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Analysis of long non coding transcripts in wild type and knock-out mutant Pseudorabies virus strains

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Introduction: Pseudorabies virus (PRV) belongs to the Alpha herpesvirinae subfamily. PRV is a commonly used transsynaptic neural tracer and it is also a used as a model for the study of transcriptional gene regulation. The viral DNA genome is double-stranded encodes 70 protein coding genes.Two latency associated long non-coding transcripts (LAT and AST) were previously described and characterized. In this project we have identified and characterized two formerly unknown long non coding RNAs of PRV.

Materials & Methods: Immortalized porcine kidney 15 (PK-15) cells were infected with Kaplan (Ka),ep0 knock-out (KO) mutant or VHS KO-mutant strain of PRV. Total RNA were purified and cDNAs were synthesized from the samples. For the analysis of the abundance of the novel transcripts, Illumina sequencing and Real-Time RT PCR techniques were used. The kinetics of these transcripts was characterized by PacBio sequencing and Real-Time RT PCR. The effects of the KO mutations on the expression of the novel transcripts were tested by Real-Time RT PCR.

Results: Two novel polyadenylated, 3'-coterminal non protein coding RNAs were identified using Illumina and PacBio sequencers as well as Northern-blot analysis. These transcripts (termed CTOs) located between the ul21 and ul22 genes close to the replication origin of the viral genome. The length of the short intergenic lncRNA (CTO-S) is 286 base pairs. The longer (CTO-L) transcript overlaps OriL and it is 2615 bp. CTO-L originates from the promoter of the ul21 gene. The expression levels of the two CTOs were higher than that of the wt PRV in the vhsKO virus. Our data show that the early protein 0 (encoded by the ep0 gene) exerts a down-regulatory effect on the transcription of CTOs throughout the whole life cycle of PRV.

Biography

Zsolt Csabai is a PhD student at the Department of Medical Biology on the University of Szeged, Hungary. He has earned his M.Sc. (Molecular Biology–Biotechnology–Microbiology MSc Program) in University of Szeged. His area of reserch is virus transcriptome analysis with qPCR technique.

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