Gene Polymorphisms and Serum Levels of Pro- and Anti-Inflammatory Markers in Dengue Viral Infections

Rosimar Neris Martins Feitosa,1 Antonio Carlos Rosário Vallinoto,1 Pedro Fernando da Costa Vasconcelos,2 Raimunda do Socorro da Silva Azevedo,2 Vânia Nakauth Azevedo,1 Luiz Fernando Almeida Machado,1 Sandra Souza Lima,1 Marluísa de Oliveira Guimarães Ishak,1 and Ricardo Ishak 1

Abstract

Pro- and anti-inflammatory markers (tumor necrosis factor [TNF]-α, TNF-β, interferon [IFN]-γ, interleukin [IL]-6, IL-8, IL-10, and C-reactive protein [CRP]) were investigated in 80 patients infected with dengue viruses, 100 patients presenting with febrile illness but negative for dengue, and 99 healthy subjects. Immunoenzyme methods were used for quantitative assays in the plasma. Polymorphisms of TNF-α, TNF-β, IFN-γ, IL-6, IL-8, and IL-10 genes were assessed by polymerase chain reaction (PCR)-restriction fragment length polymorphism and allele-specific oligonucleotide (ASO)-PCR for the IFN-γ. The highest mean serum levels of TNF-α, IFN-γ, IL-8, and CRP were observed in dengue-positive individuals. TNF-β, IL-6, and IL-10 levels were significantly higher in the dengue-negative individuals. No cytokine expression pattern was evidenced according to virus serotype. Genotypic frequency distributions were statistically significant for the polymorphisms of TNF-α and IFN-γ among positive, negative, and control dengue groups and IFN-γ among groups DENV-1, DENV-2, DENV-3, and controls. Modulation of cytokine expression and polymorphisms is a complex matter and needs further explanation considering the ethnic origins of the Brazilian population.

Introduction

Dengue virus is a member of the genus Flavivirus in the family Flaviviridae. Its genome is composed of a positive-sense, single-stranded RNA molecule, and there are four known serotypes as follows: DENV-1, DENV-2, DENV-3, and DENV-4 (15,76). An infection with any of the four agents results in a disease known as dengue, which is the most common arbovirus disease in the world (80). Specifically, dengue is endemic in all continents, except Europe, and causes approximately 100 million infections annually, including 500,000 cases of dengue hemorrhagic fever (DHF) (29).

Among the several mechanisms that have been described for DHF pathogenesis, the most studied corresponds to the shift of the cytokine-secreting profile of T helper (Th) lymphocytes (16,17). The pathogenesis of the disease caused by dengue virus is not yet fully elucidated, and some associations between increased cytokine levels and different clinical manifestations have been reported (12,16,32). Plasma levels of cytokines, such as tumor necrosis factor (TNF)-α, interferon (IFN)-γ, interleukin (IL)-2, IL-6, IL-8, and IL-10, and C-reactive protein (CRP), are also significantly increased in DHF patients compared to dengue fever (DF) patients (5,12,39,49,67).

Polymorphisms in the promoter regions of several cytokine genes can affect gene transcription and, consequently, cytokine production, which might be associated with disease outcome (23,68). Several studies have shown that cytokine gene polymorphisms can influence the evolution of several pathologies, such as autoimmune diseases, neoplasias, and post-transplant complications, and they are thus considered important markers of susceptibility to several diseases (10,20,23,35,70,74,75).

The present study investigated the frequency of polymorphisms and the expression of pro- and anti-inflammatory response markers (TNF-α, TNF-β, IFN-γ, IL-6, IL-8, IL-10, and CRP) and their possible association with dengue viral infections.

Materials and Methods

Characterization of the groups

During the dengue epidemic period, which ranged from October 2006 to April 2007, 180 persons presenting with a