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#### FECAL PROTEOMICS IN PARASITISED DOGS – A PILOT STUDY IN TRICHURIS VULPIS NATURALLY INFECTED ANIMALS

Silvia Vincenzetti (1), Giacomo Rossi (1), Fabrizia Veronesi (2), Manuela Diaferia (2), Alessandra Gavazza (1), Andrea Marchegiani (1), Beniamino Tesei (1), Gianni Sagratini (3), Massimo Ricciutelli (3), Luca Scortichini (4), Stefania Pucciarelli (1), Matteo Cerquetella (1)

(1) University of Camerino, School of Biosciences and Veterinary Medicine. (2) University of Perugia, Department of Veterinary Medicine. (3) University of Camerino, School of Pharmacy. (4) DVM, Maiolati Spontini

(AN), Italy.

Corresponding author: M. Cerquetella (matteo.cerquetella@unicam.it)

The present pilot study aimed at deepening the knowledge on fecal proteomics in dogs; it has been chosen to study the fecal proteome from dogs affected by helminthoses, using T. vulpis as a model of infection, since it is localized and harms even severely large bowel, thus expecting damages more evident in fecal samplings. The feces from 15 animals, tested positive only to *T. vulpis* by flotation in NaNO<sub>3</sub> solution (density 1.350) and having a Mc Master fecal egg count (FEC) higher than 100 eggs for gram (epg) [1], were stored at -20°C for two-dimensional electrophoresis (2DE) analysis. All fecal samples were from dogs submitted to routine coprological screening (i.e. floatation, Baermann, coproantigen) for endoparasites at the Parasitic Laboratory of the Veterinary Teaching Hospital of Perugia and did not receive any anthelmintic treatment in the last two months. Four out of the 15 parasitized dogs showed diarrhea, and only three hematochezia; any other clinical signs in addition to those due to the parasitosis were recorded. The experimental design of the present proteome analysis is based on the complete sample pooling strategy [2]. Preparation of fecal samples for 2DE, and liquid chromatography-tandem mass spectrometry (LC-MS/MS) analysis were performed as described previously [3-5]. Among the spots expressed in our samples, only those that showed a normalized quantity greater than  $20 \times 10^3$  were selected for subsequent analysis by LC-MS/MS. Seven proteins/enzymes (plus 3 isoforms for myosin), or their fragments, were identified in feces: Cu-Zn superoxide dismutase, titin isoform X1, immunoglobulin lambda-1 light chain isoform X8, alkaline phosphatase, myosin, enolase, and albumin. Of interest, considering the parasitosis, is the finding of albumin, myosin, and titin (involved in muscle development and contraction) which presence is possibly the direct expression of the damage caused by the sub-epithelial localization of the parasite as well as of the normal mucosal turnover (for albumin) [4,6-8]. Also, the enzyme Cu-Zn superoxide dismutase is noteworthy, as it acts as enzymatic antioxidants during oxidative stress [9], considering that an increased level of oxidative stress has been reported in human chronic intestinal helminthic parasitosis e.g., roundworms and whipworms infections [10-11]. The main limitation of the present pilot study is the absence of the comparison with a healthy control group, which was however out of our aim. Fecal proteomics is a technique that is showing interesting perspectives in the study of gastrointestinal diseases in the dog. The present study provides indications that this technique has the potential to identify the presence of a GI damage, suggesting its study also in other digestive disorders, for possible diagnostic/prognostic/monitoring markers discovery.

[1] MAFF. Manual of Veterinary Parasitological Laboratory Techniques. ADAS, HMSO, UK, 1986.

[6] Bowman DD. Georgi's Parasitology for Veterinarians. 7th ed. Saunders Company,

2002.

<sup>[2]</sup> Vincenzetti et al. Biomarkers mapping of neuropathic pain in a nerve chronic constriction injury mice model. Biochimie, 158:172-9, 2019.

<sup>[3]</sup> Cerquetella et al. Fecal proteomic analysis in healthy dogs and in dogs suffering from food responsive diarrhea. The Scientific World Journal, 2019:2742401, 2019.
[4] Cerquetella et al. Fecal proteome in clinically healthy dogs and cats: Findings in pooled faeces from 10 cats and 10 dogs. Vet Rec Open, 8:e9:1-7, 2021.

<sup>[5]</sup> Rossi et al. Clinicopathological and Fecal Proteome Evaluations in 16 Dogs Presenting Chronic Diarrhea Associated with Lymphangiectasia. Vet Sci, 19;8(10):242, 2021.

<sup>[7]</sup> Taylor et al. Veterinary Parasitology. 3th ed. Blackwell Publishing, 2007.

<sup>[8]</sup> Guevel et al. Quantitative proteomic analysis of dystrophic dog muscle. J Proteome Res, 10(5):2465-78, 2011.

<sup>[9]</sup> McMichael MA. Oxidative stress, antioxidants, and assessment of oxidative stress in dogs and cats. J Am Vet Med Assoc, 231(5):714-20, 2007.

<sup>[10]</sup> Saraymen et al. Serum malondialdehyde level in patients infected with Trichuris trichiura. Saudi Med J, 25(12):2046-7, 2004.

<sup>[11]</sup> Chandramathi et al. Elevated levels of urinary hydrogen peroxide, advanced oxidative protein product (AOPP) and malondialdehyde in humans infected with intestinal parasites. Parasitology, 136(3):359-63, 2009.