RECOMBINANT LUMPY SKIN DISEASE VIRUS AS A VACCINE VECTOR

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Lumpy skin disease (LSD) is a viral highly contagious transboundary disease of cattle, less often - sheep, goats and buffaloes accompanied by fever, swelling of the subcutaneous connective tissue and organs, the formation of skin nodes, damage to the eyes, mucous membrane of the respiratory and digestive tracts. Mortality in lumpy skin disease does not exceed 10%, however, the financial damage is great, it is manifested by a decrease in gains and milk yields, as well as the inability to use cowhides. Since there are no specific treatments for LSD, vaccination is the most effective way to control and eradicate LSD. Vaccination with live attenuated vaccines is a key element in the prevention of LSD. The mechanism of virus attenuation by long-term passaging in sensitive systems remains unclear. Targeted inactivation of virulence genes is the most promising way to obtain attenuated viruses.

In this study, we sequentially knocked out four virulence genes in the lumpy skin disease virus (LSDV) genome using homologous recombination under transient dominant selection conditions. The knockout of the LSDV005, LSDV008, LSDV142 genes was carried out by completely deleting the coding sequence; the reading frame was broken in the LSDV066 gene due to the insertion of a foreign sequence. The presence of deletions and insertions was confirmed by both PCR and viral genome sequencing (GenBank ID: ON005067). Recombinant LSDV Atyrau-5BJN(IL18) stably expressed interleukin 18 in vitro in lamb testicles cells. It was found that the recombinant LSDV Atyrau-5BJN(IL18) retained its genetic stability for ten passages and replicated efficiently in lamb testicles cells as well as bovine and saiga kidney cells. Knockout of four genes did not affect virus replication in vitro. In vivo experiments with cattle have shown that injection of the LSDV Atyrau-5BJN(IL18) at a high dose does not cause disease in animals or other deviations from the physiological norm. Immunization of cattle with the LSDV Atyrau-5BJN(IL18) induced the production of virus-neutralizing antibodies in titers of 4–5 log2. The challenge did not cause disease in immunized animals. The knockout of four virulence genes resulted in attenuation of the virulent LSDV without loss of immunogenicity. LSDV Atyrau-5BJN(IL18) demonstrated high safety and immunogenicity. The ability to stably express foreign genes demonstrates the potential of using this virus also as a vaccine vector.

This research was funded by the Science Committee of the Ministry of Science and Higher Education of the Republic of Kazakhstan, grants number AP08856376