

Characterization of monoclonal antibody B7 which neutralizes the cytotoxicity of pseudomonas exotoxin A. Clinical and Diagnostic Lab

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摘要

Abstract

A nontoxic *Pseudomonas aeruginosa* exotoxin A (PE), which has the carboxyl-terminal 38 amino acid residues of native PE deleted, was used as an antigen to immunize BALB/c mice, which were then challenged with native PE in order to raise monoclonal antibodies (MAbs) that can neutralize PE cytotoxicity. A murine MAb against PE, designated MAb B7, was established. MAb B7 was characterized in terms of its ability to neutralize PE cytotoxicity, epitope mapping, inhibition of PE receptor binding, and influence on cellular processing of PE and ADP-ribosylation activities. We found that MAb B7 could neutralize PE cytotoxicity in cell culture and in BALB/c mice. The epitope recognized by MAb B7 was mapped to the carboxyl-terminal amino acid residues 575 to 595 of PE. Consistent with the results of epitope mapping, MAb B7 did not block PE receptor-binding activity or the cellular processing of PE but strongly inhibited the ADP-ribosylating activity of PE. In addition, MAb B7 retained strong binding to PE even at pH 4.0, indicating that the complex of MAb B7 and PE is stable in the phagolysosome. On the basis of these observations, the neutralization of PE cytotoxicity by MAb B7 could be due to its binding to the carboxyl terminus of PE. As a result, MAb B7 may interfere with the interaction of the carboxyl-end amino acid residues REDLK of PE with cellular factors. However, we could not rule out the possibility that MAb B7 directly blocks the ADP-ribosylation activity of PE in the cytosol.