the Shapiro-Wilk W-test. Normally distributed data were tested using Student's t-test and non-normally distributed data using Wilcoxon's sum rank test. In 84% of the cases, MC interrupted SVT (92.3% of OAVRT and 66.6% of FAT) with no statistical difference between the two groups (p 0.15). In 100% of the cases the last deflection before the interruption of the SVT was a P' wave. The mean duration of the pause after the interruption was 930.06 ± 500.91 msec (1146.9 ± 383.86 msec OAVRT, 333.75 ± 182.34 msec FAT) and significant longer in OAVRT. In 100% of cases, after the pause sinus rhythm (SR) was present and SVT re-occurred in 5119.25 ± 2962.28 msec (5000.83 ± 3262.07 msec OAVRT, 5296.5 ± 825.19 msec FAT). After MC, P'R intervals progressively prolonged in 100% of patients (OAVRT: pre-MC 135.69 ± 11.94; post-MC 143.81 ± 22.98 msec; FAT: pre-MC 10.0 ± 27.93; post-MC 118.5 ± 44.37 msec), suggesting a possible concealed conduction of the artificial potential along the AV node. RP' interval remained unchanged in 100% of the cases (OAVRT: pre-MC 83.69 ± 20.32, post-MC 85.09 ± 24.57 msec; FAT: pre-MC 88.5 ± 16.97, post-MC 84.5 ± 27.58 msec) suggesting no effect on the accessory pathway retrograde conduction or atrial ectopic focus discharge rate. No complications occurred. Based on these

results, MC can be considered a successful and safe procedure for transient interruption of SVTs in dogs, it allows the comparison of P-QRS-T waves during SVT and during SR that can lead to the possible diagnosis of underlying electrogenic mechanism.

Conflicts of interest: No conflicts of interest reported.

ESVC-O-10

EVALUATION OF CARDIAC TROPONIN I FOLLOWING DUAL CHAMBER PACEMAKER IMPLANTATION IN DOGS WITH ATRIOVENTRICULAR BLOCK. N.J. Wiedemann, A. Stosic, E. Henrich, E. Hassdenteufel, N. Hildebrandt, M. Schneider. Small Animal Clinics Justus-Liebig University, GIESSEN, Germany

Cardiac Troponin I (cTnI), a myocardial regulatory protein, is released into circulation following myocardial injury. It can be used as a marker for the degree of myocyte injury, disease progression or offers prognostic information. An elevation of cTnI has been described in dogs with significant bradyarrhythmias. The cause of elevation remains unclear as values post single chamber pacemaker implantation decreased but remained above the reference range in majority of cases. Aim of this prospective study was to evaluate if AV synchronous pacing after dual chamber pacemaker implantation reduces and possibly normalizes cTnI and whether other factors influencing cTnI can be determined.

Only dogs with permanent high grade second or third degree atrioventricular block and clinical signs of bradyarrhythmia requiring pacemaker implantation were prospectively included. All dogs received a dual chamber pacemaker (DDD or DDDR). Fifteen dogs were presented between April 2010 and September 2012 (n = 9 female, n = 6 male). Serum for cTnI were taken prior to pacemaker implantation and on short-term follow-up (113 ± 28 days). Samples for cTnI were separated and frozen at -20°C within 30 minutes and measured by an ultrasensitive assay (ADVIA Centaur TnI Ultra, Siemens Diagnostics) in an external laboratory. On initial and follow-up presentation complete echocardiography was conducted. The Index of diastolic left ventricular diameter (LVDd-I) was measured as published. The event recording of the pacemaker was analysed at short term follow-up. Data were tested for normal distribution. cTnI and LVDd-I were compared between initial and follow-up examination.

Main presenting complaints were syncope (n = 13/15), exercise intolerance (n = 7/15) and panting (n = 5/15). One dog showed signs of congestive heart failure, two dogs were suspicious for atrial myocarditis. The mean age and weight was 7.72 ± 3.41 years and 29.92 ± 11.96 kg, respectively. On initial presentation cTnI was 0.76 ng/ml (range 0.03 - 13.8 ng/ml). On re-check the cTnI concentration decreased significantly to 0.11 ng/ml (0.03 - 1.36 ng/ml; P = 0.0107). Despite AV synchronous stimulation cTnI remained elevated in most dogs (12/15). The left ventricular dimensions decreased (p = 0.002), suggesting efficacy of physiologic pacing.

We identified significant decreased cTnI and left ventricular size at short-term follow-up. Further follow-up is warranted to show longterm benefits of physiologic pacing. Causes for increased cTnI concentrations remain unclear and warrant further investigation.

Conflicts of interest: No conflicts of interest reported.

ESVC-O-11

MEXILETINE SERUM LEVELS WITH TWICE DAILY DOSING IN COMBINATION WITH SOTALOL IN HEALTHY DOGS. K.F. Scollan, D.D. Sisson. Oregon State University, CORVALLIS, United States of America

Sotalol and mexiletine are commonly used oral antiarrhythmic medications to treat ventricular arrhythmias in dogs. Previous human and veterinary studies have indicated enhanced efficacy of combination therapy over monotherapy with these drugs. One of the major limitations of using mexiletine in dogs is the require-

ment of dosing every eight hours, which is impractical or impossible for many dog owners. A previous veterinary study revealed higher mexiletine serum levels when administered in combination with sotalol compared to levels measured with mexiletine monotherapy. We hypothesized that therapeutic serum levels of mexiletine may be maintained with twice daily administration when used in combination with sotalol.

The objective of this study was to assess mexiletine serum levels when administered twice daily in combination with sotalol in normal dogs. The study protocol was approved by the Animal Care and Use Committee of Oregon State University. Purpose bred dogs, four Beagles and two hounds, determined to be healthy by normal physical and echocardiographic examinations and bloodwork, received twice daily sotalol (2.5 mg/kg) and twice daily mexiletine (8-10 mg/kg) orally for ten days. On the eleventh day, whole blood was drawn at 0, 1, 2, 3, 4, 6, 8, 10, and 12 hours post pill with uninterrupted drug administration. Mexiletine serum levels were measured at all time points while sotalol serum levels were measured at 0, 3, 6, and 12 hours post pill. Serum drug concentrations were measured by high performance liquid chromatography at a commercial laboratory.

Median peak serum mexiletine concentration measured at 3 hours post pill was 1200 ng/mL (range 707-1407) and median trough concentration measured 12 hours post pill was 296.5 ng/mL (range 200-644). Mexiletine serum levels were found to remain within the described therapeutic range for humans, 500-2000 ng/mL, for the duration of 12 hours in three of the six dogs with twice daily administration. These three dogs included the largest three dogs within the group of six (two hounds and one Beagle). Comparing body weight subgroups, dogs greater than 10 kg ($n = 3$) had significantly higher peak and trough serum mexiletine levels compared to those less than 10 kg ($p = 0.050$ and 0.038 , respectively).

Our results indicate that therapeutic serum mexiletine levels are maintained with twice daily dosing when used in combination with sotalol in dogs greater than 10 kg. The therapeutic implications and efficacy of mexiletine serum levels in affected dogs should be investigated.

Conflicts of interest: No conflicts of interest reported.

ESVC-O-12

PATHOLOGIC MANIFESTATIONS AND CLINICAL CORRELATES IN CANINE DEGENERATIVE MITRAL VALVE DISEASE AT SURGICAL BIOPSY. J. Lee, M. Mizuno, T. Mizuno, K. Harada, T. Sawada, A. Shinoda, S. Uchida, M. Uechi. Nihon University, FUJISAWA, Japan

Degenerative mitral valve disease (DMVD) is the leading cause of congestive heart failure (CHF) in small breed dogs. Although myocardial histopathologic change is key process of developing CHF, its clinical relevance has not been fully evaluated. The aims of this study were to characterize pathologic alterations and investigate their relationship to clinical aspects of naturally occurred DMVD in dogs.

Biopsies from left atrial appendage and ventricle (LA and LV) and lung were taken from 117 surgical mitral valve repair cases. Specific pathologic changes were assessed in LA and LV (fatty replacement, myocardial vacuolization, infiltrated immune cells, increased perinuclear space, enlarged nuclei, hypertrophy, interstitial fibrosis), as well as in lung (alveolar septal thickening, heart failure cells and hyperplasia of type II pneumocytes), by using either semi-quantitative scoring or computer-based digitizing system. The quantified pathologic findings were analyzed in terms of the relationship to the severity (ISACHC) and the most commonly used clinical indicators, such as vertebral heart score, left ventricular end-diastolic dimension (LVEDd), left atrial-to-aortic diameter ratio (LA/Ao), fractional shortening (FS), ejection fraction (EF), and velocity of E and A wave.

Substantial pathologic progressions were found in all ISACHC groups except for the heart failure cell significantly increased only in class III ($p < .0001$). In a paired comparison of LA and LV, LA had significantly more severe myocardial fatty replacement, immune cell infiltration, and interstitial fibrosis than LV ($p < .0001$). Meanwhile, cardiomyocytes in LV were more hypertrophied than those in LA ($p < .0001$). Regression analysis

adjusted by age and body weight revealed that LVEDd was well associated with fatty replacement ($p = .033$, $R^2 = .584$) and myocardial vacuolization ($p = .003$, $R^2 = .588$) in LA. Interstitial fibrosis was negatively related to EF in both LA ($p = .012$, $R^2 = .231$) and LV ($p = .036$, $R^2 = .205$). However, FS showed no relations to any of the pathologic variables. Also, heart failure cell had relations to LA/Ao ($p = .005$, $R^2 = .174$), E wave ($p = .023$, $R^2 = .161$), and A wave ($p = .015$, $R^2 = .187$).

In DMVD, severe pathologic changes may be progressed from very early stage of CHF. Not only mechanical injury of LA but different hemodynamic strain resulting from mitral regurgitation can cause pathologic disparity between LA and LV. LVEDd and EF may be relatively effective in predicting myocardial pathologic state.

Conflicts of interest: No conflicts of interest reported.

ESVC-O-13

DIAGNOSTIC VALUE OF ECHOCARDIOGRAPHIC VARIABLES TO IDENTIFY PULMONARY ARTERIAL HYPERTENSION IN DOGS WITH MYXOMATOUS MITRAL VALVE DISEASE. A.E.M. Tidholm¹, I. Ljungvall², J. Häggström², K. Höglund³. ¹Albano Animal Hospital, DANDE-RYD, Sweden, ²Dept of Clinical Sciences, Faculty of Veterinary Medicine, UPPSALA, Sweden, ³Dept of Anatomy, Physiology and Biochemistry, UPPSALA, Sweden

Pulmonary arterial hypertension (PAH) is a common sequela to myxomatous mitral valve disease (MMVD) in dogs and may influence survival. Dogs with PAH may present without measurable tricuspid regurgitation (TR), and it would be useful to identify other echocardiographic variables that can identify PAH in such dogs. The aim of the study was to correlate estimated right ventricular systolic pressure (RVSP), which is equal to pulmonary arterial systolic pressure (PASP) in the absence of pulmonic stenosis (PS), with various echocardiographic variables in dogs with MMVD and measureable TR.

A total of 104 dogs of 42 different breeds with MMVD and detectable TR, but no evidence of PS, were included in the study. According to ACVIM classification of congestive heart failure (CHF), 34 dogs were classified with CHF and 70 dogs without CHF. TR velocities and subjective estimations of right atrial (RA) pressures were used to indirectly assess RVSP. The RA pressure was arbitrarily considered 5 mm Hg in the absence of RA enlargement and 10 mm Hg in the presence of RA enlargement. None of the dogs presented with signs of right-sided CHF. RVSP ranged from 15 to 100 mm Hg and dogs were dichotomized in 2 groups; 1) $TR \leq 2.9$ m/s corresponding to $RVSP \leq 34$ mm Hg ($n = 53$) and 2) $TR > 2.9$ m/s corresponding to $RVSP > 34$ mm Hg ($n = 51$). There were no significant differences between groups regarding age, body weight, sex and percentage increase in left ventricular internal diameter in end-diastole (LVIDd inc%). Heart rate ($P = 0.0019$) and left atrial diameter to aortic diameter ratio (LA/Ao) ($P = 0.0052$) were significantly higher in group 2 compared to group 1.

Estimated RVSP was correlated to decreasing pulmonary artery (PA) acceleration time to deceleration time ratio (AT/DT), and to right ventricular internal diameter in end-diastole (RVIDd) correlated to body weight. There was no correlation between RVSP and tricuspid annular plane systolic excursion (TAPSE) or PA flow velocity.

In the multiple regression analysis with RVSP as dependent variable, AT/DT ($P < 0.0001$) and RVIDd/kg^{0.33} ($P = 0.0075$) remained significant in the final model, which had an adjusted R^2 - value of 0.27

In conclusion, presence of PAH may be suspected with decreasing AT/DT ratio and increasing RVIDd/kg^{0.33} in dogs with MMVD.

Conflicts of interest: No conflicts of interest reported.

ESVC-O-14

LONGITUDINAL RIGHT VENTRICLE STRAIN AND STRAIN RATE BY TWO DIMENSIONAL FEATURE TRACKING ECHOCARDIOGRAPHY IN HEALTHY DOGS. C. Locatelli¹, A. Mavropoulou², I. Spalla¹, G. Riscuzzi¹, P. Crepaldi¹, P.G. Brambilla¹, C. Bussadori², ¹Università degli Studi di Milano, MILAN, Italy, ²Clinica Veterinaria Gran Sasso, MILAN, Italy

Although multiple methods have been described for quantitative right ventricle (RV) assessment using standard transthoracic echocardiography, assessment of RV structure and function remains mostly qualitative in clinical practice. Several 2D based techniques have been developed to assess myocardial function in a non-invasive and angle independent way. Their difference is based on the type of algorithm employed. The first and more investigated method is known as 'speckle tracking' and the second one is known as 'feature tracking' (FTE). In FTE, the software is based on a dedicated algorithm that follows frame by frame, the endocardial border; actually this processing system is based on a mono-dimensional technology.

The aim of the study was to determine reference values for longitudinal RV, global and regional (6 segments: basal, middle, and apical septum and basal, middle, and apical lateral wall), peak systolic strain (St) and strain rate (SR) using FTE in clinically healthy dogs.

Cine-loops were acquired from left parasternal standard 4 chamber apical view optimized for RV with 2-7.5 MHz probes with a MyLab50 Gold system (Esaote, Florence) and analyzed with regards to St and SR using the XStrain software, based on a FTE algorithm. For all variables, a mean of 3 measures was used for the statistical analysis. Influence of age, gender, body weight, heart rate (HR) and type of breed ('non athletic' versus 'athletic') on St and SR variables was tested by multivariate analysis of variance. One-way analysis of variance was used to investigate differences between myocardial segments.

Sixty dogs, 39 'non athletic' and 21 'athletic' were prospectively included in the study. Twenty-nine were females and thirty-one were males, with a mean (\pm SD) age of 5 ± 3.5 years, body-weight of 20.8 ± 11.2 kg, and HR of 114.9 ± 27.5 beats per minute.

Reference values were established for every myocardial segment. Body weight and type of breed showed a statistically significant effect on most St and SR variables analyzed ($P < 0.05$). RV septal longitudinal St and SR variables were significantly higher at base compared to apex ($P < 0.05$) and in the lateral wall compared to septum ($P < 0.05$).

In conclusion, FTE assessment of longitudinal RV St and SR was feasible in healthy dogs revealing a septal longitudinal apex to base gradient. Most St and SR variables has been shown to be influenced by body weight and type of breed ('non athletic' versus 'athletic'). Further studies are needed to confirm these results.

Conflicts of interest: No conflicts of interest reported.

ESVC-O-15

ECHOCARDIOGRAPHIC ASSESSMENT OF THE CANINE RIGHT HEART: REFERENCE INTERVALS AND REPEATABILITY. J.M. Gentile, J.A. Abbott, Virginia Tech, BLACKSBURG, United States of America

There is a paucity of published, normative echocardiographic data that relate to structure and function of the canine right heart. Accordingly, we sought to define echocardiographic reference intervals for dimensions of the canine right heart and to describe the measurement variability of echocardiographic indices of right heart structure and function.

Forty-five healthy dogs of diverse somatotype and body weight (BW) were subject to echocardiographic examination by a single operator. Six of these dogs were randomly selected to undergo twice daily echocardiographic examinations by two operators on three non-consecutive days. To describe repeatability of echocardiographic indices used to estimate pulmonary artery pressure, four clinically stable, client-owned dogs with tricuspid valve regurgitation (TR) also were subject to repeated echocardiographic

examinations. Raw data from healthy dogs were subject to logarithmic (\log_{10}) transformation and the allometric relationship between body size and echocardiographic variables was determined; reference intervals intended to include 95% of the population were defined. A mixed model was developed to evaluate the fixed effects of operator, day, time-within-day and the random effect of subject. Measurement variability was described by calculation of the repeatability coefficient (RC) from variance components and also expressed as within-day, between-day and between-operator% coefficients of variation (CV).

The strength of the linear relationships between \log_{10} transformed variables and BW was variable; the range of coefficients of determination (R^2) was 0.055-0.872. For linear dimensions, the scaling exponent of derived allometric equations was between 0.14 and 0.39. For measurements of chamber area, scaling exponents were between 0.58 and 0.71. Two hundred-seven [91%] of 228 calculated CV were less than 15. CV from M-mode variables generally were higher than those from two dimensional echocardiography (2DE). The range of within-dog/between-day/between-operator RC for linear dimensions was 2.8-19.2 mm, and for areas, 495.1-655.3 mm². The analogous RC for peak TR velocity was 0.88 m/s. The within-dog/within-operator/between-day RC for peak TR velocity was 0.32 m/s. The range of CV for TR velocity was 3.2-7.1.

Reference intervals for echocardiographic dimensions and indices of right heart function are proposed. Selected linear dimensions and areas obtained from 2DE have "acceptable" repeatability. When the same operator obtains serial measurements, changes in TR velocity that exceed 0.32 m/s are outside of the expected range of measurement variation.

Conflicts of interest: No conflicts of interest reported.

This study was funded by a grant provided by the not for profit Virginia Veterinary Medical Association - Veterinary Memorial Fund; this does not represent a conflict of interest.

ESVC-O-16

CLINICAL EVALUATION OF A SECOND-GENERATION CANINE NT-PROBNP ASSAY. J.S. Buch¹, M.A. Oyama², J.E. Rush³, P. Fox⁴, T.P. Nguyenba⁵, B.J. Bulmer³, S.M. Cunningham³, H. Kellihan⁶, S. Gordon⁷, B.K. Lefbom⁸, T.C. DeFrancesco⁹, J.M. Macgregor¹⁰, L.B. Lehmkuhl⁵, R.L. Stepien⁶, W.A. Brown¹¹. ¹IDEXX Laboratories, WESTBROOK, ME, United States of America, ²University of Pennsylvania, PHILADELPHIA, PA, United States of America, ³Tufts University, NORTH GRAFTON, MA, United States of America, ⁴Animal Medical Center, NEW YORK, NY, United States of America, ⁵MedVet, WORTHINGTON, OH, United States of America, ⁶University of Wisconsin, MADISON, WI, United States of America, ⁷Texas A&M University, COLLEGE STATION, TX, United States of America, ⁸Chesapeake Veterinary Cardiology Associates, VIENNA, VA, United States of America, ⁹North Carolina State University, RALEIGH, NC, United States of America, ¹⁰InTown Veterinary Group, WOBURN, MA, United States of America, ¹¹Veterinary Cardiology Consultants, NOVI, MI, United States of America

NT-proBNP is increased in dogs with heart disease, can differentiate symptomatic respiratory disease from heart failure, and stratifies heart disease severity. However, the commercially-available canine NT-proBNP assay has a limited dynamic range which reduces the clinical utility of the biomarker in canine patients with severe heart disease. To address this limitation, a new second-generation assay for canine NT-proBNP has been developed to extend the upper limit of quantitation from 3,000pmol/L to 40,000pmol/L. A method comparison was conducted to retrospectively evaluate the clinical performance of the second-generation NT-proBNP assay using samples from a previous multicenter trial (NT-proBNP Concentrations in Dogs with Respiratory Signs, Heart Disease, and Heart Failure: Preliminary Results from a Prospective Multicenter Field Study, Oyama et al, ECVIM 2010).

Patients recruited from 11 referral centers were prospectively assigned to one of 4 groups based upon physical exam, ECG, radiographs, and echocardiography: Group 1) healthy controls; Group 2) asymptomatic heart disease; Group 3) respiratory signs

attributable to congestive heart failure; Group 4) respiratory signs attributable to primary respiratory or airway disease. Dogs with cardiac disease were further categorized by the attending DVM as having insignificant (I), mild/moderate (M) or severe (S) cardiac disease. Of 272 plasma samples originally collected and retained at -80°C, 190 had sufficient volume for inclusion in this comparison study.

NT-proBNP ranged from <50 to >3,000 pmol/L and 29 to 19,873 pmol/L on the first-generation and second-generation assays, respectively. A Bland-Altman analysis indicated an overall bias of 68 pmol/L between assays that was statistically insignificant ($P = 0.1588$). For both assays, NT-proBNP between the 4 patient groups was differentiated with statistical significance; $P < 0.0001$. Using a cutoff of 1,800 pmol/L for both assays, NT-proBNP equally distinguished CHF from primary respiratory disease with sensitivity of 89.2% and specificity of 67.4% (AUC=0.7957 and AUC=0.8573 for first and second-generation assays, respectively). For dogs diagnosed with cardiac disease, NT-proBNP similarly distinguished disease severity with statistical significance. Median NT-proBNP by group for current assay was: Group I, 626 [IQR: 403-911], $n = 20$; Group M, 1232 [579-2334], $n = 61$; Group S, 3001 [2680-3001], $n = 61$; $P < 0.0001$. Median NT-proBNP by group for second-generation assay was: Group I, 677 [510-1045], $n = 20$; Group M, 1301 [724-2050], $n = 61$; Group S, 3742 [2269-5685], $n = 61$; $P < 0.0001$.

A new second-generation assay for canine NT-proBNP demonstrates equivalent clinical performance to its predecessor while offering an increased dynamic range to better support the clinical utility of NT-proBNP in canine patients with severe heart disease.

Conflicts of interest: The presenting author is an employee of IDEXX Laboratories which offers a commercial assay for canine NT-proBNP.

ESVC-O-17

EFFECTS OF COMBINATION OF ACEPROMAZINE/BUTORPHANOL ON CONVENTIONAL ECHOCARDIOGRAPHIC MEASUREMENTS AND GLOBAL STRAIN IN HEALTHY DOGS. G. Santarelli¹, M.J. Fernández del Palacio², J. Talavera². ¹Veterinary Teaching Hospital - Murcia University, MURCIA, Spain, ²Department of Veterinary Medicine and Surgery. University of Murcia, Spain., MURCIA, Spain

Pharmacological tranquilization is sometimes necessary to aid and simplify the echocardiographic examination in dogs and obtain scanings of good quality. The effect of sedation with acepromazine (ACE) and butorphanol (BUT) on echocardiographic variables including global strain (GS) has not been reported previously in dogs. We hypothesized that the combination of ACE-BUT has no or minimal effect on cardiac function.

Six volunteer-owned healthy dogs (1.5-10 years-old; 16-35 kg body weight) of different breeds (2 Labrador Retrievers, 1 Golden Retriever, 1 Dalmatian, 1 Spanish Podenco hound dog, 1 Mongrel) were used for the study. Transthoracic echocardiographic examination (2D, M-mode, color flow Doppler, spectral Doppler, TDI) was performed using a commercial echocardiographic system (iE33, Philips) before and 30 minutes after sedation with a combination of ACE (0,02 mg/kg) and BUT (0,2 mg/kg) administered intramuscularly. Radial and longitudinal global strain of the left ventricle (LV) were determined via 2D speckle tracking echocardiography offline analysis (QLAB 9 software application package) using parasternal short axis images at the midpapillary muscle level and left parasternal long-axis apical images. The following parameters were measured/calculated: heart rate (HR), LV M-mode measurements, shortening fraction; 2D LV outflow tract and left atrial dimensions, LA/AO, LV volumes, ejection fraction, cardiac output; mitral, aortic and pulmonary blood flow measurements, myocardial performance index, systolic time intervals; TDI velocities (septal, LV lateral mitral annulus and right ventricular tricuspid annulus) eas index, E/e'; LV radial and longitudinal GS. Statistical analysis were performed with a computer software EPSS. Differences in the parameters prior to and after sedation were assessed with either

the paired Student's test or the Wilcoxon signed rank sum test. $P < .05$ were considered statistically significant.

The ACE-BUT combination was well tolerated and a good degree of sedation requiring minimal or no restraint was achieved, except for a dog that presented panting before and after sedation. The following parameters were significantly modified after sedation: decreased HR (from 114 ± 11 to 90 ± 14 beats/min) ($P.028$), LV diastolic volume (from $52,7 \pm 14,2$ to $43,9 \pm 11,6$ ml) ($P.028$), LV radial GS (from -28 ± 2 to $-21 \pm 11\%$) ($P.046$); increased LV end-diastolic free wall thickness (from $0,97 \pm 0,9$ to $1,05 \pm 0,5$ cm) ($P.028$).

In conclusion, sedation with ACE-BUT at the doses used in this study showed little influence on echocardiographic measurements and cardiac function.

Conflicts of interest: No conflicts of interest reported.

ESVC-O-18

PULMONARY VEIN TO PULMONARY ARTERY RATIO IS AN ECHOCARDIOGRAPHIC INDEX OF LEFT CONGESTIVE HEART FAILURE IN DOGS WITH MYXOMATOUS MITRAL VALVE DISEASE. A.C. Merveille¹, M. Cabrero¹, E. Krafft¹, A.L. Etienne¹, P. Jaspers², S. Gomart², G. Bolen¹, C. Clercx¹, K. Mc Entee². ¹University of Liège, LIÈGE, Belgium, ²Université Libre de Bruxelles, BRUSSELS, Belgium

Early recognition of left congestive heart failure (CHF) in dogs with myxomatous mitral valve disease (MMVD) is of clinical importance. Thoracic radiography is considered the clinical gold standard while several Doppler-echocardiographic variables of left ventricular filling have been tested successfully. Recently, a novel echocardiographic index: the pulmonary vein to pulmonary artery diameter ratio (PV/PA) has been described and might be helpful to distinguish dogs in CHF from those in a compensatory state. The aim of this study was to investigate if PV/PA could predict CHF in dogs with MMVD. For this prospective study, 35 dogs were recruited, including 6 healthy dogs and 29 dogs with MMVD including ISACHC stage 1 (15), stage 2 (7) and stage 3 (7), based on physical examination echocardiography-Doppler (in all dogs) and thoracic radiographs (in 22 dogs). Radiographs were assessed for signs of CHF, using a composite score based on cardiomegaly signs, venous congestion and lung pattern, by two readers blinded regarding echocardiographic results. PV/PA was measured in bi-dimensional (BD) and M-mode (MM) at the end of the T wave by two investigators blinded to radiographs results. Inter-observer variability for PV/PA was identical in BD and MM (coefficient of variation of 9% in each). PV/PA BD was highly correlated to PV/PA MM ($r = 0.815$, $P < 0.001$). PV/PA in MM was lower in healthy dogs (1.04 ± 0.1) compared to dogs with MMVD stage 1 (1.57 ± 0.08 , $P < 0.05$) and gradually increased with ISACHC stages (2.06 \pm 0.2 in stage 2, 2.90 \pm 0.3 in stage 3, $P < 0.05$). In BD, PV/PA in healthy and MMVD stage 1 dogs differed from MMVD stages 2 and 3 ($P < 0.01$). PV/PA in BD and MM were correlated to other echocardiographic left ventricular filling indices such as the indexed left atrial (LA/Ao) and left ventricular sizes (LVIDd/Ao, EDVI) ($P < 0.001$) and to E/A ratio ($P < 0.001$). PV/PA ratio was increased in dogs with a composite radiographic score ≥ 2 compared to dogs with a score < 2 . Areas under the receiver-operating characteristic curve showed that a cut-off value of 1.75 for the PV/PA ratio in MM and BD predicted CHF in dogs with MMVD with a sensitivity of 100% and 89% and a specificity of 83% and 92%, respectively. PV/PA ratio is a simple, reproducible, sensitive and specific echocardiographic index and contributes to assess CHF status in dogs with MMVD.

1. Biretoni et al., «A Novel Echocardiographic Index in the Dog: Pulmonary Vein: Pulmonary Artery Diameter», 20th ECVIM-CA Congress, 2010

Conflicts of interest: No conflicts of interest reported.

ESVC-O-19

HEART SPECIFIC GENE EXPRESSION PROFILING IN PERIPHERAL BLOOD OF DOGS WITH CHRONIC HEART FAILURE. G. Kiss, F. Manczur. Faculty of Veterinary Science, Szent István University, BUDAPEST, Hungary

High-throughput gene expression analyzing methods (e.g. microarray) are widely used today to study complex disease processes, and as such, in cardiological research. The major obstacle of these studies is the limited availability of heart tissue samples. Previous studies showed that lymphocytes in peripheral blood can reflect the expression profiles of several organs including the heart in humans. This is due to interaction of blood cells with every tissue and its microenvironment in the body. Blood based expression profiles may offer a powerful tool for screening molecular events in the heart during the disease courses.

Our research aimed to investigate correlation of gene expression (mRNA) profiles between the heart and the peripheral blood in dogs with chronic heart failure. Heart tissue and blood samples were collected for RNA isolation from animals that were used in other purpose approved human medical research projects. Tachypacing induced cardiomyopathy dogs with severely decreased contractility ($n = 6$) were compared to healthy controls ($n = 4$) in pooled samples with equal gender ratio. Whole blood was obtained in special sampling tubes suitable for immediate total RNA stabilization and storage. Cardiac left ventricular free wall (transmural) tissue samples were collected into RNAlater solution after euthanasia. Isolated total RNA quantity and quality was controlled by spectrophotometry and capillary electrophoresis and proved proper for gene expression profiling. Gene expression levels were measured by dual color microarray method (oligonucleotide microarray 4x44K format covering the total canine transcriptome). Results showed 1150 differently expressed transcripts (fold change ≥ 2 ; Student's t-test with Benjamini-Hochberg multiple test correction, $p \leq 0.05$; two technical repeats) between heart failure and control groups in heart samples. From these transcripts 123 (10,7%) could be identified similarly also in the blood samples. This subgroup consist of numerous mRNA transcripts related to extracellular matrix proteins, inflammatory and stress response, connective tissue turnover, apoptosis and contractility (e.g. versican, osteopontin, heat shock protein-90, S100-protein, CD163, IL-8, IL-17, alpha-2 collagen type-I, matrix metalloproteinase-1, mitogen-activated protein kinase, chemokine ligand 2, fibroblast growth factor, myosin heavy chain-7). Pathway analysis resulted fibrosis as the main significant process. This finding is in line with major characteristic of the tachypacing induced model described in the literature (decreased contractility, fibrosis, remodeling). Our results suggest that blood may reflect heart-specific gene expression changes in dogs with heart failure.

Conflicts of interest: No conflicts of interest reported.

ESVC-O-20

BIOPTERIN STATUS IS ASSOCIATED WITH DISEASE SEVERITY AND HUMAN CARDIOVASCULAR RISK FACTORS IN DOGS WITH MYXOMATOUS MITRAL VALVE DISEASE. M.J. Reimann¹, J. Häggström², A. Mortensen¹, J. Lykkesfeldt¹, J.E. Möller², L.H. Olsen¹. ¹University of Copenhagen, FREDERIKSBERG, Denmark, ²Swedish University of Agricultural Sciences, UPPSALA, Sweden, ³Odense University Hospital, ODENSE, Denmark

Endothelial dysfunction represents a therapeutic target and has been suggested to be associated with myxomatous mitral valve disease (MMVD) in dogs. Tetrahydrobiopterin (BH4) is an important cofactor for production of the endothelium-derived vasodilator nitric oxide (NO). Under conditions of oxidative stress, BH4 is oxidised to the biologically inactive form dihydrobiopterin (BH2). Thus, plasma levels of BH2 and BH4 have been suggested to reflect endothelial function. The aim of the study was to determine plasma concentrations of BH2 and BH4 in dogs with different degrees of naturally occurring MMVD.

Clinical examination including echocardiography was performed in 84 privately-owned dogs (13 control dogs (Beagles), 57 cavalier King Charles spaniels with different degrees of MMVD

and 14 dogs of different breeds with clinical signs of congestive heart failure (CHF) due to MMVD).

Plasma levels of BH2 and BH4 were measured by high performance liquid chromatography (HPLC) and fluorescence detection. Differences in BH2, BH4 and BH4/BH2-ratio between disease groups were tested using multiple linear regression.

Dogs in CHF had significantly higher BH4 and BH2 levels than other dog groups ($P < 0.009$).

BH2 and BH4/BH2 levels were found to increase with advancing age ($P < 0.04$). Females had higher levels of BH4 and BH4/BH2 ($P < 0.0001$). Other risk factors such as passive smoking ($P = 0.01$) and increased body weight ($P = 0.02$) were associated with decrease in BH4 levels.

In conclusion, age, gender, body weight, passive smoking and plasma and cardiac status correlate with plasma BH2 and BH4 concentrations in dogs.

Conflicts of interest: I am employed by the University of Copenhagen but affiliated with a research center: LIFE PHARM - that is partly financed by Novo Nordisk A/S.

ESVC-O-21

PREVALENCE OF PHYSIOLOGIC HEART MURMURS IN HEALTHY YOUNG ADULT DOGS. A. Drut, T. Ribas, F. Floch, C. Cottin, B. Rannou, L. Freyburger, S. Franchequin, J.L. Cadoré, I. Bublot. VetAgro Sup Lyon, MARCY L'ETOILE, France

Physiologic heart murmurs (PHM) originate through normal flow patterns in the absence of abnormalities of the heart and great vessels. They are described in young animals less than 6 months and in pathological conditions altering the pattern of blood flow (anemia, fever, hypertension...). PHM are documented in adult boxers and athletic dogs but their prevalence in the general population of adult dogs is unknown. Distinguishing between physiologic and pathologic murmurs in otherwise healthy animals may be challenging for the clinician. The objective of the study is to assess the prevalence of PHM in healthy young adult dogs.

A pilot study performed between November 2011 and April 2012 on 25 staff- or student-owned dogs suggested a high prevalence of PHM in this population.

The objective of the main study is to evaluate the prevalence of PHM in a larger population of dogs. About a hundred of overtly healthy dogs between 1 and 5 years of age will be enrolled up to July 15th, 2013, via preventive medicine consultations. All dogs undergo complete physical examination and cardiac auscultation by three observers blinded to each other (AD, TR, JLC). Animals are submitted to complete blood count, serum total protein and blood pressure measurements to exclude abnormalities potentially causing heart murmurs. Each murmur is timed, graded, and the point of maximal intensity is recorded. To exclude any cardiac abnormality, a complete echocardiographic examination is performed on the same day of auscultations on every dog with a murmur auscultated by at least one observer. The echocardiographer (IB) is blinded to the murmur characteristics.

So far 36 dogs have been enrolled but 4 were excluded. A murmur was heard in 16/32 dogs. Based on echocardiography, 13/16 murmurs were considered PHM. Of the 3 remaining dogs, two had accelerated aortic flow velocity and one had MVD. Four PHM were auscultated by three observers, 3 PHM were heard by two observers and 6 PHM were noted by only one observer. All PHM were systolic and low grade ($\leq 3/6$). The point of maximal intensity was the left base and/or manubrium in 11/13 dogs. In the 2 remaining dogs, observers did not reach agreement on localization. There was no significant difference in body weight between dogs with PHM (mean 21 kg ± 11) and dogs without murmur (mean 22 kg ± 10).

The final results including prevalence and characteristics of the PHM, correlation between observers and population characteristics will be presented.

Conflicts of interest: No conflicts of interest reported.

ESVC-O-22

SPECKLE TRACKING ECHOCARDIOGRAPHY IN DOGS WITH PATENT DUCTUS ARTERIOSUS. I. Spalla¹, C. Locatelli¹, G. Riscuzzi¹, P. Brambilla¹, C. Bussadori². ¹Università degli Studi di Milano, MILAN, Italy, ²Clinica Veterinaria Gran Sasso, MILAN, Italy

Patent Ductus Arteriosus (PDA) is one of the most common congenital heart defects in the dog and it causes marked left ventricular volume overload (LVvo), leading to cardiac remodeling and finally to congestive heart failure. Pulmonary to systemic flow ratio (Qp:Qs ratio) is one of the criteria described for quantification of PDA shunt in veterinary medicine. PDA induced LVvo can also be quantified by standard transthoracic echocardiography-derived volume equations (TTE -V). Speckle-tracking echocardiography (STE) is an angle independent imaging modality increasingly used to evaluate regional as well as global cardiac function in different clinical scenarios. The aim of our study was to assess global circumferential, radial and longitudinal strain (S) and strain rate (Sr) STE values in 20 dogs with PDA and 10 healthy young dogs, and compare these data to standard echocardiographic parameters of shunting and LVvo.

All dogs underwent a complete echocardiographic study. Cine-loops from right parasternal short axis views (mitral valve, papillary muscle and apical level) and from left parasternal 4 chamber view were acquired and analysed with regards to St and Sr using the Xstrain TM software (Esaote, Florence). For all variables, a mean of 3 beats was used for the statistical analysis.

Dogs with PDA showed a statistically higher TTE-V as compared to normal dogs ($p < 0.001$). A statistical difference was found when global S and Sr were compared in dogs with pda vs control dogs. Mean global circumferential S and Sr were higher ($p < 0.01$) in dogs with pda as compared to healthy control dogs. Mean global radial S and Sr were higher ($p < 0.03$) in dogs with pda as compared to healthy control dogs. Longitudinal S and Sr were higher ($p < 0.001$) in dogs with pda as compared to healthy control dogs. These findings suggest that PDA is associated with an increase in all S and Sr values and is a complementary tool together with standard TTE-V in order to better assess cardiac function in these patients.

Conflicts of interest: No conflicts of interest reported.

ESCG-O-1

EVALUATION OF SERUM METABOLITE CONCENTRATIONS AND FECAL MICROBIOTA IN DOGS WITH IDIOPATHIC INFLAMMATORY BOWEL DISEASE BEFORE AND AFTER TREATMENT. Y. Minamoto¹, C.C. Otoni², B.C. Guard¹, M.E. Markel¹, R.M. Heilmann¹, J.M. Steiner¹, A.E. Jergens², J.S. Suchodolski¹. ¹Gastrointestinal Laboratory, Texas A&M University, COLLEGE STATION, United States of America, ²Iowa State University, AMES, United States of America

The combination of an altered composition of the GI microbial community (dysbiosis), underlying host genetic susceptibility, and environmental factors are suspected to contribute to the pathogenesis of idiopathic inflammatory bowel disease (IBD). Despite well documented evidence that intestinal microbes play a pivotal role in the pathogenesis of IBD, the actual mechanisms of the host-microbe interactions remain elusive, but are believed to be mediated in part by microbial products (metabolites) derived from the GI microbiota. However, only limited information is available about serum metabolite concentrations and the fecal microbial communities in dogs with IBD. The aim of this study was to evaluate serum metabolite concentrations and the fecal microbiota in dogs with IBD before and after medical therapy for this disorder.

Serum and fecal samples were collected from healthy dogs ($n = 10$), and from dogs with IBD ($n = 12$) before and after 21 days of standard medical therapy (e.g., elimination diet and administration of anti-inflammatory drugs). The disease status was assessed using the clinical disease activity index (CIBDAI). Fecal DNA was evaluated for fecal microbial composition using 454-pyrosequencing of the 16S rRNA gene and quantitative PCR (qPCR). Serum metabolites were profiled using a GC-TOF/MS

platform and analyzed using MetaboAnalyst 2.0. Sequence analysis was performed using QIIME v1.6.

Before treatment, the abundance of *Blautia*, *Faecalibacterium*, and *Turicibacter* was significantly decreased in dogs with IBD ($p = 0.037, 0.048, 0.029$, respectively) relative to healthy dogs. Unifrac analysis revealed clustering between healthy and diseased dogs (ANOSIM, $p < 0.05$). While the CIBDAI was significantly decreased, no significant differences in the proportions of bacterial groups were found before and after treatment. Serum analysis revealed 157 annotated metabolites. Serum concentrations of several metabolites differed significantly between healthy dogs and dogs with IBD (decreased in IBD: tyrosine, aminomalonic acid, 4-hydroxyproline, trans-4-hydroxyproline, naphthalene, and proline; increased in IBD: xylitol and allantoin). However no significant changes in concentrations of these metabolites were observed before and after treatment.

In conclusion, altered serum concentrations of some metabolites and a fecal dysbiosis were observed in dogs with IBD. Although a clinical improvement was observed after treatment, this was not accompanied by significant changes in fecal microbiota or serum metabolite concentrations. A longer period of treatment or a different approach to medical therapy (i.e., pre-/probiotics) may be needed for dogs to recover from fecal dysbiosis associated with IBD.

Conflicts of interest: No conflicts of interest reported.

ESCG-O-2

SERUM HOMOCYSTEINE CONCENTRATIONS IN HYPOCOBALAMINEMIC AND HYPOFOLATEMIC GREYHOUNDS. N. Grützner, S.B. Keyser, C.S. Bridges, R.M. Heilmann, J.S. Suchodolski, J.M. Steiner. Gastrointestinal Laboratory, COLLEGE STATION, United States of America

A recent search of the Gastrointestinal Laboratory (GI Lab) database showed that hypcobalaminemia is frequently observed in Greyhounds. It is unknown if a malabsorption of cobalamin or other micronutrients (e.g., folate) due to chronic gastrointestinal disease plays a role in the cause for this finding in Greyhounds. Cobalamin and folate are absorbed in the distal and proximal small intestine, respectively. In humans, low serum cobalamin and folate concentrations have been associated with an increase in serum homocysteine (HCY) concentration, which reflects a lack of the intracellular availability of both vitamins. Increased serum HCY concentrations are associated with cardiovascular and thrombotic diseases in humans, and these conditions have also been described in Greyhounds. Therefore, the aims of this study were to evaluate 1) the frequency of hypofolatemia in hypcobalaminemic Greyhounds, and 2) serum HCY concentrations in hypcobalaminemic and hypofolatemic Greyhounds.

Submissions from Greyhounds ($n = 423$) to the GI Lab (2006-2010) for analysis of serum cobalamin and folate concentrations were reviewed. Hypcobalaminemic Greyhounds (serum cobalamin concentrations below the lower limit of the reference interval [RI]: 251-908 ng/L) were identified and investigated for the proportion of dogs with concurrent hypofolatemia (serum folate concentration $< 7.7 \mu\text{g/L}$, RI: 7.7-24.4 $\mu\text{g/L}$) by calculating the odds ratio (OR) and 95% confidence interval (CI). Also, serum samples from 44 Greyhounds, submitted to the same laboratory between October 2012 and March 2013, were used to measure serum HCY concentrations (RI: 5.0-22.1 $\mu\text{mol/L}$) by gas chromatography/mass spectrometry. A Mann-Whitney *U* test was used to compare serum HCY concentrations in hypcobalaminemic and hypofolatemic Greyhounds with those in normocobalaminemic and normofolatemic Greyhounds. A $p < 0.05$ was considered significant.

In this study, hypofolatemia was more frequently observed in hypcobalaminemic Greyhounds than in normocobalaminemic Greyhounds (OR [CI]: 1.8 [1.2-2.6], $p = 0.0064$). Although not significant, serum HCY concentrations were higher in hypcobalaminemic and hypofolatemic Greyhounds ($n = 12$, median: 42.7 $\mu\text{mol/L}$) compared to normocobalaminemic and normofolatemic Greyhounds ($n = 32$, median: 27.8 $\mu\text{mol/L}$; $p < 0.0670$). Hyperhomocysteinemia was detected in 11 (92%) hypcobalaminemic and hypofolatemic Greyhounds and 19 (59%) normocobalaminemic and normofolatemic Greyhounds.

Hypocobalaminemia in Greyhounds was associated with hypofolateamia, and increased serum HCY concentrations were observed in hypocobalaminemic and hypofolatemic Greyhounds, but also in Greyhounds with normal serum concentrations of both vitamins, suggesting a lack of these vitamins at the intracellular level. The functional implication of these findings in Greyhounds warrants further studies.

Conflicts of interest: No conflicts of interest reported.

ESCG-O-3
PREVALENCE OF CLOSTRIDIUM PERFRINGENS ENTEROTOXIN AND CLOSTRIDIUM DIFFICILE TOXIN A/B IN DOGS WITH IDIOPATHIC ACUTE HAEMORRHAGIC DIARRHOEA SYNDROME. S. Unterer¹, J. Suchodolski², K. Busch¹, Y. Minamoto², J. Steiner², R.S. Müller¹, K. Hartmann¹. ¹Clinic of Small Animal Medicine, Ludwig-Maximilians-Universität Munich, Germany, MUNICH, Germany, ²Gi Lab, Texas A&M University, COLLEGE STATION, United States of America

An association between clostridial pathogens and idiopathic acute haemorrhagic diarrhoea syndrome (AHDS) in dogs has been described. However, clinical relevance of the presence of *Clostridium* (*C.*) spp. and their toxins in feces has not been evaluated so far. Thus, the aim of this study was to evaluate association between severity of clinical signs and presence of *C. perfringens* enterotoxin (CPE) and *C. difficile* toxin A/B (CDT-A/B) in feces of dogs with AHDS.

Fecal samples of 54 dogs with idiopathic AHDS were tested by qualitative CPE and CDT-A/B ELISA. In addition, genotyping was performed to detect enterotoxin genes of *C. perfringens* (*cpe*) and toxin B genes of *C. difficile* (*tcdB*). Severity of clinical signs, duration of hospitalization, mortality rate, and selected laboratory parameters (hematocrit, banded neutrophils, bilirubin, albumin) were compared between dogs with toxin-positive and -negative as well as toxin gene-positive and -negative fecal samples.

CPE was found in 13/54 (24.1%) fecal samples of dogs with idiopathic AHDS, *cpe* in 35/54 (64.8%), and both CPE and *cpe* in 13/54 (24.1%). CDT-A/B was detected in 2/54 (3.7%), *tcdB* in 10/54, and both CDT-A/B and *tcdB* in 1/54 (1.9%) dogs. No significant difference could be detected in any parameter evaluated, neither between CPE-positive and -negative, nor between *cpe*-positive and -negative dogs.

The results of this study suggest that CPE and CDT-A/B play no significant role in dogs with idiopathic AHDS, and there is no value of measuring these toxins to predict severity or outcome of the disease.

Conflicts of interest: No conflicts of interest reported.

ESCG-O-4
A PILOT STUDY ON THE EFFECT OF PROBIOTIC TREATMENT ON GENE EXPRESSION IN INTESTINAL BIOPSIES FROM DOGS WITH FOOD-RESPONSIVE CHRONIC ENTEROPATHY. S. Schmitz, B. Glanemann, O.A. Garden, D. Werling, K. Allenspach. Royal Veterinary College, NORTH MYMMS, United Kingdom

Canine chronic enteropathies (CE) are thought to be partially caused by an aberrant immune response towards the intestinal microbiome. In humans and mice, administration of probiotics can alleviate colitis severity and/or prevent relapse by induction of a more "tolerant" microenvironment. *In-vitro* effects of probiotic *Enterococcus faecium* NCIMB 10415 E1707 (EF) have been demonstrated both in canine whole blood and *ex-vivo* cultured canine duodenal biopsies. The aim of this study was to investigate the *in-vivo* effects of EF on gene expression in duodenal and colonic biopsies of dogs diagnosed with food-responsive CE (fCE). Dogs were recruited to receive EF in a double-blinded, placebo-controlled manner in addition to an exclusion diet (hydrolysed protein). Duodenal and colonic biopsies were taken

before and 6 weeks after start of treatment. RNA extraction from biopsies and reverse-transcriptase quantitative PCR was performed in a SYBRGreen-based assay for the following genes: TLR2/4/5/9, IL-17A, IL-22, IL-23p19, RORC, IL-2, IL-12p35, TNF α , IL-4, IFN γ , IL-10, TGF β , IL-1 β , IL-18, NLRP3, casp-1 and PPAR γ ; using a 10-fold dilution of plasmid controls for each gene to assess assay efficiency. Relative quantification of gene expression was performed using four reference genes for the duodenum and 5 for the colon. For comparison of gene expression before/ after treatment and between treatment groups, Wilcoxon matched-pairs signed rank test and Mann Whitney tests were performed, respectively. Significance was set at $p < 0.05$.

Twelve dogs finished the clinical trial, 7 received EF and food (EFF) and 5 placebo (PF). Six weeks after the beginning of treatment, significantly reduced gene expression was found only in duodenal tissues of the PF group for TLR4, TNF α , RORC, IL-1 β , IL-18 and PPAR γ . In the colon, significant increases in the expression of all tested genes apart from Th17- and Th1 cell cytokines and TGF β , RORC and casp-1 were seen only in the PF group.

This is the first study demonstrating a profound and site-specific effect of treatment in canine fCE. Down-regulation of several genes of the innate and adaptive immune system were found in the duodenum, whereas gene expression for nearly all examined genes was increased in colonic tissues of the placebo group, despite clinical improvement seen in all dogs. However, the number of dogs in this study was comparably small, making a bias due to sample size possible. Similar investigations, complemented by the assessment of protein expression, should be performed in a larger number of dogs suffering from different types of CE.

Conflicts of interest: The work presented here is part of a PhD supported by Protexin Veterinary Ltd (Somerset, UK) and the BBSRC.

ESCG-O-5
THE ROLE OF SIGNAL TRANSDUCER AND ACTIVATION OF TRANSCRIPTION (STAT) 3 IN DOGS WITH CHRONIC ENTEROPATHIES. N. Luckschander¹, U. Reichart², B. Richter³, K. Allenspach⁴, E. Haas¹, A. Bilek¹, M. Müller⁴, J.G. Thalhammer¹. ¹Internal Medicine, VIENNA, Austria, ²Institute for Animal Breeding and Genetics, Biomodels Austria, VIENNA, Austria, ³Institute of Pathology and Forensic Medicine, University of Veterinary Medicine, VIENNA, Austria, ⁴Department of Clinical Sciences and Services, The Royal Veterinary College, HATFIELD HERTS, United Kingdom

Chronic intestinal inflammatory disorders, such as inflammatory bowel disease (IBD), are observed with increasing frequency in canine patients. The molecular pathogenesis of canine IBD is multifactorial and seems to involve similar pathogenetic mechanisms as in human IBD and mouse models of human IBD. Despite the multifactorial pathogenesis of this disease, various cytokines are expressed correlating to the grade of tissue inflammation and/or damage providing the opportunity to find markers for the disease outcome. In human IBD and murine IBD models the transcription factor STAT3 is activated in epithelial cells by interleukin 22, IL-6 and IL-10 and contributes substantially to wound healing and epithelial regeneration. Although the expression and activation of STAT3 in the intestinal mucosa in human chronic enteropathy patients as well as in mouse models is well studied there are no data available in dogs.

Thus the aim of this project was to investigate the role of STAT3 in canine IBD.

Ten dogs with chronic gastrointestinal signs were included into this prospective study. Two dogs which were euthanized due to non gastrointestinal diseases and 2 Beagle dogs served as controls. The patients were assessed given a clinical score using the CIBDAI and CCECAI scoring system and underwent gastrointestinal endoscopy. Using mucosal biopsy specimens from duodenum, immunohistological stainings were performed to detect phosphorylated STAT3.

Activated STAT3 was found to be exclusively expressed in non-epithelial cells, e.g. macrophages and lymphocytes in the intestinal stroma. In human IBD patients and in IBD mouse models, activated STAT3 is mainly found in epithelial cells and

is a hallmark of the inflammation and regeneration process in the intestinal epithelium. This discrepancy could be due to the fact that in canine IBD, the epithelium is largely intact and regeneration of the epithelium may play a minor role in the pathogenesis of the disease.

Conflicts of interest: No conflicts of interest reported.

ESCG-O-6
DEVELOPMENT AND VALIDATION OF A SIMPLIFIED ENDOSCOPIC ACTIVITY SCORE FOR CANINE CHRONIC ENTEROPATHIES. J.E. Slovak, C. Otoni, C. Wang, A.E. Jergens. Iowa State University, Ames, United States of America

Endoscopy is the gold standard to evaluate chronic enteropathies but the reliability of the assessment of endoscopic lesions is unclear. Previous studies have provided conflicting results as to the benefit of endoscopic activity in the diagnosis and management of intestinal disease. Moreover, mucosal criteria have varied between trials which may create confusion and confound accurate interpretation between endoscopists. The aims of the present study were to develop and validate an endoscopic index for assessing the severity of chronic enteropathy and to compare the role of operator experience in mucosal interpretation.

In total, 95 endoscopic images of inflammatory and normal mucosa from dogs with chronic enteropathies were displayed to 3 expert and 5 novice endoscopists. Each picture was assessed independently by the endoscopist for inflammatory changes using established indices (ie, hyperemia, granularity, friability, lymphatic dilation, erosions, masses) or interpreted as normal mucosa. Expert endoscopists were defined as individuals with advanced clinical training and active operator participation in gastrointestinal endoscopy over the preceding 24 months. Novice endoscopists had minimal endoscopic training or lacked consistent endoscopic operator experience over the same 24 month period. A mixed effects logistic regression model used mean values to compare agreement between novice and expert endoscopists in each organ. A *P* value < 0.05 was regarded as indicating statistical significance.

The agreement rate between expert or novice endoscopists was excellent or good for recognition of 6 out of 7 endoscopic signs or patterns (granularity, friability, erosions, lymphatic dilation, masses, normal mucosa), but was poor for hyperemia. While significant differences in mucosal assessment of the stomach (*P* = 0.17) and colon (*P* = 0.89) between operator groups were not observed, a significant difference (*P* < 0.007) was observed for the small intestine. Repeat duodenal assessment aided by use of a test set of representative mucosal lesions improved the overall scores of novice endoscopists to near that of experienced endoscopists (*P* = 0.053). For both operator groups, duodenal hyperemia showed the greatest chance (*P* < 0.021) of inter-observer disagreement as compared to mucosal abnormalities in other organs.

Our results indicate that trained operators can identify most endoscopic lesions of mucosal inflammation. A reliable overall scoring of endoscopic severity should be based on the presence of granularity, friability, lymphatic dilation, erosions and masses but not hyperemia. Novice endoscopists can identify some but not all well-defined endoscopic appearances but improve with operator experience.

Conflicts of interest: No conflicts of interest reported.

ESCG-O-7
SERUM PANCREAS-SPECIFIC LIPASE CONCENTRATIONS IN DOGS WITH UPPER GASTROINTESTINAL FOREIGN BODIES. M.R. Trehy¹, D. Batchelor¹, P.J. Noble¹, P. Silvestrini¹, C.M. Elwood², I. Battersby², F. Adam³, A.J. German¹. ¹Small Animal Teaching Hospital, NESTON, United Kingdom, ²Davies Veterinary Specialists, HIGHAM GOBION, United Kingdom, ³North Downs Specialist Referrals, BLETCHINGLEY, United Kingdom

Upper gastrointestinal foreign bodies and acute pancreatitis have a similar clinical presentation and rapid distinction between

the two conditions is important in emergency cases. One diagnostic option is measurement of serum canine pancreas-specific lipase (cPLI), the most sensitive and specific biomarker of pancreatic inflammatory disease. However, data regarding its performance in clinical settings are sparse. Our aim was to assess serum cPLI concentrations in dogs with upper gastrointestinal foreign bodies.

In this retrospective, observational, cohort study, case records of dogs referred to three referral centres (January 2007 to December 2012) were reviewed. Dogs were included if a gastric or small intestinal foreign body had been removed at coeliotomy, and serum had been submitted for cPLI measurement. Signalment, presenting clinical signs, clinicopathological and diagnostic imaging findings, foreign body location and outcome were recorded.

In total, 61 medical records were retrieved and 36 dogs met the inclusion criteria. Of these 36 dogs, 28 (group A) had increased PLI (13 with Spec cPL[®] >400 µg/l; 2 with Spec cPL[®] 201-400 µg/l; 13 with an abnormal SNAP[®]cPL[TRADEMARK]), and 8/36 dogs had a normal cPLI (group B). There was no difference between groups in signalment, presenting signs, foreign body location, or survival to discharge. A diagnosis of concurrent pancreatitis, based on clinical or sonographic findings, was made in 2/28 dogs in group A.

Conclusion: Serum cPLI frequently is increased in dogs with upper gastrointestinal foreign bodies. When increased cPLI is detected, gastrointestinal foreign body must remain a differential diagnosis, and should be excluded by appropriate diagnostic imaging.

Conflicts of interest: No conflicts of interest reported.

ESCG-O-8
IDIOPATHIC ACUTE HEMORRHAGIC DIARRHOEA SYNDROME IN DOGS: 108 CASES. F. Mortier, T.-E. Yoo, K. Strohmeier, K. Hartmann, S. Unterer. Ludwig-Maximilians-Universität, MUNICH, Germany

No prospective descriptive studies including large numbers of dogs with idiopathic acute hemorrhagic diarrhoea syndrome (AHDS) are published so far. The aim of this study was to describe signalment, history, clinical signs and laboratory values in dogs with AHDS.

One hundred and eight dogs were prospectively enrolled. Inclusion criterion was acute hemorrhagic diarrhoea (< 3 days). Dogs with any disease known to cause bloody diarrhoea were excluded. Clinical assessment was performed by calculation of the 'AHDS activity index' (maximum index 18). The hospital population served as control group.

Dogs with AHDS had a significantly lower median body weight (9.8 kg) and age (5 years) compared to control dogs (20 kg; 10 years) (*p* < 0.001). No sex predilection could be observed. Predisposed breeds were Yorkshire Terrier, Miniature Pinscher, Miniature Schnauzer, Miniature Poodle, and Maltese dogs. The syndrome was more likely to occur during the winter months (*p* < 0.001). Vomiting preceded the onset of bloody diarrhoea in 80% of cases. Median AHDS index was 12 (range 3-17). Hematocrit was generally high (mean 56%; range 33%-76%), but exceeded 60% only in 31% of dogs. Hematocrit of 48% of dogs was above the reference range, as was monocyte (50%), segmented (60%) and band neutrophil count (45%).

AHDS occurs more commonly in middle-aged small breed dogs and during winter. Hematocrit was elevated in 48% of cases, but did not exceed 60% in two thirds of dogs.

Conflicts of interest: No conflicts of interest reported.

ESCG-O-9

PROTEIN-LOSING ENTEROPATHY COMMONLY CO-EXISTS WITH HIGH FECAL FAT OUTPUT IN GERIATRIC CATS WITH IDIOPATHIC MALABSORPTION AND PERSISTS FOLLOWING CORRECTION OF SUBNORMAL SERUM COBALAMIN CONCENTRATION. D. A. Williams¹, G. Czarnecki-Maulden². ¹University of Illinois, URBANA, United States of America, ²Nestle Purina PetCare, 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 primary objective of this study was to determine if this common malabsorptive enteropathy in geriatric cats is associated with enteric protein loss as assessed by assay of fecal α_1 -proteinase inhibitor (α_1 -PI). A secondary objective was to determine if correction of subnormal serum cobalamin concentration improved any observed abnormalities in fecal α_1 -PI.

The study evaluated 15 cats older than 12 years of age with increased fecal fat (>20%) but without exocrine pancreatic insufficiency (EPI) as assessed by assay of serum trypsin-like immunoreactivity (fTLI). Enteric protein loss was assessed by species-specific immunoassay of fecal α_1 -PI as recently described (Burke KF et al. Vet. J. 2012 <http://dx.doi.org/10.1016/j.tvjl.2012.09.019>). Serum cobalamin and fTLI were assayed by competitive binding assay, both performed through the GI Laboratory at Texas A&M University and evaluated using reference ranges derived by that laboratory. Serum albumin was assessed using routine serum biochemical analysis.

All cats had abnormally high fecal fat (>20%) but serum fTLI was above the lower limit of the reference range, eliminating EPI as the cause of the steatorrhea. Serum cobalamin was subnormal (<290 ng/L) in 4 of the 15 cats, while fecal α_1 -PI was above the reference range (> 1.6 μ g/g) in 11 cats (range 2.3 to 12.6 μ g/L). Two months after cobalamin supplementation was initiated serum cobalamin was above the lower limit of the reference range in all cats, but fecal α_1 -PI was abnormally high (range 2.0 to 11.1 μ g/L) in 13 of the 15 cats. All cats had normal serum albumin concentrations at all time points evaluated.

It is concluded that the enteropathy characterized by nutrient malabsorption in geriatric cats is also associated with enteric protein loss as assessed by fecal α_1 -PI concentration. Serum albumin remained normal despite protein-losing enteropathy in the cats evaluated in this study, and correction of subnormal serum cobalamin concentrations in these cats did not improve abnormal fecal α_1 -PI concentrations.

Conflicts of interest: David Williams is a consultant with, and founder of, the GI Laboratory that performed some of the assays utilized in this study.

ESVE-O-1

EVALUATION OF INSULIN-LIKE GROWTH FACTOR 1 (IGF-1), TOTAL THYROXINE (TT4), FELINE PANCREATIC LIPASE IMMUNOREACTIVITY (fPLI) AND URINARY CORTICOID CREATININE RATIO (UCCR) IN CATS WITH DIABETES MELLITUS IN SWITZERLAND AND THE NETHERLANDS. S. Schäfer¹, H.S. Kooistra², A. Künzle¹, K. Macha¹, S. Moser¹, M. Prins³, J. Schmid¹, R. Sprecher¹, J. Suchodolski³, J.M. Steiner³, C.E. Reusch¹. ¹Vetsuisse Faculty, University of Zurich, ZURICH, Switzerland, ²Faculty of Veterinary Medicine, Utrecht University, UTRECHT, The Netherlands, ³College of Veterinary Medicine and Biomedical Sciences, Texas A&M University, TEXAS, United States of America

It is assumed that approximately 80% of diabetic cats have type 2 diabetes mellitus (DM) and that in the remaining cats diabetes is caused by underlying diseases. Recent studies suggest, that the percentage of so-called secondary diabetic forms, especially DM due to acromegaly and pancreatitis, is much higher than previously suspected. The aim of this study was to evaluate circulating concentrations of IGF-1, TT4, fPLI and the UCCR

as an estimate for the prevalence of acromegaly, hyperthyroidism, pancreatitis and hypercortisolism in DM cats in two different countries.

Private veterinarians in Switzerland (CH) and the Netherlands (NL) were asked to provide blood and urine samples from diabetic cats treated with insulin for at least 4 weeks. IGF-1 was analyzed by IGFBP blocked RIA, TT4 by Immulite[®] 1000, fPLI by ELISA Spec fPL and UCCR by in house RIA for urine corticoids. IGF-1 > 1000 ng/dl was considered suspicious for acromegaly, the respective cut-off values for hyperthyroidism and pancreatitis were TT4 > 45 nmol/l and fPLI > 5.3 μ g/l. Hypercortisolism was considered to be excluded when UCCR < 42 x 10⁻⁶.

Blood samples were available from 225 cats, urine samples collected at home were available from 119 cats.

IGF-1 concentrations ranged from 15-2471 ng/ml (median 584), 17.8% of the cats had IGF-1 > 1000 ng/ml (CH 12.5%, NL 21%). TT4 concentrations ranged from 6.4-143 nmol/l (median 23), 4.5% of the cats had elevated TT4 concentrations (CH 2.5%, NL 5.9%). fPLI ranged from 0.7-51 μ g/dl (median 4.4) and was elevated in 43% of the cats (CH 39%, NL 46%). UCCR ranged between 0.4-139 x 10⁻⁶ (median 17.6 x 10⁻⁶); in 85% of cats UCCR was < 42 x 10⁻⁶ (CH 91%, NL 84%). These findings did not differ significantly between countries.

The prevalence of increased IGF-1 was lower than previously reported. Since a high IGF-1 does not necessarily reflect acromegaly, CT studies are underway to evaluate the cats for a mass in the pituitary gland. The prevalence of an increased TT4 was comparable to the prevalence of hyperthyroidism in the non-diabetic elderly cat population. Increased UCCR was a relatively frequent finding. However, as none of the cats showed appropriate clinical signs, we hypothesize that hypercortisolism is a rare cause of feline DM. The increased fPLI in 43% of the cats may reflect subclinical pancreatitis, emerging during the course of DM.

Conflicts of interest: No conflicts of interest reported.

ESVE-O-2

ROUTINE KIDNEY PARAMETERS, GLOMERULAR FILTRATION RATE AND URINARY CYSTATIN C IN CATS WITH DIABETES MELLITUS, CATS WITH CHRONIC KIDNEY DISEASE AND HEALTHY CATS. D. Paeppe¹, L.F.E. Ghys², P. Smets², H.P. Lefebvre³, S. Croubels², J. Delanghe⁴, E. Meyer³, S. Daminet². ¹University of Ghent, MERELBEKE, Belgium, ²Faculty of Veterinary Medicine, Ghent University, MERELBEKE, Belgium, ³école nationale de vétérinaire de Toulouse, TOULOUSE, France, ⁴Faculty of Health Medicine and Life Sciences, Ghent University, GHENT, Belgium

Diabetes mellitus (DM) is increasingly common in humans and cats. Diabetic nephropathy is a frequent and serious complication in human diabetic patients, but little is known in cats.

This study compared kidney function between cats with DM, cats with chronic kidney disease (CKD) and age-matched healthy cats by measuring routine kidney parameters [serum creatinine (sCreat), serum urea (sUrea), urinary specific gravity (USG), urinary protein-to-creatinine ratio (UPC)], glomerular filtration rate (GFR) and urinary cystatin C (uCysC). Additionally, these parameters were compared between recently (<1 month) and not recently diagnosed diabetic cats.

Thirty-six cats with DM (19 recently, 17 not recently diagnosed), 10 cats with CKD and 10 healthy control cats were prospectively recruited. GFR was evaluated by exo-iohexol clearance in 17 diabetic, all CKD and all healthy cats. In all cats but two diabetic cats, uCysC was measured with a human particle-enhanced nephelometric immunoassay, validated to measure feline cystatin C, and expressed as uCysC/urinary creatinine ratio (uCysC/uCreat). Statistical analysis was performed using ANOVA.

Diabetic cats had significantly lower sCreat (mean \pm SD 120 \pm 43 versus 243 \pm 80 μ mol/L) and uCysC/uCreat (6 \pm 31 versus 173 \pm 242 mg/mol) and significantly higher USG (1.033 \pm 0.012 versus 1.018 \pm 0.006) and GFR (2.0 \pm 0.7 versus 0.8 \pm 0.3 mL/min/kg) compared with CKD cats. Compared with healthy cats, diabetic cats only had significantly lower USG

(1.033 ± 0.012 versus 1.046 ± 0.008). None of the evaluated parameters significantly differed between recently and not recently diagnosed diabetic cats.

In conclusion, based on the evaluation of routine kidney parameters, GFR and uCysC as tubular marker, feline DM has no negative impact on kidney function.

Conflicts of interest: No conflicts of interest reported.

ESVE-O-3

INVESTIGATION OF BREED DIFFERENCES AND DLA GENETIC INFLUENCE ON DEVELOPMENT OF ANTI-INSULIN ANTIBODIES IN DIABETIC DOGS. B. Catchpole¹, A.L. Holder¹, W.E.R. Ollier², L.J. Kennedy². ¹Royal Veterinary College, HATFIELD, United Kingdom, ²Centre for Integrated Genomic Medical Research, University of Manchester, MANCHESTER, United Kingdom

Diabetes mellitus is a common endocrine disease in dogs, which requires administration of exogenous insulin to control hyperglycaemia. Insulin therapy can stimulate an immune response and some dogs develop anti-insulin antibodies (AIA). Generation of an immune response would require antigen presentation of insulin peptide epitopes by major histocompatibility complex (MHC) molecules, which in dogs, are encoded by the dog leukocyte antigen (DLA) genes. The hypothesis for the current study was that breed differences in AIA reactivity exist that might reflect a DLA genetic influence on antibody responses to insulin therapy.

Blood samples from diabetic dogs were recruited via the UK Canine Diabetes Register, and sera were made available for research following completion of routine diagnostic testing, with informed owner consent. Indirect ELISA against bovine or porcine insulin was used to determine the AIA status in newly-diagnosed diabetic dogs (n = 109) and diabetic dogs treated with Caninsulin[®] (n = 594), Insuvel[®] lente (n = 348) or Insuvel[®] PZI (n = 50). DLA alleles and haplotypes for all diabetic dogs were determined by sequence-based typing for DLA-DRB1, -DQA1 and -DQB1 loci.

Only three of the 109 newly-diagnosed diabetic dogs were AIA positive. Significantly greater insulin reactivity was seen in insulin-treated diabetic dogs. Fifty two per cent of diabetic dogs treated with Insuvel lente were AIA positive, compared to 12% of dogs treated with Caninsulin, suggesting that bovine insulin is more immunogenic than porcine insulin. The greatest insulin reactivity was seen in Insuvel PZI treated dogs, with 74% being AIA positive, suggesting that addition of protamine enhances insulin immunogenicity. Breeds such as the dachshund, Cairn terrier, miniature schnauzer and Tibetan terrier were more likely to develop AIA, whereas cocker spaniels were less likely to seroconvert during insulin therapy.

Three DLA haplotypes appeared to show an association with AIA status. DLA-DRB1*00901/DQA1*00101/DQB1*008011 was associated with being AIA positive in Insuvel lente-treated dogs (p < 0.05), whereas DLA-DRB1*001501/DQA1*00601/DQB1*02301 was associated with being AIA positive in Caninsulin-treated dogs (p < 0.01). DLA-DRB1*00601/DQA1*005011/DQB1*00701 showed an association with being AIA negative (p < 0.05). These results suggest that DLA genes might influence AIA responses in treated diabetic dogs and that inter-breed variability in the DLA haplotypes expressed, results in breed differences in immune responses to insulin therapy.

Conflicts of interest: This study was undertaken by ALH as part of an MRes degree, which was co-funded by MSD Animal Health and The Kennel Club Charitable Trust, as part of their sponsorship of the UK Companion Animal Diabetes Register. Neither funding body had any involvement in the conduct of the study or the data analysis that could inappropriately influence or bias the content of the paper.

ESVE-O-4

SF-1 EXPRESSION IN CANINE CORTISOL-SECRETING ADRENOCORTICAL TUMORS. M.F. van den Berg, M.J. Kool, J.A. Mol, H.S. Kooistra, S. Galac. Faculty of Veterinary Medicine, Utrecht University, UTRECHT, The Netherlands

Hypercortisolism due to an adrenocortical tumor (AT) is characterized by ACTH-independent hypersecretion of cortisol and uncontrolled tumor growth. In lack of explanations, both processes are still characterized as autonomous.

The transcription factor SF-1 plays an important role in adrenal development and physiology. It is an obligate activator of steroidogenic enzymes and essential for adrenocortical proliferation. Studies in human adrenocortical cell cultures and transgenic mice have demonstrated that an increased SF-1 dosage stimulates adrenocortical cell proliferation and triggers tumor formation in mice. It has been suggested that modulation of SF-1 activity may represent an important therapeutic target in ATs.

We report on a screening of 37 canine cortisol-secreting ATs (26 carcinomas and 11 adenomas) for the mRNA expression of SF-1 (quantitative RT-PCR) and describe the association of SF-1 mRNA expression with malignancy and clinical outcome. In addition, protein expression and SF-1 localization were determined by immunohistochemistry (IHC). Fifteen normal canine adrenals served as controls.

The results of mRNA expression analysis demonstrated a significant upregulation (P = 0.03) of SF-1 in carcinomas, whereas the mRNA expression of adenomas did not differ significantly from that of normal adrenals. IHC staining of SF-1 was predominantly nuclear through the whole adrenal cortex and in all ATs. The staining pattern between adenomas and carcinomas did not differ. In 6 out of the 17 dogs, which were available for follow-up, there was recurrence of clinical signs of hypercortisolism within 2 years after adrenalectomy. Besides biochemically confirmed hypercortisolism, in 5 of the 6 dogs metastasis were detected by diagnostic imaging. In this group of dogs, SF-1 mRNA expression was significantly higher (P = 0.05) than in dogs which were disease-free at least 2 years after adrenalectomy.

In conclusion, significant upregulation of SF-1 in adrenocortical carcinomas and the association between high SF-1 levels and poor outcome post adrenalectomy, suggest a role of SF-1 in the pathogenesis of canine ATs. Further research focusing on pharmacological modulation of SF-1 activity is warranted and might provide a new therapeutic approach in adrenal hypercortisolism.

Conflicts of interest: No conflicts of interest reported.

ESVE-O-5

ACTIVATING MUTATIONS OF GNAS IN CANINE CORTISOL-SECRETING ADRENOCORTICAL TUMORS. M.M.J. Kool, S. Galac, C.G. Spandauw, H.S. Kooistra, J.A. Mol. Utrecht University, UTRECHT, The Netherlands

Cushing's syndrome or hypercortisolism is one of the most common endocrinopathies in dogs. Adrenal hypercortisolism is a result of an adrenocortical adenoma or carcinoma and represents about 15% of cases of spontaneous hypercortisolism in dogs. It is characterized by the adrenocorticotropin (ACTH)-independent cortisol hypersecretion. In lack of explanations, the mechanism leading to hypersecretion of cortisol is characterized as autonomous.

In the normal adrenal cortex, cortisol production is regulated by melanocortin 2 receptor (MC2R) signaling. Upon ACTH binding to the MC2R, the stimulatory G protein alpha subunit (G α) activates adenylyl cyclase, producing cyclic AMP (cAMP). This, in turn, activates protein kinase A (PKA), resulting in target gene transcription through activated transcription factors and the subsequent initiation of steroidogenesis.

The aim of the present study was to investigate whether ACTH-independent hypersecretion of cortisol in canine ATs may result from aberrant activation of the MC2R-cAMP-PKA signaling pathway. Mutation analysis was performed of the genes encoding the stimulatory G protein alpha subunit (*GNAS*), *MC2R* and PKA regulatory subunit 1A (*PRKARIA*) in 44 cortisol-secreting ATs (14 adenomas and 30 carcinomas), derived from dogs diagnosed with ACTH-independent hypercortisolism.

Mutation analysis of *GNAS* showed the presence of 7 different missense mutations, all known for their ability to constitutively activate the pathway. Activating mutations were present in 14 of 44 ATs and were located in codon 201 (8 carcinomas and 3 adenomas), in codon 203 (1 adenoma) and in codon 227 (2 carcinomas). No functional mutations were found in *MC2R* and *PRKARIA*.

We conclude that activation of cAMP signaling by means of an activating *GNAS* mutation is a frequent event in canine cortisol-secreting ATs and is likely to play a crucial role in both ACTH-independent cortisol production and tumor formation. To our best knowledge, this is the first report of potentially causal mutations in canine cortisol-secreting ATs.

Conflicts of interest: No conflicts of interest reported.

ESVE-O-6

INCIDENTAL ADRENAL LESIONS: ULTRASONOGRAPHIC FINDINGS IN 3748 DOGS (2007-2010). A.K. Cook. Texas A&M College of Veterinary Medicine and Biomedical Sciences, COLLEGE STATION, United States of America

Incidental adrenal lesions (IALs) are routinely identified on imaging studies in canine patients. This study describes the prevalence of IALs in dogs undergoing ultrasonographic (US) examinations by veterinary radiologists. Patient demographics and outcomes are also reported.

Reports of US exams performed at Texas A&M Veterinary Medical Teaching Hospital between January 2007 and June 2010 were reviewed. Reports were excluded if a complete examination of the abdominal cavity was not performed, or if a scan had been performed within the previous 2 months. Adrenal glands were classified as abnormal if a nodule or mass was described, or if the width of either adrenal was ≥ 10 mm. Information regarding signalment, clinical findings, and outcome was collected from the medical record. Patients were determined to have an IAL if there was no clinical suspicion of adrenal disease prior to imaging at this institution and the adrenal abnormality appeared unrelated to on-going medical problems.

A total of 4,150 US examinations (performed on 3,748 dogs) met the inclusion criteria. An IAL was noted in 151 patients (4.0%), with distinct nodules/masses reported in 146 dogs and non-specific enlargement noted in 5. Lesions were noted in the left gland in 74 (49%), the right in 60 (39%) and both glands in 17 dogs (11%). Vascular invasion was noted in 10 patients. Genders were equally represented (75 female and 76 males). Body weights ranged from 2-56 kg, with a median of 21 kg. Age ranged from 3.5-16.9 years, with a median of 11.25 years. Adrenalectomy was performed in 6 dogs; diagnoses included adenoma (2), carcinoma (2), and pheochromocytoma (2). Necropsy examinations were performed in 14; findings included cortical hyperplasia (5), cortical adenoma (2), cortical carcinoma (1), pheochromocytoma (1), cortical atrophy (1), and grossly normal (4). Proven malignant masses ranged in size from 20-46 mm; benign lesions were all < 20 mm. Various contemporaneous conditions were reported, including malignancy in 43 (28%) of patients.

These findings suggest that IALs in dogs have a similar prevalence to that reported in human patients, and are often noted in dogs with concurrent neoplastic diseases. Based on this small data set, IALs are equally likely to be benign or malignant, although a mass ≥ 2 cm is probably malignant.

Conflicts of interest: No conflicts of interest reported.

ESVE-O-7

MANAGEMENT OF HYPERTHYROIDISM IN CLIENT-OWNED CATS WITH AN IODINE-RESTRICTED FOOD. M. van der Kooij¹, I. Becnárová², H.P. Meijer², E. Teske¹, H.S. Kooistra¹. ¹Utrecht University, UTRECHT, The Netherlands, ²Hill's Pet Nutrition, Europe, PRAGUE, Czech Republic

Feline hyperthyroidism is one of the most common endocrine disorders in middle-aged and elderly cats. Currently, three

treatment options are available: (i) anti-thyroid medication, (ii) thyroidectomy and (iii) radioiodine therapy. Recently, a new treatment option consisting of an iodine-restricted food was introduced in the management of feline hyperthyroidism. Previous studies showed that the food is a safe and effective method for decreasing circulating total thyroxine (TT4) concentration and managing hyperthyroidism in a research colony of cats¹. The objective of this study was to evaluate the effects of an iodine-restricted food on circulating TT4 concentrations and clinical parameters in client-owned hyperthyroid cats.

A prospective, multi-center, non-controlled, open-label study was performed. Two hundred and twenty-five hyperthyroid cats were enrolled in the study and were transitioned to the iodine-restricted food. Data from questionnaires completed by veterinarians and owners were recorded at weeks 0, 4 and 8, as well as circulating TT4, urea and creatinine concentrations. The study group consisted of 89 males and 136 females with a median age of 15 years (range 4-21 years). One group (n = 113) had been previously diagnosed and treated with anti-thyroid drugs, and the other group (n = 112) included newly diagnosed cats.

Circulating TT4 concentrations decreased to within the reference range at week 4 ($P < 0.0001$) and did not change significantly between weeks 4 to 8. Significantly more cats with an increased TT4 at week 8 had poor owner compliance and/or palatability compared to cats with a normalized TT4, indicating that these factors may be responsible for the less than desired result in these cats. Clinical parameters (vomiting, PU/PD, hyperactivity, polyphagia, weight loss, hair coat quality and quality of life) improved at 4 weeks ($P < 0.0001$). Circulating creatinine concentration significantly ($P = 0.001$) decreased from week 0 to week 4, which may be explained by the lower heat-processed meat content of the iodine-restricted food compared to regular cat food. No significant differences were observed between the previously diagnosed and newly diagnosed cats at any time-point. The most common reported reasons for incomplete data were poor palatability of the food and owner compliance. No short-term side effects were detected.

In conclusion, in client-owned cats with hyperthyroidism an iodine-restricted food is a valuable management option to normalize circulating TT4 concentrations and improve symptoms and signs of hyperthyroidism within 4 weeks.

Reference: Melendez LD *et al.*: Titration of dietary iodine for reducing serum thyroxine concentrations in newly diagnosed hyperthyroid cats. AbstractEN-16, ECVIM Forum Abstracts, 2011

Conflicts of interest: M. van der Kooij received a study grant from Hills Pet Nutrition Europe to complete this manuscript. Iveta Becnárová and Hein P Meyer are employees of Hills Pet Nutrition Europe. No further conflicts of interest are reported.

ESVE-O-8

SURVIVAL TIMES FOR CATS WITH HYPERTHYROIDISM TREATED WITH A FIXED LOW-DOSE OF IODINE 131I. M. Vagny¹, L. Desquilbet², E. Reyes-Gomez³, F. Delisle⁴, P. Devauchelle⁴, I. Rodriguez-Piñero⁴, D. Rosenberg⁴, P. de Fornel-Thibaud⁴. ¹CHV Fregis, ARCUEIL, France, ²Clinical Epidemiology and Biostatistics Unit, National Veterinary School Alfort, MAISONS ALFORT, France, ³Anatomical Pathology Unit, National Veterinary School of Alfort, MAISONS ALFORT, France, ⁴Micen Vet referral centre, CRETEIL, France

Hyperthyroidism is the most common endocrine disorder in geriatric cats. Several treatment modalities are used for this disease, including radioiodine (¹³¹I). Both individual ¹³¹I dose determination using radiotracer kinetic studies or scoring systems, and fixed relatively high ¹³¹I dose (i.e. 5.0 mCi) administration are effective and associated with prolonged survival times. The latter method is less complicated but has the potential to expose patients and veterinary personnel to unnecessary levels of radiation.

The aim of the study was to retrospectively evaluate the efficacy of a fixed low ¹³¹I dose (mean \pm SD: 3.3 mCi \pm 0.1) for treatment of hyperthyroid cats, assess outcome and identify factors associated with survival.

Medical records of one single ^{131}I referral centre were retrospectively examined. The following data were collected at diagnosis and at follow-up, when available: serum total thyroxine (TT4), urea, creatinine concentrations, ALPK and ALT activities. In addition, age, sex, weight at admission and at follow-up, reason for ^{131}I and any prior treatment with antithyroid drugs were recorded. Death, whatever the cause, was the event in survival analysis. Median survival time (MST) was determined by Kaplan-Meier product-limit method (admission as day 0). Multivariate Cox models were used to investigate independent risk factors for death.

Ninety-six cats were included in the study. Male to female ratio was 1:1.4, median age at admission was 13 years (interquartile range [IQR] 11-14). Median initial follow-up time was 7 weeks (IQR 5-9). At diagnosis, serum TT4 concentration, ALPK and ALT activities were above the reference range in 96/96, 29/56 and 52/65 cats, respectively. No cat had serum creatinine concentration above reference range at diagnosis. At follow-up, serum TT4 concentration, ALPK and ALT activities were below the upper range value in 94/96, 52/54 and 48/57 cats, respectively. Serum creatinine concentration above reference range was identified in 12/72 cats. MST was 3 years; the 1- and 2-year survival rates after ^{131}I therapy were 90% and 78%, respectively. Low body weight (≤ 3.1 kg; adjusted hazard ratio [aHR], 5.88; 95% confidence interval [CI], 2.22-16.67; $p < 0.01$) and male gender (aHR, 2.63; 95% CI, 1.01-7.14; $p = 0.04$) were independently associated with death, whereas age, prior treatment with anti-thyroid drugs, and azotemia were not.

This study suggests that a relatively fixed low ^{131}I dose treatment is effective for feline hyperthyroidism and that long-term survival can be achieved. Considering any cause of death, low body weight and male gender were significantly associated with shorter survival times.

Conflicts of interest: No conflicts of interest reported.

ESVE-O-9

PROGNOSTIC FACTORS IN DOGS WITH NEWLY DIAGNOSED PITUITARY DEPENDENT HYPERCORTISOLISM. S. Corradini, D. Floriano, F. Dondi, P.F.B. Famigli-Bergamini, F. Fracassi. University of Bologna, OZZANO DELL'EMILIA, Italy

Pituitary dependent hypercortisolism (PDH) is one of the most frequent endocrinopathies in dogs but prognostic factors are largely unknown. The aim of the present study was to determine the prognostic value of different clinical and clinicopathological variables evaluated at the time of diagnosis, before treatment, in dogs with PDH. Medical records from one referral center were retrospectively evaluated. The diagnosis of hypercortisolism was confirmed using the LDDS test and/or ACTH stimulation test in dogs with compatible clinical signs. The differentiation between adrenal dependent hypercortisolism and PDH was based on the ultrasonographic appearance of the adrenal glands, the results of the LDDS test and concentrations of endogenous ACTH. Only newly diagnosed dogs with PDH, subsequently treated with trilostane, were included. The following variables were entered into a univariate Cox Regression analysis to determine if they were associated with survival: age, sex, neutered status, clinical signs, concomitant diseases, hypertensive status, serum cortisol concentrations, routine biochemical and haematological parameters, urine specific gravity, urine protein:creatinine ratio (UPC), and urine albumin:creatinine ratio (UAC). Those that showed possible association ($p < 0.2$) with survival were then included in a stepwise multivariate Cox regression analysis. Statistical significance was set at $p < 0.05$. Eighty-two dogs were eligible for inclusion in the study. The median age at diagnosis was 10.1 years (range: 4.3-16.9). There were 36 male (5 castrated) and 46 female (19 spayed). Median survival time was 461 days (range: 8-3256). Variables related to survival were age, urea, potassium, phosphate, lymphocytes and basal serum cortisol concentrations. In the multivariable model survival was associated with serum potassium (OR: 2.08, CI95%: 1.24-3.49, $p = 0.006$) and phosphate (OR: 1.31, CI95%: 1.03-1.68, $p = 0.029$) concentrations. Hyperphosphatemia was observed in 30 cases (36.6%, median phosphate 5.5 mg/dl, range: 4.9-7.5) and hyperkalemia in 6 dogs

(7.3%, median potassium 5.8 mEq/L, range: 5.6-6.2). Phosphate concentrations were positively correlated with post-ACTH cortisol concentrations ($r = 0.33$, $p = 0.006$), alanine aminotransferase ($r = 0.37$, $p = 0.002$) and gamma glutamyl transferase activities ($r = 0.38$, $p = 0.001$) and negatively correlated with total calcium concentrations ($r = -0.40$, $p < 0.001$) and urinary specific gravity ($r = -0.35$, $p = 0.005$). Potassium concentrations were positively correlated only with serum alkaline phosphatase activity ($r = 0.29$, $p = 0.017$). We conclude that serum phosphate and potassium concentrations appear to be of prognostic importance in dogs with PDH. UPC appears unrelated to survival in canine PDH, unlike in dogs with chronic kidney disease, despite the marked proteinuria associated with this disease.

Conflicts of interest: No conflicts of interest reported.

ESVE-O-10

ASSOCIATION BETWEEN ACTH STIMULATION TEST, CLINICAL SIGNS, AND LABORATORY PARAMETERS IN DOGS WITH HYPERADRENOCORTICISM TREATED WITH TRILOSTANE. A. Wehner¹, S. Gloeckner¹, C. Sauter-Louis¹, D. Kruse², C. Stockhaus², K. Hartmann¹. ¹Ludwig Maximilian University, MUNICH, Germany, ²Small Animal Clinic Haar, HAAR, Germany

Trilostane is the medical treatment of choice in dogs with hyperadrenocorticism. Regular treatment monitoring is recommended, and dose adjustments are mainly based upon results of ACTH stimulation tests. However, a discrepancy between results of ACTH stimulation test and clinical response to treatment is sometimes observed. The aim of this study was to investigate the association between ACTH stimulation test results (baseline and ACTH-stimulated cortisol concentration) and clinical and laboratory parameters in dogs with hyperadrenocorticism currently treated with trilostane.

Based upon consultation of endocrinologic experts, a standard operating procedure protocol and a disease-specific questionnaire for owners concerning severity of clinical signs were developed for dogs with hyperadrenocorticism. Twenty five dogs with hyperadrenocorticism that were treated with trilostane were included. Sixtyone events in these dogs consisting of history, owner questionnaire, physical exam findings, complete blood count, biochemical profile, urinalysis, and ACTH stimulation test results were collected. Using spearman's rank correlation coefficient, correlations of baseline and ACTH-stimulated cortisol concentration with clinical and laboratory parameters were calculated.

While there was a correlation of baseline cortisol with ACTH-stimulated cortisol concentration ($r = 0.596$; $p < 0.001$, $n = 61$), there was no correlation of clinical signs and laboratory parameters neither with baseline nor with ACTH-stimulated cortisol concentration.

ACTH stimulation test does not reflect control of clinical signs. Therefore, dose adjustments of trilostane should not be based on results of ACTH stimulation tests. However, ACTH stimulation test is still the only method to assess overdosing of trilostane and risk of iatrogenic hypocortisolism.

Conflicts of interest: No conflicts of interest reported.

ESVE-O-11

THE MEASUREMENT OF 21-HYDROXYLASE ANTIBODIES IN DOGS VIA ENZYME-LINKED IMMUNOSORBENT ASSAY. M.R. Rick¹, K.R. Refsal¹, D.M. Callewaert², T. Rader². ¹Michigan State University, LANSING, United States of America, ²Oxford Biomedical Laboratories, ROCHESTER-HILLS, United States of America

Most cases of hypoadrenocorticism (HoAC) in humans result from immune-mediated adrenalitis. Commercially available assays, using recombinant human 21-hydroxylase (21-OH) as the antigen, are used as screening tools for the identification of anti-adrenal autoantibodies (ABs) in serum. Presence of these ABs

constitutes the primary criteria for an early diagnosis of adrenitis. In dogs, such a test is not commercially available. Using recombinant canine 21-OH as the antigen in Western Blot (WB) and enzyme-linked immunosorbent assay (ELISA), we previously were able to show that approximately 30% of dogs with naturally occurring primary HoAC had measurable ABs against 21-OH. A limitation of this study was small sample size. Further, the differences in optical densities (OD) in the ELISA between specific binding to 21-OH and nonspecific binding (NSB) were narrow. The aim of the present study was to decrease the NSB OD, without decreasing the specific OD, with the overarching goal to improve the performance of the 21-OH AB ELISA. To achieve this, sera from one dog that was immunized with canine 21-OH were used as negative (pre-immunization) and positive (post-immunization) controls. Using these, combinations of four different amounts of canine 21-OH for plate coating, 12 different blocking buffers, and eight different assay buffers were compared. Once the best combination was established, a total of 59 serum samples from dogs with HoAC, 20 serum samples from healthy dogs, and from dogs with hyperadrenocorticism receiving treatment with mitotane ($n = 31$) and trilostane ($n = 24$), were analysed. Coating ELISA plates using 0.1 M sodium bicarbonate, pH 9.6, containing 2.5 $\mu\text{g/mL}$ recombinant purified canine 21-OH, gave an adequate OD, and increasing its concentration did not increase the OD. Using a commercial non-mammalian protein blocking buffer significantly decreased NSB OD, without decreasing the specific OD. This positive effect was enhanced with the use of a particular proprietary assay buffer. Considering ODs above the calculated upper limit (MEAN+2SD) for healthy dogs as positive, 13 of the 59 dogs with HoAC were classified as true positives, and one healthy dog was classified as false-positive, showing that the ELISA had a high specificity (95%), but a low sensitivity (22%). Including dogs with hyperadrenocorticism in these calculations, numbers changed slightly (92 and 22%, respectively). These data suggest that the ELISA, in its current form, provides a high level of confidence in positive results. We therefore strongly believe that this assay has potential for assisting in the early diagnosis of adrenitis in dogs.

Conflicts of interest: Dr. Callewaert and Ms Rader are employed by a commercial biotechnology company.

ESVE-O-12

STABILISATION OF DOGS WITH PRIMARY HYPOADRENOCORTICISM, COMPARING ONCE DAILY VERSUS TWICE DAILY ORAL DOSING OF FLUDROCORTISONE

ACETATE. E. Roberts¹, L.A. Boden², I.K. Ramsey¹. ¹SMALL ANIMAL HOSPITAL, UNIVERSITY OF GLASGOW, GLASGOW, United Kingdom, ²University of Glasgow, GLASGOW, United Kingdom

Canine hypoadrenocorticism is routinely treated with long term fludrocortisone acetate in Europe. There has been little research on the pharmacokinetics of fludrocortisone and opinions vary as to the starting dose frequency. This study compared the stabilisation of three groups of dogs with primary hypoadrenocorticism: those starting and continuing on once daily (SID) dosing, those starting and continuing on twice daily (BID) dosing and those starting on SID and then switching to BID.

Cases of primary hypoadrenocorticism that had been confirmed by ACTH stimulation were included in this retrospective study providing they had been started on fludrocortisone, had follow up data available for more than 1 week (including electrolytes) and had records of dosage changes. The time taken (in months) to stabilise, and any changes to treatment, were recorded for each dog. Stabilisation was defined by normal electrolyte concentrations and clinical evaluation. The dosing regimen of fludrocortisone was determined by individual clinicians. Gender, age at diagnosis, breed predisposition and dose frequency were examined with Kaplan-Meier survival curves and in a multi-variable Cox proportional hazard model.

Four dogs started on SID dosing failed to stabilise at all during the study period including two dogs that were subsequently unsuccessfully switched to BID dosing. Thirty-three dogs remained eligible after these exclusions. The median time of the whole cohort to become stable after diagnosis was 4 months (interquartile range (IQR) 0-12 months). Dogs who were started and continued on SID dosing ($n = 18$) had a median stabilisation period of 4 months (IQR 0-14 months). Dogs that were started and continued on BID dosing ($n = 5$) had a median stabilisation period of 7 months (IQR 4-13 months). Ten dogs which failed to stabilise on SID dosing after a median time of 6 months (IQR 3-11 months) were switched to BID dosing and were subsequently found to stabilise at a median time of 0 months after the change of dose frequency. No dogs during the study were changed from BID to SID dosing. There was no significant difference between stabilisation times of the SID and BID groups. However once changed from SID to BID the stabilisation time was significantly quicker than either group. Breed, age and gender, included in the model as confounders, were not statistically significant.

The differences identified in this study suggest that larger studies would be useful. Dogs treated SID that do not stabilise quickly may be rapidly stabilised using BID dosing.

Conflicts of interest: No conflicts of interest reported.