

production of both the gRNA and Cas9-encoding mRNA and b) incorporating a smaller saCas9 or cjCas9 nuclease. These measures will reduce the vector size and likely increase the vector titer.

We have compared different CRISPR-Cas systems for their efficiency in terms of antiviral activity and viral titer. Superior antiviral activity is reported for saCas9 compared to cjCas9, which can achieve full HIV inactivation in cell culture with only a single gRNA. We demonstrated that reduction of the vector size (smaller Cas9 nuclease and dual-polymerase active H1-promoter) increases the vector titer. This greatly facilitates the use of viral vectors with a limited packaging capacity. These results are important in the path towards the formulation of a cure strategy.

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## Construction of a lentiviral vector-carrying SARS-CoV-2 spike gene for COVID-19

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A new pandemic disease named COVID-19 has emerged in December 2019 in China spreading rapidly to other countries. This highly contagious disease is transmitted through respiratory droplets mostly affecting the lungs of the infected individuals. Consequently, the development of an effective vaccine is urgently needed to prevent the spread of SARS-CoV-2 virus. As suggested previously, genetic immunization is one of the most effective tools for vaccination. The *ex vivo* transfer of viral antigens to dendritic cells was the first method used in dendritic cell-based vaccines against infectious diseases. With the discovery of viral vectors, antigen encoding genes are delivered to dendritic cells by recombinant viruses. Among the viral vectors tested lentiviral vectors with improved biosafety profile appeared superior in the transduction of dendritic cells. We chose spike protein of SARS-CoV-2 as the antigen due to its role in infectivity and its potential to induce an effective immune response as shown in previously studies concerning SARS-CoV and MERS-CoV. A multisite gateway reaction was set up between entry and destination vectors to generate an expression clone carrying Spike gene (pLentiSpike). DNA sequencing and restriction enzyme analysis were utilized to confirm the orientation and the sequence of the transgene. Then, immunocytochemical staining of transfected cells with an Anti-SARS-CoV-2 spike glycoprotein antibody demonstrated Spike protein expression from the expression plasmid. Transient transfection of the transfer and packaging plasmids resulted in the generation of a lentivirus carrying spike protein encoding gene.

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## Combined antibacterial effect of polyethyleneimine and halloysite nanotubes

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The search for agents inhibiting the growth and development of pathogenic microorganisms is promising. It is known that polycation polyethyleneimine and its derivatives inhibit the growth of biofilms, in particular, the growth of *Candida albicans*. In addition, it has been shown to inhibit the growth of *Bacillus subtilis* biofilms on a substrate made of halloysite mineral and surface-active compounds. However, there are no studies evaluating the combined effect of polyethyleneimine and halloysite nanotubes on the growth and development of *E. coli*. The aim of this work was to study the effect of the combined effect of halloysite nanotubes at a concentration of 600 µg / ml, as well as polyethyleneimine (1%) on the growth of the planktonic form of *E. coli* for 96 hours. The object of the study was the gram-negative bacterium *Escherichia coli*. There are certain strains of *E. coli* that can cause infections by colonizing medical devices such as urethral and intravascular catheters, joint prostheses and shunts, and prostheses. Therefore, it is important to study the effect of various therapeutic agents to inhibit the growth of *E. coli*. Initially, using laser Doppler velocimetry, data on the zeta potential and hydrodynamic parameters of the studied compounds were obtained, which amounted to  $423.9 \pm 8.351$  nm and  $-36 \pm 0.819$  mV for halloysite nanotubes and  $55.63 \pm 1.241$  nm and  $+11.7 \pm 0.289$  mV for polyethyleneimine. The reported study was funded by Russian Foundation of Basic Research (grant 18-29-25057).

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## Adenoviral vector design and construction for SARS-CoV-2 vaccine development

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COVID-19 is a contagious and a fatal respiratory illness caused by SARS-CoV-2. The outbreak was declared a public health emergency of international concern on January 30 2020, and a pandemic on March 11 2020 by The World Health Organization. Since, vaccination is the most effective way for protection against such infectious diseases, outstanding efforts from various laboratories have focused on the development of effective and safe vaccines. The production of a vaccine requires the understanding of the molecular structure of the virus, as the selection of a proper antigen and the vector type is essential to induce an optimum immune response following vaccination. Compared to other structural proteins, the spike protein of SARS-CoV-2 appeared to be the most promising antigen candidate in vaccine development due to its essential role in viral transduction through interaction with the Angiotensin-Converting Enzyme 2 (ACE2) receptors. Viral vector-based vaccines induce both an effective cellular immune response and a humoral immune response owing to their natural adjuvant properties via transduction of immune cells. Accordingly, adenoviral vectors with well-known structure, serve as effective gene carriers to induce immune response via gene transfer. To generate a viral vector based vaccine candidate, we first constructed an adenoviral expression vector (pAd5Spike) encoding spike protein via Gateway cloning technology. 293T cells were transfected with pAd5Spike to confirm spike protein expression by immunostaining. The pAd5Spike plasmid was then used to produce the first generation adenovirus vector encoding spike protein (Ad5Spike).

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