

SLC11A1 genetic variation between Holstein Friesian and Brown Swiss cattle, its influence on NRAMP1 expression and the potential association with bovine tuberculosis resistance.

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Background

- o Bovine Tuberculosis (bTB) is caused by Mycobacterium bovis.
- Cattle breeds show varying incidence of bTB susceptibility ^[1].
- o Brown Swiss (BS) cattle show more resistant responses to intracellular bacteria compared to Holstein Friesians (HF)^[2], and therefore maybe more resistant to bTB.
- o The solute carrier family 11 member 1 (SLC11A1) gene encodes for natural resistance-associated macrophage protein 1 (NRAMP1) which confers resistance against intracellular bacteria such as *M.bovis*^[4,5,6]. Study aims: to compare SLC11A1 polymorphisms between Ο HF and BS cattle and assess how these impact NRAMP1 protein expression.

Results

Genetic Analysis

Three non-synonymous SNPs were identified in the SLC11A1 gene.

• SNP1 c.650C>T





Methods

Study population: o SLC11A1 sequencing: 15 BS and 15 HF o NRAMP1 ELISA:

- SNP2 c.961G>A
- SNP3 c.1066C>G

SNP1 and SNP2 were present at very low frequencies (<10%) in both HF and BS cattle.

For SNP3 the alternative G allele was present at a higher frequency in BS (30%) compared to HF (13%), but this was not significant.







Macrophages from HF and BS do not produce significantly different amounts of NRAMP1 protein.

CG/GG SNP3 Genotypes Macrophages from cattle which are CG/GG at SNP3 produce significantly more NRAMP1 protein (p=0.0496) compared to those which are the CC genotype.

Conclusions

- o Breed did not appear to influence SLC11A1 genotype or macrophage NRAMP1 protein production.
- The presence of the alternative G allele at SNP3 (c.1066C>G) was significantly associated with higher amounts of NRAMP1 protein expression in macrophages.
- o NRAMP1 has previously been shown to play a vital role in innate immune responses against intracellular pathogens^[7].
- Therefore, the presence of the G allele at SNP3 (c.1066C>G) could influence resistance to bTB.
- o Further studies with larger animal populations are necessary to confirm our findings o If confirmed, these findings could be considered for incorporation into cattle breeding programmes seeking to breed cattle resistance to bTB

References

[1] Ameni, G., Aseffa, A., Engers, H., Young, D., Gordon, S., Hewinson, G., Vordermeier, M. (2007) 'High prevalence and increased severity of pathology of bovine tuberculosis in Holsteins compared to zebu breeds under field cattle husbandry in central Ethiopia', Clinical and Vaccine Immunology, 14(10), 1356-1361.

[2] Gibson, A. J., Woodman, S., Pennelegion, C., Patterson, R., Stuart, E., Hosker, N., Siviter, P., Douglas, C., Whitehouse, J., Wilkinson, W., Pegg, S. A., Villarreal-Ramos, B., Werling, D. (2016) 'Differential macrophage function in Brown Swiss and Holstein Friesian cattle', Veterinary Immunology and Immunopathology 181, 15-23.

[4] Ruiz-Larranaga, O., Garrido, J. M., Manzano, C., Iriondo, M., Molina, E., Gil, A., Koets, A. P., Rutten, V. P. M. G., Juste, R. A., Estonba, A. (2010) 'Identification of single nucleotide polymorphisms in the bovine solute carrier family 11 member 1 (SLC11A1) gene and their association with infection by Mycobacterium avium subspecies paratuberculosis', Journal of Dairy Science, 93(4), 1713-1721.

[5] Balasubramaniam, S., Kumar, S., Sharma, A., Mitra, A. (2013) 'Microsatellite (GT)n polymorphism at 3'UTR of SLC11A1 influences the expression of brucella LPS induced MCP1 mRNA in buffalo peripheral blood mononuclear cells', Veterinary Immunology and Immunopathology, 152(3-4), 295-302.

[6] Liu, K., Zhang, B., Teng, Z., Wang, Y., Dong, G., Xu, C., Qin, B., Song, C., Chai, J., Li, Y., Shi, X., Shu, X., Shang, Y. (2017) 'Association between SLC11A1 (NRAMP1) polymorphisms and susceptibility to tuberculosis in Chinese Holstein cattle', Tuberculosis, 103, 10-15.

7] Johnson, E., Wessling-Ressnick, M. (2012) 'Iron metabolism and the innate immune response to infection', Microbes and infection, 14(3), 207-216.