Rumen fluke, *Calicophoron daubneyi*, are gastro-intestinal parasites infecting grazing livestock worldwide. Heavy infections of immature flukes result in animal morbidity and mortality. Rumen fluke prevalence has rapidly increased across the UK and Ireland. However, our understanding of parasite-host interactions, particularly in terms of how this parasite modulates host immune responses to aid its survival, is limited. Related pathogenic helminths are shown to release extracellular vesicles (EVs), containing immune modulators (both predicted and validated), allowing them to manipulate the host immune responses to facilitate survival. Adult rumen flukes are demonstrated to produce and release EVs, yet their actions on the host immune system have not yet been characterised. Historically, the effect of infectious diseases on the host have been investigated through traditional *in vitro* models, such as cell culture models. However, such models cannot account for the multiple cell types present within tissues and therefore cannot fully represent natural infections. As a result, there is increasing interest in the development of tissue explant models, whereby animal tissues are maintained *in vitro*.

This research project has optimised an *in vitro* explant model of the bovine rumen for exploring host-parasite interactions. Stimulation of the model with *E. coli* derived liposaccharide (LPS), a common bacterial antigen known to stimulate immune responses in alternative tissue models and bovine epithelial cell cultures, was conducted to validate the ability of the model to mount an immune response. Rumen tissue explants (n=3) were cultured with LPS or without (Control) for 48 hours before harvesting and preserved for downstream proteomic analysis via GeLC proteomics. Principle component analysis (PCA) was used to assess the whole proteomic profiles from control and LPS treated explants. The majority of variance between samples was attributed to biological replication (as explained by PC1-2), determining a strong effect of individual animals on protein expression within the model. However, assessment of PC3 and 4 demonstrated differences in profiles between control and LPS treated explants. Differential expression analysis (via gProfiler) identified that several proteins with immune function were differentially expressed following LPS treatment, including complement factor B, galactin 3 and TNF alpha induced protein 8, and explaining the differences in overall protein profiles as determine by PCA analysis. As such, our rumen explant model was demonstrated to be capable of mounting an immune response *in vitro* following LPS stimulation and was therefore deemed suitable for investigating immune functions, presenting a novel mechanism for studying the immunomodulatory effects of rumen fluke EVs at the tissue level. Rumen explants have now been stimulated with a physiologically relevant dose of Rumen fluke EVs and samples will be analysed via our established proteomic pipeline.