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Immunofluorescent staining and mirna expression of HCT-8 and HT-29 cell lines upon cryptosporidium infection (Article)

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Abstract

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The protozoan *Cryptosporidium* mainly infects epithelial cells of the colorectal region. The infection of this species can be observed and confirmed by immunofluorescent staining method in vitro. In addition, the onco-microRNA (miRNA) and tumour suppressor miRNA expression pattern of the epithelial cells upon *Cryptosporidium* infection may reveal possible effect of *Cryptosporidium* in the onset and progression of cancer. In this study, two epithelial colorectal cancer cell lines HCT8 and HT29 were infected with *Cryptosporidium parvum* (*C. parvum*) and observed using direct immunofluorescent technique via fluorescein conjugated Vicia Villosa Lectin (VVL) staining. Real Time-quantitative Polymerase Chain Reaction (RT-qPCR) was performed to determine the expression of miR-21 and miR-145 in infected cell lines. Data normalisation of the miRNA expression was carried out using reference gene RNU44. The immunofluorescent micrographs exhibited the *Cryptosporidium* infection on both HCT-8 and HT-29 cell lines with green fluorescence. Upregulation of miR-21 and downregulation of normalised expression of miR-145 was observed in both of the cell lines upon *Cryptosporidium* infection. We found the expression of miR-21 to be increased and miR-145 greatly decreased in both cell lines upon infection. Our current observation suggests that, *C. parvum* can potentially infect the colorectal cancer cell lines and play critical role in regulating onco-miRNAs and tumor suppressor miRNAs. © 2018, Malaysian Society of Applied Biology. All rights reserved.

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