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Abstract

This invention relates to a method for producing virus RNA polymerases of RNA viruses, more specifically, virus RNA polymerases of RNA viruses free of virus genomic RNA.

The methods described in this invention includes the procedures for preparation of cDNAs for the genes for the component proteins of RNA polymerase of an RNA virus, incorporation of the cDNA into baculovirus genome to construct recombinant virus, and the infection of insect cells with the recombinant virus to express RNA polymerase. In this method, it is recommended that all species of the recombinant viruses, each of which is designed for expressing each of the above-mentioned component protein genes of RNA polymerase, are coinfecting into insect cells. Thus, cDNA is prepared for each of the component proteins of RNA polymerase and incorporated into baculovirus genome to construct recombinant virus for independently expressing the corresponding protein. In addition, the RNA viruses described above include influenza virus especially. In this method, RNA polymerase may be tagged to facilitate the subsequent purification. Furthermore, the tagged RNA polymerase described above may be purified using an adsorbent to trap the RNA polymerase at the tag. Another subject matter of this invention is to artificially prepare a complex of the component proteins of RNA polymerase of an RNA virus, or, in other words, to prepare RNA polymerase free from RNA virus genomic RNA. The RNA viruses described here include influenza virus, and the component proteins of RNA polymerase include PA, PB1 and PB2.