

### NECROTIC ENTEROCOLITIS ASSOCIATED WITH *CLOSTRIDIUM PERFRINGENS* IN LORIKEETS (*TRICHOGLOSSUS HAEMATODUS*)

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**Introduction:** *Clostridium perfringens* is the cause of necrotic enteritis (NE) in chickens. A similar disease has occasionally been reported in psittacines. We diagnosed several outbreaks of NE in lorikeets (*Trichoglossus haematodus*).

**Materials and Methods:** A retrospective study in lorikeets with lesions similar to poultry NE necropsied between 2005 and 2018 was performed. Thirty-three out of the 65 lorikeets necropsied during this period showed lesions of NE. Analysed data included clinical histories, necropsy findings, histopathology and bacteriological analyses. In addition, PCR detection of toxin genes was performed on DNA extracted from formalin-fixed and paraffin wax-embedded (FFPE) intestinal lesions.

**Results:** NE was the most commonly diagnosed cause of death in lorikeets in our material. Lorikeets with NE showed severe fibrinonecrotizing enteritis and/or colitis. Bacteriology was performed in 19 out of the 33 lorikeets with NE and *C. perfringens* was isolated from intestinal samples of 15 lorikeets subjected to anaerobic culture. Toxin typing (genes *cpa*, *cpb*, *ctx*, *iap*, *cpb2* and *cpe*) was performed in nine lorikeets by PCR and the gene for the  $\alpha$ -toxin (*cpa*) was the only toxin gene detected. However, not all recently suggested toxin-typing genes were included in routine investigations. Additional PCRs to detect *C. perfringens* toxin genes including *netB* in FFPE intestinal samples are currently being undertaken.

**Discussion:** This study indicates that NE is an important cause of mortality in lorikeets and suggests that it is associated with *C. perfringens* enteric infection. Analysing the presence of additional genes is required to further classify *C. perfringens* according to the recently adapted toxin-typing scheme.

### FELINE MORBILLIVIRUS IN PIEDMONT: A PATHOLOGICAL AND MOLECULAR STUDY

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**Introduction:** Feline morbillivirus (FeMV) was recently described in domestic cats from Asia, America and Europe. Most of the positive cats are affected by lower urinary tract disease or nephritis. This study aimed to investigate the presence of FeMV infection in domestic cats from the Piedmont region (North-Western Italy) and, possibly, to correlate it with inflammatory kidney lesions.

**Materials and Methods:** Urine ( $n = 153$ ) and kidney ( $n = 50$ ) samples were collected by two veterinary clinics. Molecular investigations were performed by real time RT-PCR and positive samples were submitted for sequencing and phylogenetic analysis. Formalin-fixed and paraffin wax-embedded sections of kidney were examined by standard methods.

**Results:** Cats were mainly European breed, aged from 7 months to 20 years, of both sexes. FeMV RNA was detected in 8.5% of urine samples and in 8.0% of the tested kidneys. Phylogenetic analysis showed that the identified FeMV sequences cluster within a clade of German and Turkish isolates, not related to the first Italian strain, Piuma/2015. FeMV RNA was detected both in healthy cats (84.6%) and in cats with clinical signs compatible with urinary tract pathologies (15.4%). The histopathological investigation carried out on kidney tissues revealed the presence of tubulointerstitial nephritis in both positive (75.0%) and negative cats (68.0%).

**Discussion:** This study represents the first epidemiological investigation of FeMV infection in North-Western Italy. The prevalence of the virus seems to be low and not closely related to CKD or inflammatory kidney lesions.

### EFFECT OF CHESTNUT WOOD AND FLUBENDAZOLE ON MORPHOLOGY OF SMALL INTESTINE AND LYMPHOCYTES IN PERIPHERAL BLOOD, SPLEEN AND JEJUNUM IN CHICKENS

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**Introduction:** Flubendazole is an anthelmintic belonging to the benzimidazole group. The mechanism of action of flubendazole is disruption of microtubule structure and function. Tannins (chestnut extract) may reduce the number of gastrointestinal parasites in birds and have beneficial effects on the digestion. The aim of this experiment was to follow the effect of sweet chestnut on the morphology of small intestine and lymphocytes in peripheral blood, spleen and jejunum in chickens during application of Flimabend (flubendazole).

**Materials and Methods:** The experiment was conducted on a broiler chicken farm. Twenty-four chickens, 40 days old, were included in the trial. Chickens were randomly divided into four groups of six chickens each: Fli (Flimabend®), Far (Farmatan®), Far + Fli and C (control). Extract of sweet chestnut (Farmatan®) was added to water, for 6 h per day for 5 days starting at the age of 43 days. Chickens in the Fli group received Flimabend individually per os for 7 days starting at the age of 40 days.

**Results:** There was a mild increase in leucocytes, lymphocytes, monocytes, leucocyte common antigen CD45, IgM<sup>+</sup> and IgA<sup>+</sup> cells in peripheral blood after administration of Flimabend. Subpopulations of lymphocytes (CD3<sup>+</sup>, CD4<sup>+</sup>, CD8<sup>+</sup>, IgM<sup>+</sup>) were increased in the jejunum after application of the drug. Administration of Farmatan revealed the opposite effect in immunocompetent cells, proving an anti-inflammatory effect. Morphology of villi was also negatively influenced by administration of Flimabend. This suggests that administration of Flimabend induces a mild inflammatory process in the intestine.

**Discussion:** Administration of chestnut extract demonstrated a beneficial effect on systemic immunity during application of Flimabend.

### HISTOLOGICAL ASPECTS OF CORNEAL CROSS LINKING IN EQUINES

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**Introduction:** Despite advances in therapy of ocular disorders in horses, surgical intervention is frequently required, due to poor response to medical treatment. Corneal crosslinking (CXL), a recently introduced method of treatment in veterinary ophthalmology, induces new chemical bridges between proteins and other molecules within the corneal stroma, based on physicochemical reactions after exposure to riboflavin and ultraviolet light. In this study, histopathology of the treated cornea is used to evaluate the efficacy of the CXL procedure.

**Materials and Methods:** Fixation of the entire globe versus corneal tissue only and formaldehyde versus Davidson's fixative, were compared in eyes from three horses. Paraffin wax-embedded tissue sections were analysed after HE, PAS and Alcian blue staining, as well as immunohistochemistry using antibodies against laminin and collagen type I.

**Results:** Corneal tissue collapsed once removed from the globe before fixation and fixation in 10% neutral buffered formalin created microfolds, both hampering histological evaluation. Therefore, entire globe fixation in Davidson's fixative is the selected method of sample treatment. PAS-stained crosslinked corneas revealed a change in colour of the treated area, indicating a possible change in composition of extracellular matrix. The basement of corneal epithelium was clearly visible in the immunohistochemistry for laminin.

**Discussion:** Preliminary results in establishing a practical protocol to assess the penetration depth of CXL indicate that the protein-based staining methods seem useful; however, quantitative analyses might be needed to support these preliminary findings. Furthermore, a method to assess stromal cell necrosis needs to be involved to determine the penetration depth of CXL protocols.