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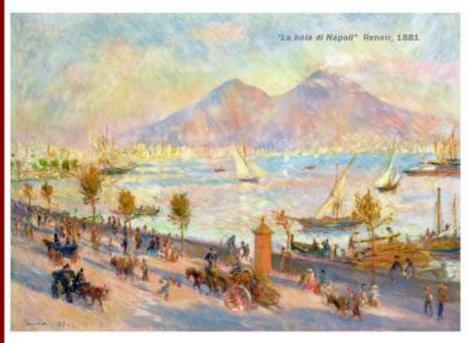
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## INVESTIGATION OF HUMAN TNF-α-308G/A SINGLE NUCLEOTIDE POLYMORPHISM IN DOGS PRESENTING IDIOPATHIC INFLAMMATORY BOWEL DISEASE OR FOOD RESPONSIVE DIARRHEA – PILOT STUDY

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Idiopathic inflammatory bowel disease (IBD) and food responsive diarrhoea (FRD) and are two common canine chronic enteropathies (CEs) [1-3]. Even if the pathogenesis of IBD is not yet clearly defined, it is probably due to environmental factors, microbiome, mucosal immune system dysfunctions, and to the genetic susceptibility of the host [1,2,4]. The authors hypothesize that the presence of the TNF-α-308G/A single nucleotide polymorphism (SNP) in dogs affected by CEs could be of aid in predicting the severity of such diseases, or even a possible predisposition, similarly to what previously reported in man for IBD [5]. In 2007 it was described that coding sequences for TNF-α in man (Homo sapiens) are similar for the 90.8% to those of Canis familiaris [6]; by using a human genetic test (GENOKIT®, registered trademark of BIOAESIS srl.), we collected buccal swabs in 13 dogs previously diagnosed with IBD (n=5) or FRD (n=8) [7]. Genomic DNA was extracted from the buccal swabs of the 13 samples. The TNF- $\alpha$ -308G/A SNP was genotyped using polymerase chain reaction-restriction fragment length polymorphism (PCRRFLP) assay. The enzyme digestion of the amplified product of sample 5 FRD gave the heterozygotes result: the genomic DNA of dog 5 FRD has both the TNF- $\alpha$ -308G allele and the TNF- $\alpha$ -308A. The subsequent sequence analysis confirmed the presence of both the guanine and the adenine in the corresponding -308 position. In canine medicine no studies correlating the TNF- $\alpha$  gene's polymorphisms and CEs were ever performed [8,9]. On the contrary, some studies investigated TNF expressions in dogs CEs, even if with discordant results [1,10-14]. The detection of the TNF- $\alpha$  (-308G/A) polymorphism only in one dog (and only in one of the two swabs) is probably due to the fact that only in this patient we obtained an adequate amount of DNA from the buccal swabs. Unfortunately, this single finding (gene and polymorphism) (however, the latter, for the first time in the dog) did not allow us to make any considerations about the correlation between its presence and the other variables considered (CIBDAI [15], albumin concentration, histopathology). Nevertheless, in our opinion this pilot study paves the way for future studies in this direction, possibly performed on a lager cohort of dogs, of selected breeds, by also including different substrates (e.g. blood), to confirm whether the test performed in the present study could be used successfully in dogs (for both gene and polymorphism), and to confirm the eventual presence of the polymorphism in diseased (or healthy) patients.

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