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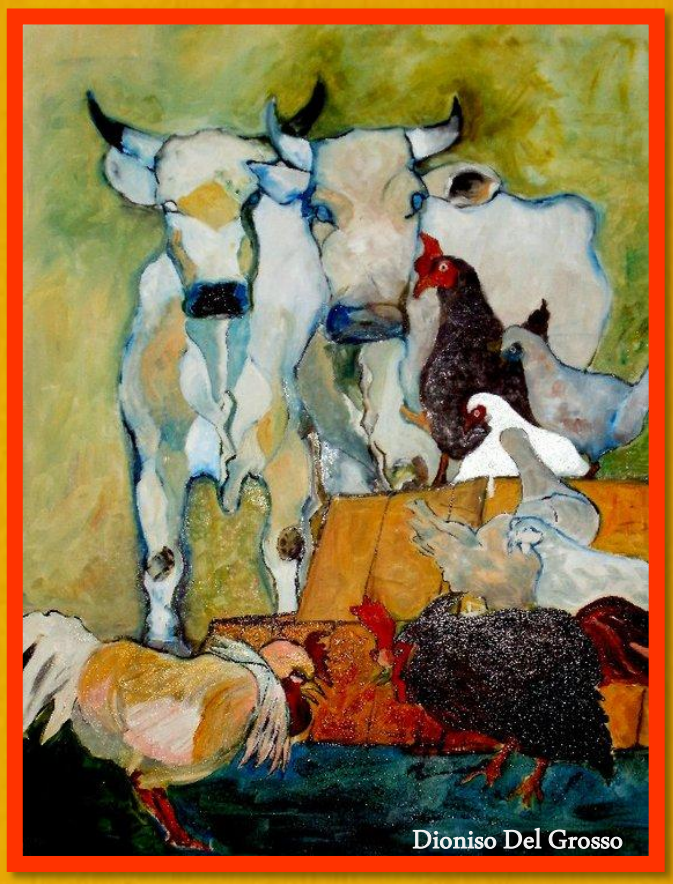

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XVI Convegno SIRA

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CHROMOGENIC IN SITU HYBRIDIZATION FOR THE DIAGNOSIS OF FELINE HERPESVIRUS-1 ASSOCIATED DERMATITIS

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Felid herpesvirus type 1 (FHV-1) is a worldwide pathogen mainly responsible of upper respiratory tract infection, ocular disease and dermatitis in felids [1]. The FHV-1-associated dermatitis is a facial and nasal dermatitis commonly seen on the dorsal and lateral muzzle, nasal planum and periorbital areas. These lesions overlaps with other feline dermatoses including hypersensitivity disorders, granuloma complex and cutaneous adverse food reaction [2]. Positive FHV-1 PCR results cannot guarantee an active role of FHV-1 in development of skin lesion because of latent infection, widely spread in cats and therefore conventional PCR possess limited clinical values [3]. The aim of this study was to correlate the presence and the amounts of FHV-1 viral genomes on feline tissues, assessed by conventional and qPCR assays, to the visualization of FHV specific nuclear signal of infected cells by chromogenic in situ hybridization (CISH).

Twenty-two formalin fixed, paraffin embedded skin samples from cats with facial dermatitis were retrieved, and divided in four groups: 1) samples with a diagnosis of herpesvirus dermatitis (n=5); 2) samples with non-herpetic facial dermatitis (n=6); 3) samples with facial dermatitis of ambiguous nature (n=7); 4) samples from healthy cats (n=4). Data on conventional PCR and qPCR by the ΔΔCq method were available for all the cases. DNA extraction was performed using DNeasy Blood and Tissue kit (Qiagen, Hilden, Germany) and the extracted DNAs were amplified using specific set of primers amplifying two viral gene targets: glycoprotein B (gB) and thymidine kinase (TK). The probe synthesis was performed amplifying an 80 bp fragment of gB gene using DIG DNA labelling mixture (Roche) HotStartTaq plus PCR kit (Qiagen). CISH was performed in automation on Ventana BenchMarck ULTRA (Roche, USA). All the cases of group 1 and 2/7 of group 3 were positive by both qPCR and CISH; all samples of group 2 and 4 were negative by both methods. Some of the cases that were negative by both qPCR and CISH, scored positive to conventional PCR (2/6 group 2; 6/7 group 3; and 1/4 group 4).

To the authors' knowledge this is the first time that conventional PCR, qPCR assay by the ΔΔCq method and CISH are simultaneously applied for the diagnosis of FHV-1 associated dermatitis in cats. Both qPCR and CISH methods, resulted to be more specific than conventional PCR, and sensitive to provide a correct diagnosis for FHV-1 associated dermatitis, particularly when histological features are not conclusive.

[1] Parzefall B, Schmahl W, Fischer A et al. Evidence of feline herpesvirus-1 DNA in the vestibular ganglion of domestic cats. Vet J 2010, 184:371-372. [2] Gaskell R, Dawson S, Radford A et al. Feline herpesvirus. Vet Res 2007, 38:337-354. [3] Persico P, Roccabianca P, Corona A, et al. Detection of feline herpesvirus 1 via polymerase chain reaction and immunohistochemistry in cats with ulcerative dermatitis, eosinophilic granuloma complex reaction patterns and mosquito byte hypersensitivity. Vet Dermatol 2011, 22:521-527.