Analyzing the Complex Carbohydrate Structures on the Plasma Membrane of the Cultured Mosquito Cells Using Fluorescently Labeled Lectins

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Every year, three million people across the world are killed due to mosquito-transmitted diseases. Carbohydrate structures found on plasma membranes of cells can act as receptors for viruses to initially infect the mosquito, and to replicate by budding throughout the mosquito. The purpose of this investigation is to characterize the repertoire of complex carbohydrate structures on the plasma membrane of Ag55 and C6/36 cultured mosquito cells, that may act as virus receptors. It was hypothesized that fluorescently labeled plant lectins could be used to label carbohydrate structures. The cells were first maintained, then labeled with lectins using two methods: fixation or live fixation. Using a confocal microscope, the cells were imaged, and analyzed by creating graphs and charts from the images. A high concentration of N-Acetylgalactosamine, Gluclose and Galactose was seen on the membranes of the cells by a high intensity from jacalin, LCA, and SBA lectins. The investigation was expanded through a biochemical analysis. The C6/36 cell plasma membrane was first isolated, then the protein was isolated. From the protein, a Western Blot was performed. The proteins were labeled with a biotinylated lectin and alkaline phosphatase avidin; the biotin-avidin reaction was colored. It was concluded that proteins with a mass of about 40 kDa or higher had a higher concentration of the carbohydrate. Particularly, proteins with a mass of 280 kDa had a high concentration of the carbohydrate with a low concentration of protein, exhibiting a high carbohydrate to protein ratio. Advancements in characterization of possible virus receptors contribute to the later development of a molecule to block the virus from bonding with the receptor, therefore preventing viral transmission.

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