Analysis of structure-function of RNA polymerase of influenza A virus

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Influenza A virus has RNA-dependent RNA polymerase consisting of PB1, PB2, and PA. The RNA polymerase complex is integral to both transcription and replication of the vRNAs. In the present study, the relationship of structure-function of RNA dependent RNA polymerase of influenza A virus was analyzed using monoclonal antibodies.

To provide information on structure-function of RNA polymerase, monoclonal antibodies to polymerase proteins, PB2, PB1, and PA, were generated and these epitopes were mapped. Using the monoclonal antibodies, at least 4 different epitopes were defined on the PB 2 molecule. Antibodies recognizing the region of amino acids 206-259 and C-terminal proximal region inhibited the transcription of viral genome. Especially, monoclonal antibodies recognizing the region of amino acids 660-759 strongly inhibited transcription primed either by rabbit globin mRNA or ApG and endonuclease activity of PB2. Antigenic analysis revealed that the epitopes recognized by these monoclonal antibodies were highly conserved among human, equine, pig, and avian strains. It is possible that the region including these epitopes recognized by monoclonal antibodies inhibiting the transcription is responsible for the transcription of viral genome. Furthermore, it is revealed that the epitope recognized by 2 group IV antibodies are conserved among equine strains.

Using PB 1 monoclonal antibodies, 8 epitopes were defined on the molecule. According to the results in indirect immunofluoresence assay, it is indicated that the epitopes recognized by the monoclonal antibodies which reacted with PB 1 in immunoprecipitation assay but not in indirect immunofluoresence assay are masked by some host proteins in virus-infected cells. Two monoclonal antibodies bound to the N-terminal proximal region (amino acids 1-100) inhibited the transcription primed either by rabbit globin mRNA or ApG. It is indicated that these monoclonal antibodies affect the trancriptional activity of PB1, interfere with the binding of PB 1 to vRNA, and / or affect on the structure of polymerase complex.

Epitope mapping of PA using anti-PA monoclonal antibodies reveals that at least10 different epitopes are located on the PA molecule.

In the present study, the function-structure of polymerase proteins of influenza A virus was analyzed. The results provide important information for better understanding of the nature of influenza virus RNA-dependent RNA polymerase.

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