International Journal of Immunogenetics, 39, 314-320

The pathogenesis of Johne's disease (JD), caused by Mycobacterium avium subsp. paratuberculosis (MAP), is complex and has not been completely understood yet. In the present study, we analysed the polymorphism in the exon-2 of the caprine major histocompatibility complex (MHC) Class II DRB region and its association with resistance or susceptibility to JD. A total of 203 Jamunapari goats, which is an Indian endangered breed highly susceptible to JD, kept at a single farm were studied. On the basis of clinical signs, microscopic examination, faecal culture, ELISA and diagnostic PCR, 60 and 143 goats were classified as resistant and susceptible to JD, respectively. PCR-based restriction fragment length polymorphism (PCR-RFLP) with two enzymes, PstI and TaqI, was used to assess variation in the DRB gene(s) in all 203 goats studied. Two di-allelic single nucleotide polymorphisms (SNPs), here referred as P and T, were tested. In each of them, three genotypes were found in the group analysed. The minimum allele frequencies (MAFs) were 0.233 and 0.486 for the P and T SNPs, respectively. Statistically significant associations between alleles, individual genotypes and composed genotypes of both SNPs were found. The frequency of p and t alleles, of individual pp and tt and of composed pptt genotypes were significantly higher (Pcorr < 0.001) in the resistant group as compared to the susceptible group, while the P and T alleles were associated with susceptibility (Pcorr < 0.001). In heterozygous genotypes, susceptibility was dominant over resistance. The effects of both SNP on resistance and susceptibility were comparable and composed heterozygous genotypes showed intermediate levels of susceptibility in terms of the odds ratio and P-values calculated.

Comparative Immunology Microbiology and Infectious Diseases, 35, 303-307

Routine cultivation methods are able to distinguish between isolates of the Mycobacterium avium and the Mycobacterium tuberculosis complex. However, molecular tools are needed to further identify the several subspecies in the M. avium complex, especially for the subspecies avium and silvaticum. A rapid technique using Hhal restriction digestion of a 349 bp amplification product of the 85B antigen (alpha-antigen) gene was used for the identification of M. avium subsp. silvaticum in a three-year-old gelding presenting with caseose, necrotizing, granulomatous lesions. The result was confirmed by sequencing of the 85B antigen gene. (c) 2012 Elsevier Ltd. All rights reserved

1261 Rastislav, M., Mangesh, B. (2012) *BoLA-DRB3 exon 2 mutations associated with paratuberculosis in cattle* 
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A single nucleotide polymorphism at the antigen recognition site of the bovine leucocyte antigen (BoLA) DRB3 gene was assessed in healthy and Mycobacterium avium subsp.
paratuberculosis (MAP) - infected cattle, in order to determine if there was a correlation between mutations and altered susceptibility to infection. Of a sample of 200 animals, 19.6% were found to be infected with MAP. PCR - single strand conformational polymorphism analysis of the BoLA DRB3 gene found 19 genotypes (16 in the heterozygous and three in homozygous state, respectively). Four mutations, Val53Glu (OR 453.7), Val53Leu (OR 453.7), Asp57His (OR 1.944) and Arg84Gly (OR 1.458), were linked with increased susceptibility to infection, whereas, Asp57Asn (OR 0) and Phe60Tyr (OR 0.453) were associated with increased resistance. The findings indicate potentially important mutations in the protein-binding site of DRB3, which may be crucial to the activation of an appropriate immune response against MAP. (C) 2011 Elsevier Ltd. All rights reserved

No evidence that wild red deer (Cervus elaphus) on the Iberian Peninsula are a reservoir of Mycobacterium avium subspecies paratuberculosis infection
Veterinary Journal, 192, 544-546

The potential role of red deer (Cervus elaphus) as a reservoir of Mycobacterium avium subspecies paratuberculosis (MAP) infection is largely unknown. A total of 332 wild red deer were investigated using postmortem examination, bacteriology and serology. Only three animals (1.12%) were found to have lesions on histopathological examination and no MAP bacteria were recovered on culture. The results suggest it is unlikely that wild red deer make a significant contribution to the maintenance of MAP infection in the region. The cross-reactivity of the ELISAs used indicates this diagnostic modality is ineffective in the detection of MAP infection in this species. The implications of these results for the control of this important pathogen in both livestock and wildlife are discussed. (C) 2011 Elsevier Ltd. All rights reserved

Mycobacterium avium complex - the role of potable water in disease transmission
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Mycobacterium avium complex (MAC) is a group of opportunistic pathogens of major public health concern. It is responsible for a wide spectrum of disease dependent on subspecies, route of infection and patients pre-existing conditions. Presently, there is limited research on the incidence of MAC infection that considers both pulmonary and other clinical manifestations. MAC has been isolated from various terrestrial and aquatic environments including natural waters, engineered water systems and soils. Identifying the specific environmental sources responsible for human infection is essential in minimizing disease prevalence. This paper reviews current literature and case studies regarding the wide spectrum of disease caused by MAC and the role of potable water in disease transmission. Potable water was recognized as a putative pathway for MAC infection. Contaminated potable water sources associated with human infection included warm water distribution systems, showers, faucets, household drinking water, swimming pools and hot tub spas. MAC can maintain long-term contamination of potable water sources through its high resistance to disinfectants, association with biofilms and intracellular parasitism of free-living protozoa. Further research is required to investigate the efficiency of water treatment processes against MAC and into construction and maintenance of warm water distribution systems and the role they play in MAC proliferation

New publications in the CROHN’S DISEASE AND PARATUBERCULOSIS database (719-720)
Nucleotide binding and oligomerization domain-containing protein 2 (NOD2/Card15) is an intracellular protein that is involved in the recognition of bacterial cell wall-derived muramyl dipeptide. Mutations in the gene encoding NOD2 are associated with inherited inflammatory disorders, including Crohn disease and Blau syndrome. NOD2 is a member of the nucleotide-binding domain and leucine-rich repeat-containing protein gene (NLR) family. Nucleotide binding is thought to play a critical role in signaling by NLR family members. However, the molecular mechanisms underlying signal transduction by these proteins remain largely unknown. Mutations in the nucleotide-binding domain of NOD2 have been shown to alter its signal transduction properties in response to muramyl dipeptide in cellular assays. Using purified recombinant protein, we now demonstrate that NOD2 binds and hydrolyzes ATP. Additionally, we have found that the purified recombinant protein is able to bind directly to muramyl dipeptide and can associate with known NOD2-interacting proteins in vitro. Binding of NOD2 to muramyl dipeptide and homo-oligomerization of NOD2 are enhanced by ATP binding, suggesting a model of the molecular mechanism for signal transduction that involves binding of nucleotide followed by binding of muramyl dipeptide and oligomerization of NOD2 into a signaling complex. These findings set the stage for further studies into the molecular mechanisms that underlie detection of muramyl dipeptide and assembly of NOD2-containing signaling complexes.


Previous studies have established that Mycobacterium tuberculosis heat shock protein 65 (mHSP65) plays an important role in immune-associated diseases as an autoimmune factor. Some overlapping epitopes of mHSP65 may serve as initiators of both atherosclerosis and other autoimmune-associated diseases. In the present study, atherosclerosis was significantly enhanced in high-cholesterol diet (HCD)-fed New Zealand white rabbits immunized with mHSP65(91-105) compared with PBS-immunized or BSA-immunized rabbits. Immunizing wild-type C57BL/6J mice with mHSP65(91-105) induced the aortic endothelial injury. Although western blot demonstrated that specific antibodies against mHSP65(91-105) can cross-react with recombinant human heat shock protein 60, specific antibodies against mHSP65(91-105) had no direct effects on HUVECs in vitro. Laser scanning confocal microscopy showed that mHSP65(91-105) localized in the cytoplasm of HUVECs, even when HUVECs were heat shocked at 42A degrees C. mHSP65(91-105)-specific splenic cells secreted more IFN-gamma than controls. Also, adoptive transfer of mHSP65(91-105)-specific splenic cells can accelerate atherosclerosis in idlr (-/-) mice. We can conclude that the (auto)immune response to mHSP65(91-105) accelerates atherosclerosis in animal models, and that the response of Th1 plays an important role in this progress.