

TITLE: PHENOTYPIC CHARACTERIZATION OF A BRAZILIAN STRAIN OF *MEGALOCYTIVIRUS* IN BF-2 CELLS

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Megalocytivirus, member of family *Iridoviridae*, is a genus that consists of double-stranded DNA virus with an icosahedral capsid of 120-200nm. Some virions of this genus can present an external lipid envelope around the capsid. Megalocytiviruses have a worldwide distribution and are responsible for a high rate of morbidity and mortality in many wild and commercial fish around the world. Infected fish show unspecific clinical signs as anorexia, abnormal swimming and lethargy, thus impairing early diagnosis. Although the *Megalocytivirus* infection is well known, little is known about the characteristics of the viral particle. The main goal of this study was to characterize the phenotype of a Brazilian strain of *Megalocytivirus* isolated from an ornamental fish *Poecilia reticulata*. For this purpose, a pool of tissues (liver, kidney and spleen) of a *Poecilia reticulata* with positive molecular diagnosis for *Megalocytivirus* was submitted to isolation in 24-well culture plate containing BF-2 (Bluegill fry-2) cells. A cytopathic effect was observed in the second passage in BF-2 cells and the isolation was confirmed by polymerase chain reaction (*nested*-PCR) with sequential amplification of 1.362bp- and 369bp-fragments of the *Major Capsid Protein* (MCP) gene of *Megalocytivirus*. The *nested*-PCR product was sequenced and showed a high nucleotide similarity to corresponding sequences deposited in GenBank. Transmission electron microscopy of infected BF-2 cells revealed the presence of hexagonal viral particles with diameter of 150nm. Ether treatment showed that the majority of viral particles were enveloped. Three aliquots of virus-cell suspension were incubated at 25°C, 37°C and 56°C for 6h to determine the heat sensibility. There were no significant changes in the viral titers at 25°C and 37°C but the titer at 56°C was zero, which indicates that the virus is sensible to treatments at this temperature. A viral aliquot was submitted to freezing and thawing at -80°C for three times but the viral titer remained the same as control. All these characteristics are compatible with virions of *Megalocytivirus*. These results can contribute to better understanding of aetiology of *Megalocytivirus* infections in Brazil aiming also at future prevention and control actions for *Megalocytivirus* disease.

Keywords: ornamental fish, *Iridoviridae*, cell culture, viral diagnostics

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