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Author : Y Nining Sri Wuryaningsih

Detection of Dengue Virus Antigen in Monocytes to Support the Diagnosis of Dengue Hemorrhagic Fever

Abstrak :

Dengue virus infection is a major cause of morbidity and mortality in tropical and subtropical areas of the world. The immunopathological mechanism that results in severe complications of dengue virus infection, i.e. dengue hemorrhagic fever (DHF), is important to determine. Primary dengue infection induces serotype specific and serotype-crossreaction. In secondary infections with a virus a different serotype from that which causes primary infections, the presence of cross-reactive non-neutralizing antibodies, results in an increased number of infected monocytes by dengue virus antibody complexes. This, in turn, results in marked activation of serotype cross-reactive CD4 and CD8 memory CTL and result in overproduction of cytokines that affects monocytes, endothelial cells, and hepatocytes. Platelets are destroyed by cross-reactive anti-platelets autoantibodies. Dengue virus induced vasculopathy and coagulopathy must be involved in the pathogenesis of hemorrhage and the unbalance between coagulation and fibrinolysis activation increases the likelihood of severe hemorrhage in DHF/DSS. Definite diagnosis of dengue is provided by the detection of virus in acute-phase sera of patients. Virus isolation can be accomplished with mosquito cell lines or mosquito inoculation. However, these methods are time consuming and labor intensive. The reverse-transcriptase polymerase chain reaction (RT-PCR) provides a potential means of rapid diagnosis, but requires specialized facilities and equipment as well as expensive. Therefore, a rapid, simple, sensitive and economic sera is needed for clinical and epidemiological investigations. An amplified immunocytochemistry examination using streptavidinâ€“biotin is described for the detection the antigen of dengue virus on monocytes in patient's serum. Monocytes/macrphages are the major target cells for dengue virus and attachment of the dengue virus particle to the surface membrane receptor(s). This assay utilized biotinylated antibody directed against dengue antigens by anti-dengue complex monoclonal antibody and applied streptavidin peroxidase. After being incubated, 1-2 drops of DAB chromogen were added, counterstained and visualized by microscopy examination. The result was regarded as positive if the visualization was brown. The result of immunocytochemistry by streptavidin biotin was tested on 32 sera submitted routinely to our laboratory for confirmaton of dengue diagnosis. The sensitivity of the test streptavidin biotin was 88% and the specificity was 87.7%. In conclusion, the immunocytochemistry by streptavidin biotin can be used for early diagnosis of dengue infection.

Keyword :

dengue hemorrhagic fever, immunocytochemistry, streptavidin-biotin