

First observation of Parasitic viruses in *Trichomonas gallinae*

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Trichomonas gallinae is a single cell protozoan parasite that causes avian trichomonosis in a diverse array of birds especially pigeons and doves. Numerous studies show that viruses can reside within protozoan pathogens and contribute towards pathogen virulence and the closely related *Trichomonas vaginalis*, can be infected with a double-stranded RNA (dsRNA) virus which enhances its pathogenicity. However, the presence of *T. gallinae* has remained hitherto undiscovered. We screened a cryobank containing hundreds of UK passerine columbid and raptor isolates of *T. gallinae* with a wide range of genotypes for the presence or RNA virus. An initial Agarose gel-based screen of extracted RNA from different isolates of *T. gallinae* revealed an extra band of RNA in two isolates (C3 and C10). This band of RNA is consistent with the size of viral dsRNA and indicative of viral infection in *T. gallinae*. The presence of dsRNA was further verified using immune fluorescence monoclonal antibodies J2 specific to dsRNA viruses. Both these isolates were from infections which lacked demonstrable pathology and which were considered to be avirulent strains. To characterize the effect of the virus on *Trichomonas gallinae* we compared these strains with two virulent strains which lacked virus namely (A1 and C4). We observed that virus infected cells of *T. gallinae* were smaller and grew less well than non-infecting cells. Moreover, using (scanning and transmission) electron microscopic methods, we found evidence of plasma membrane disruption and granular structures which may be virus budding from the cell surface. Using negative staining of supernatants, we found icosahedral structures which may be virions. Using RNA transcriptomics, we were able to show expression of viral RNAs with 70% RNA identity to Trichomonas Virus 1. Overall our study offers new insight into parasitic pathogenesis of *T. gallinae* which in contrast to *Trichomonas vaginalis* correlates with low virulence of strains. It is to be hoped that knowledge of the virus may provide a route to novel intervention strategies for avian trichomonosis in birds.