РАЗДЕЛ IV

ВИРУСОЛОГИЯ

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EFFECTS OF HUMAN SFPQ PROTEIN ON THE INITIAL STAGES OF HIV-1 LIFE CYCLE *

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Abstract

We have demonstrated that human SFPQ protein acts as a positive regulator of the integration and postintegration repair steps of HIV-1 replication. SFPQ binds to IN via two N-terminal RGG motifs. Substitution of Arg in these motifs disrupts this binding, and SFPQ with these substitutions does not affect integration efficiency but enhances postintegration repair as well as the wild-type protein.

Human immunodeficiency virus type 1 (HIV-1) causes one of the most dangerous incurable infectious pathologies. Highly active antiretroviral therapy (HAART), aimed at suppressing the functions of viral proteins, significantly improves the quality and life expectancy of infected people, but HIV-1 can develop resistance to it. The virus uses numerous cellular proteins to ensure its replication, which opens up new directions in the development of drugs that block interactions between viral and cellular proteins. In this regard, studying the molecular mechanisms of the cellular proteins' involvement in the processes of HIV-1 replication is an important task for the creation of innovative therapeutic agents.

The SFPQ protein (Splicing Factor, Proline- and Glutamine-rich) is involved in alternative splicing, transcription and transport of mRNA in the cell [1]. SFPQ is also known to participate in the HIV-1 life cycle. It is a part of the preintegration complex (PIC), which includes viral reverse transcriptase (RT) and integrase (IN). Direct interaction of SFPQ with HIV-1 IN has been shown *in vitro* [2]. However, the molecular mechanisms of SFPQ influence on HIV-1 replication remain poorly understood. The aim of this study was to identify the role of SFPQ in the early stages of the HIV life cycle.

To determine the specific stage of the HIV life cycle affected by SFPQ, a VSV-G pseudotyped, replication-incompetent lentiviral vector encoding firefly luciferase instead of HIV-1 enzymes was used. This vector adequately mimics early stages of the viral life cycle, including reverse transcription, integration, and post-integration repair. The levels of HIV-1 single cycle infection were quantified by measuring luciferase activity. The results showed that SFPQ acts as a positive factor for integration and post-integration repair.

In order to clarify the mechanism of the positive effect of SFPQ, the interaction of recombinant SFPQ with viral enzymes (IN and RT) was analyzed using the pull-down assay, and it was found that SFPQ interacts with IN, but not with RT. Using site-directed mutagenesis and pull-down analysis, two RGG motifs in the N-terminal domain of SFPQ were identified that are responsible for the interaction with IN. The protein with the substitution of Arg residues in these motifs did not bind to IN, and its superexpression in HEK 293T cells transduced with the pseudoviral vector did not affect the efficiency of integration, but increased the level of post-integration repair almost as well as the superexpression of the wild-type SFPQ protein. Thus, it was established for the first time that the cellular SFPQ protein positively affects two stages of the HIV-1 life cycle, and its effect on integration is mediated by its interaction with IN, but not on post-integration repair.

References

1. Knott G. J., Bond C. S., Fox A. H. The DBHS proteins SFPQ, NONO and PSPC1:a multipurpose molecular scaffold // Nucleic Acids Res. 2016. Vol. 44 (9). P. 3989–4004.

2. Yadav P., Sur S., Desai D. et al. Interaction of HIV-1 integrase with polypyrimidine tract binding protein and associated splicing factor (PSF) and its impact on HIV-1 replication // Retrovirology. 2019. Vol. 16. P. 12.

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