

## **RNA interference: a functional tool for screening potential vaccine targets in the poultry red mite *Dermanyssus gallinae***

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The Avian haematophagous ectoparasite, the poultry red mite (PRM), *Dermanyssus gallinae*, affects the health and welfare of poultry, bringing substantial economic losses to the layer industry worldwide. Current acaricide-based controls are limited by ineffective application and emerging resistance. A sustainable control method or effective vaccine is therefore urgently needed by the egg laying industry. RNA interference (RNAi) as a gene knock-down tool has now been successfully established for the validation of gene function in *D. gallinae*. In this study, the aspartic protease, Cathepsin D (CatD) from *D. gallinae*, with a likely function in blood meal digestion, was selected for targeted gene knock-down by RNAi. Gene silencing was achieved through the oral delivery of target gene-specific double-stranded RNA (dsRNA) within the PRM blood meal (goose blood) via an *in vitro* feeding device. After 72hrs post-blood meal feeding using female mites, RNA was extracted for qPCR confirmation of CatD gene-knockdown. This confirmed that CatD expression was successfully knocked down by at least 65%. All PRMs were then re-fed with one more round of target gene-specific dsRNA in order to assess any phenotypic changes in terms of blood meal digestion. The experimental group of PRMs showed decreased blood digestion by 50% after 2 rounds of target gene-specific dsRNA delivery compared to the lacZ dsRNA treated controls. At the proteomic level, western blotting revealed increased haemoglobin residues and decreased levels of CatD protein in the CatD knock-down group. Previous vaccine trials have demonstrated the potential efficacy of using CatD as a target antigen and this study demonstrates the potential of using RNAi as a screening tool to identify novel vaccine targets, potentially reducing the need for large animal trials during the target selection phase.