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Evaluation of siRNAs as antivirals against MERS-CoV in cell culture.

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Abstract

The respiratory syndrome caused by Middle East respiratory syndrome Coronavirus (MERS-CoV) was identified in April 2012 from Saudi Arabia. Since then, this has spread to 27 countries with 858 deaths and 2494 laboratory confirmed cases. Currently, there is no vaccine and antiviral therapy available. The Orf 1ab gene mediates the virus replication and the expression of a particular gene can be modulated by RNAi approach. In this work, we have Insilico designed and evaluated the chemically synthesized potential siRNAs in HEK-293-T cell lines. The selected siRNAs were evaluated for cytotoxicity and antiviral activity at various concentrations (0.1-50nm) using MTT assay through Lipofectamine mediated delivery in cell culture. The antiviral activity of siRNAs was analyzed by Real-time PCR. The average CC50 were observed as 272.7 for siRNA-1, 190.1 for siRNA-2, 389.3 for siRNA-3, 261.6 for siRNA-4, 134.5 for siRNA-5, 399.4 for siRNA-6, in HEK 293 cells. The CT value of siRNAs were variable for both cell supernatant and lysate as presented in the table. The siRNA-2 and siRNA-4 were found to show better antiviral activity as compared to others. Based on results obtained in this study, it is concluded that the designing and selection protocol of designed siRNA are very helpful to minimize the possible cytotoxic effects and showed potential antiviral activity of siRNA in Vero cells. The results indicated that the potential siRNAs can be evaluated further in other cell cultures and used as antivirals against MERS-CoV.

Biography

Dr. SAYED S. SOHRAB (Professor), Joined King Fahd Medical Research Center, King Abdulaziz University, Jeddah in Feb 2012. He has 16-year experience on Molecular Virology/ biology. Currently, he has 16 accepted proposals as Principal Investigator/Co-Pi from, Ministry of Health, KACST, RDO, Deanship of Scientific Research (DSR), King Abdulaziz University. Currently, he is working on molecular diagnosis of MERS-CoV, SARS-CoV-2, Dengue Virus, Influenza, HPV & Alkhumra virus and vaccine development against MERS and Dengue virus. He has published more than 89 research papers in internationally reputed scientific journals and six book chapters edited by different editors and submitted more than 350 genome sequences in GenBank. His research interest is molecular diagnosis, gene cloning and sequencing, gene construct development for vaccines, recombinant DNA technology, and genetic diversity analysis.